Effects of essential oils and aromatherapy on cognitive function in dementia: laboratory studies and application in aged care facility residents

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Southern Cross University

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Effects of essential oils and aromatherapy on cognitive function in dementia: laboratory studies and application in aged care facility residents.

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Submitted for the Degree of Doctor of Philosophy
Australian Centre for Complementary Medicine, Education and Research
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April, 2006
I hereby certify that the work embodied in this thesis is the result of original research and has not been submitted for a higher degree to any other University or Institution

Signed:
Acknowledgements

This thesis would not have continued if I had not been offered a University scholarship to allow me to dedicate full-time attention to my studies. Thank you to everyone at the University who believed in me and my project, in particular my supervisors, Stephen Myers, John Stevens, Phil Cheras, Lesley Stevenson, David de Vries and David Lin. Special thanks go to Sister Margaret Bray, my research assistant, and Henry Sheerwater and Delilah Williams who assisted with proof-reading.

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I would not have believed at the outset had someone warned me that the last three years would have been so complex a test of character. I have emerged humbler, and a little wiser about how to undertake a three year multiple task research project. I look forward to future opportunities to further examine the efficacy of aromatherapy in dementia.
List of publications

Journal articles

Abstracts

Oral presentations


Poster presentation
“Is the MMSE a useful tool in dementia trials?” Innovations in Management of Cognitive Impairment in Older Australians, Change Champions and Australian Resource Centre for Healthcare Innovations (ARCHI), Sydney, NSW, 6-7 April 2006.
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<thead>
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<th>Abbreviation</th>
<th>Full text</th>
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<tbody>
<tr>
<td>95% CI</td>
<td>95% confidence interval</td>
</tr>
<tr>
<td>ACCMER</td>
<td>Australian Centre for Complementary Medicine, Education and Research</td>
</tr>
<tr>
<td>AChE</td>
<td>Acetylcholinesterase</td>
</tr>
<tr>
<td>ADAS-cog</td>
<td>Alzheimer's Disease Assessment Scale - cognitive subscale</td>
</tr>
<tr>
<td>AIN</td>
<td>Assistant In Nursing</td>
</tr>
<tr>
<td>ANOVA</td>
<td>Analysis of Variance</td>
</tr>
<tr>
<td>ARCHI</td>
<td>Australian Resource Centre for Healthcare Innovations</td>
</tr>
<tr>
<td>ATCI</td>
<td>Acetylthiocholine iodide</td>
</tr>
<tr>
<td>BPSD</td>
<td>Behavioural and Psychological Symptoms of Dementia</td>
</tr>
<tr>
<td>CDR</td>
<td>Clinical Dementia Rating</td>
</tr>
<tr>
<td>ChAT</td>
<td>Cholineacetyltransferase</td>
</tr>
<tr>
<td>CINAHL</td>
<td>Cumulative Index to Nursing &amp; Allied Health (database)</td>
</tr>
<tr>
<td>CMAI</td>
<td>Cohen-Mansfield Agitation Index</td>
</tr>
<tr>
<td>DALY</td>
<td>Disability-adjusted life year</td>
</tr>
<tr>
<td>DSM-IV</td>
<td>Diagnostic and Statistical Manual of Mental Disorders - Fourth Edition</td>
</tr>
<tr>
<td>DTNB</td>
<td>5,5'-dithiobis-(2-nitrobenzoic acid)</td>
</tr>
<tr>
<td>ECG</td>
<td>Electrocardiogram</td>
</tr>
<tr>
<td>GABA&lt;sub&gt;A&lt;/sub&gt;</td>
<td>Gamma-aminobutyric acid, receptor subtype A</td>
</tr>
<tr>
<td>GC-MS</td>
<td>Gas chromatography and mass spectrometry</td>
</tr>
<tr>
<td>IC&lt;sub&gt;50&lt;/sub&gt;</td>
<td>Inhibitory concentration that inhibits to 50% of the control</td>
</tr>
<tr>
<td>ICD-10</td>
<td>International Classification of Diseases, tenth edition</td>
</tr>
<tr>
<td>IGLS</td>
<td>Iterative Generative Least Squares</td>
</tr>
<tr>
<td>LDL-In</td>
<td>Immunoregulatory low-density lipoprotein</td>
</tr>
<tr>
<td>ML</td>
<td>Maximum Likelihood</td>
</tr>
<tr>
<td>mM</td>
<td>millimolar</td>
</tr>
<tr>
<td>MMSE</td>
<td>Mini-Mental State Examination</td>
</tr>
<tr>
<td>MPRC</td>
<td>Medical Plant Research Centre</td>
</tr>
<tr>
<td>MTD</td>
<td>Maximum Tolerated Dose</td>
</tr>
<tr>
<td>NINCDS-ADRDA</td>
<td>National Institute of Neurological Conditions, Disorders and Stroke – Alzheimer’s Disease and Related Disorders Association</td>
</tr>
<tr>
<td>NMDA</td>
<td>N-methyl-d-aspartate</td>
</tr>
<tr>
<td>NOAEL</td>
<td>No Observed Adverse Effect Level</td>
</tr>
<tr>
<td>NOSGER</td>
<td>Nurses’ Observation Scale of Geriatric Residents</td>
</tr>
<tr>
<td>NPI</td>
<td>Neuropsychiatric Inventory</td>
</tr>
<tr>
<td>NRACAN</td>
<td>Northern Rivers Aged Care Aromatherapy Network</td>
</tr>
<tr>
<td>NSW</td>
<td>New South Wales</td>
</tr>
<tr>
<td>PAS</td>
<td>Pittsburgh Agitation Scale</td>
</tr>
<tr>
<td>RN</td>
<td>Registered Nurse</td>
</tr>
<tr>
<td>rpm</td>
<td>revolutions per minute</td>
</tr>
<tr>
<td>SD</td>
<td>Standard deviation</td>
</tr>
<tr>
<td>Tx</td>
<td>Treatment</td>
</tr>
</tbody>
</table>
Abstract

This thesis investigates the use of aromatherapy for the improvement of cognitive function in aged care facility residents with dementia. A four-phase cascading research framework was used, comprising four phases: investigation; hypothesis formation; laboratory testing of the hypothesis; use of laboratory results in a randomised controlled trial of aromatherapy for cognitive function in aged care facility residents with dementia.

The investigation phase comprised a background literature review and an exploratory survey of existing aromatherapy use in aged care facilities in northern NSW. The hypothesis was formed that certain essential oils already being used in aged care would have potential activity against acetylcholinesterase (one of the major drug targets in dementia). The laboratory phase screened the twenty-five essential oils identified in the survey using a modified Ellman acetylcholinesterase technique. Five of the oils showed >50% acetylcholinesterase inhibition at 0.454 mg/mL (Cypress (Cupressus sempervirens), Lime (Citrus latifolia), Rosemary (Rosmarinus officinalis), Eucalyptus (Eucalyptus globulus) and Sage (Salvia officinalis)).

The final clinical trial phase compared two essential oils lotions (up to 2.7% essential oil w/w) and a placebo, with repeated cognitive testing using the well-accepted Mini-Mental State Examination (MMSE) over a three month period. One essential oil lotion contained three of the most active essential oils; the other contained three of the least active oils from the laboratory tests. Blends were used so that participants would not be distracted by being able to identify individual aromas. Lotions were applied once a day in the morning to the shoulders and neck of the 98 participants (selected from 10 facilities). Neither essential oil blend nor placebo were statistically different, and the overall decline over the treatment period was 0.9 (+/- 1.07) MMSE points.

The conclusion was that aromatherapy is not effective for short-term improvement of cognitive function in people with mild to moderate dementia (baseline scores 10-26 on MMSE). Use of stronger doses of essential oils over a six or twelve month period, and inclusion of a no-treatment arm could further test the hypothesis that some essential oils can affect cognitive function in dementia.
1 Introduction

This thesis investigates the research question: ‘Does aromatherapy affect cognitive function in aged care facility residents with dementia?’ Aromatherapy is the use of aromatic plant essential oils for the improvement of well-being, and is a popular complementary therapy in use in northern NSW aged care facilities. The choice to study dementia was influenced by the author’s growing awareness of the potential dementia epidemic facing Australia and other developed nations in the coming decades.

The decision to focus the research on the possible cognitive effects of aromatherapy on people with dementia arose out of the author’s previous experience of the use of aromatherapy in aged care, and a meeting with Dr E.K. Perry and her colleagues at the Medicinal Plant Research Centre (MPRC), Newcastle-on-Tyne, UK. The MPRC is investigating the effects of essential oils on cognitive function in dementia.

To ensure that the investigation of the topic was as comprehensive as possible, and yet targeted to answer the research question within the allowed time, a research framework was established.

The framework was divided into four phases each generated by results from the previous phase. The four phases were as follows:

1. Investigation of the research area by survey and background literature review.
2. Formation of research hypothesis and research plan.
3. Quantitative assessment of findings from the investigative phase to test hypothesis formed in phase two.
4. Testing of phase three results in a pilot trial in the research population.

The remainder of this thesis reports on the application of this framework to the research question mentioned above. The research population chosen for investigation was limited to residents of aged care facilities, mainly due to the relative ease of participant recruitment and the expectation that aged care facility environments would present less variability compared to environments of participants living in the community.
1.1 Phase One: Investigative phase

The investigative phase was designed to describe and summarise dementia and existing aged care aromatherapy practices, using a literature review and a survey of aromatherapy practices in aged care facilities in northern NSW.

1.2 Phase Two: Research hypothesis and plan

The hypothesis and research plan were generated after reflection on the background literature review and survey outcomes.

1.3 Phase Three: Quantitative assessment

The quantitative assessment involved laboratory assessment of the acetylcholinesterase inhibiting properties of essential oils identified in the survey.

1.4 Phase Four: Clinical testing of Phase Three results

The laboratory results were then used to develop appropriate essential oil blends to test the research question in a population of aged care facility residents with mild to moderate dementia.

1.5 Thesis outline

Chapters Two, Three and Four report on the first phase of the framework. Chapter Two provides an overview of dementia, its effects, social costs and management strategies used in Australia. Chapter Three follows with a definition of aromatherapy and an overview of the literature regarding the potential benefits of essential oils as agents in a dementia management strategy. A more intensive review of the possible acetylcholinesterase (AChE) inhibiting effects of essential oils and the use of aromatherapy in dementia management concludes the background to the research.

Chapter Four reports a survey of aromatherapy practices in twenty-eight aged care facilities in northern NSW. The survey had several aims, chief among those to identify the extent to which aromatherapy is thought (by nursing staff) to be useful for dementia, and to identify practices with possible application in a clinical trial. Results of the survey showed that aromatherapy is perceived as helpful for: pain management in arthritis; sleep improvement;
and improvement of behavioural and psychological symptoms of dementia. Aromatherapy was not perceived as useful for the improvement of cognitive function. Twenty-five essential oils were identified as being used regularly by aged care facilities, and limb massage and foot-baths were the most common application methods. Fifty-nine percent of all residents in the facilities surveyed received aromatherapy treatments, with 47% receiving daily treatments.

Chapter Five outlines the second phase of the framework, i.e. the research hypothesis generated from the review of aromatherapy use in dementia and the survey of aged care aromatherapy practices in northern NSW. This hypothesis was designed to enquire whether essential oils already being used in dementia care for behaviour management could also have an effect on cognitive function. The research plan emerging from the hypothesis proposed the screening of essential oils identified in the survey for AChE inhibiting properties, followed by a randomised controlled trial of aromatherapy for improvement of cognitive function in aged care facility residents with dementia. The aromatherapy blends to be used in the trial would be comprised of essential oils found to have AChE inhibiting properties in the laboratory phase.

The third phase of the framework is reported in Chapter Six. This contains reports of the laboratory assays of the essential oils identified in the survey. Appropriate essential oils were selected for a randomised controlled trial of aromatherapy for cognitive improvement in mild to moderate dementia. The chapter describes a modified Ellman cholinesterase method with preliminary methodological assessments. Results of assays on 25 essential oils used in aged care facilities are reported. Lime (Citrus latifolia), Cypress (Cupressus sempervirens) and Eucalyptus (Eucalyptus globulus) essential oils had the highest dose-dependent inhibitory activities (concentrations providing 50% inhibition (IC₅₀) in the range of 0.1mM) and were combined into an ‘active’ blend for the subsequent clinical trial. Mandarin (Citrus recutita), Ginger (Zingiber officinalis) and Lemongrass (Cymbopogon citratus) essential oils had the lowest inhibitory activity and were combined into an ‘inactive’ blend for the trial.

Chapter Seven covers the fourth phase of the framework, and describes and reports on a clinical trial testing the hypothesis that an ‘active’ aromatherapy lotion containing Lime, Cypress and Eucalyptus oil would have a beneficial effect on the cognitive function of people with dementia when massaged daily into neck and shoulders. The trial was an 18-week randomised, double-blind controlled trial with a three arm parallel group design, conducted in 10 aged care facilities over a 150 km radius in the Northern Rivers area of NSW. Results of the trial showed that neither the hypothesised ‘active’ lotion nor the control lotions had a
statistically significant effect on cognitive function as assessed by the Mini-Mental State Examination. Improvement in mean scores between the tests was noted, but was not attributable to the treatment. This suggests the need for further exploration of the frequency of testing for cognitive function tests in dementia trials to control for possible ‘learning’ effects.

Chapter Eight provides a general discussion of the results and critiques the methodologies used in the research projects, and concludes by providing suggestions for future studies on the use of aromatherapy in aged care.
2 Background: Dementia - impacts and management

Dementia is the ‘catch-all’ term for syndromes characterised by a gradual decline of higher cortical function, occurring most frequently in people over 65 years of age. Dementia is not specific to gender, ethnicity or education, (ADI, 1999), although a longer education period appears to delay cognitive decline (Chan et al., 2001). About 1% of the Australian population was estimated to have dementia in 2005 (Access-Economics, 2005).

At present no single cause of dementia has been identified, and there are several different types of dementia. There is no known prophylaxis or cure although several drugs appear to delay cognitive decline, and life-style modifications such as increasing exercise levels may help prevent development of dementia in later life (Larson et al., 2006).

The ageing of the population is a major cause for concern in developed nations, which will see an explosion of people entering the over-65 age group in the next 10-20 years. The major form of dementia, Alzheimer's disease (over 60% of diagnosed dementia cases) (Jorm, 2001), is particularly pernicious as it eventually causes deterioration of the brain areas responsible for autonomous living and self-care. Although a person with the early stages of Alzheimer’s disease can be cared for adequately at home by relatives, in the later stages of the disease, costly institutional care is required (Brodaty et al., 2003b).

This chapter describes dementia, its relative importance in terms of global burden of disease, and its pathophysiological and neuropsychological impacts on individuals, and the social impacts on families and society. Various therapeutic possibilities and management strategies are then outlined.

2.1 Dementia - impacts

Dementia is primarily a condition associated with ageing, but can be brought about by trauma, illness and substance abuse. Dementia is described as follows, in the World Health Organisation’s International Classification of Diseases:

*Dementia (F00-F03) is a syndrome due to disease of the brain, usually of a chronic or progressive nature, in which there is disturbance of multiple higher cortical functions, including memory, thinking, orientation, comprehension, calculation, learning capacity, language, and judgement. Consciousness is not clouded. The impairments of*
cognitive function are commonly accompanied, and occasionally preceded, by deterioration in emotional control, social behaviour, or motivation. This syndrome occurs in Alzheimer’s disease, in cerebrovascular disease, and in other conditions primarily or secondarily affecting the brain. (WHO, 2003)

Diagnostic criteria for Alzheimer’s disease, the most prevalent form of dementia, are outlined in the Diagnostic and Statistical Manual of Mental Disorders:

A. The development of multiple cognitive deficits manifested by both:

1) Memory impairment (impaired ability to learn new information or to recall previously learned information);

2) One or more of the following cognitive disturbances: aphasia (language disturbance) apraxia (impaired ability to carry out motor activities despite intact motor function) agnosia (failure to recognize or identify objects despite intact sensory function) disturbances in executive functioning (i.e., planning, organizing, sequencing, abstracting)

B. The cognitive deficits in Criteria A1 and A2 each cause significant impairment in social and occupational functioning and represent a significant decline from a previous level of functioning.

C. The course is characterized by gradual onset and continuing cognitive decline

D. The cognitive deficits in Criteria A1 and A2 are not due to any of the following:

(1) other central nervous system conditions that cause progressive deficits in memory and cognition (e.g., cerebrovascular disease, Parkinson’s Disease, Huntington’s Disease, subdural hematoma, normal pressure hydrocephalus, brain tumor).

(2) systemic conditions that are known to cause a dementia (e.g., hypothyroidism, vitamin B12 or folic acid deficiency, hypercalcemia, neurosyphilis, HIV infection)

(3) substance-induced conditions

E. The deficits do not occur exclusively during the course of a delirium

F. The disturbance is not better accounted for by another Axis 1 disorder (e.g., Major Depressive Disorder, Schizophrenia) (APA, 1994).

The diagnosis of Alzheimer’s disease is a diagnosis of exclusion. There is rarely an identifiable cause, so clinical diagnosis relies on assessment of cognitive impairment. Neuropathological diagnosis of Alzheimer’s disease is confirmed at autopsy.

Different types of dementia have been characterised by neuropathological and clinical criteria, but they all involve death of cortical and/or subcortical neurons and chronic neurochemical
imbalances. Individuals may also have pathologies of one or more types of dementia at the same time (WHO, 2003).

There are four major types of dementia: Alzheimer’s disease; vascular dementia; dementia with Lewy bodies and Pick’s disease. Their neuropathological and clinical features, possible causes, risk factors and the prevalence of the type of dementia are summarised below (see Table 2.1). Alzheimer’s disease comprises 50-70% of all dementia cases, and vascular dementia comprises 20-30%. Other diseases that can result in dementia are Parkinsonism, Huntington’s chorea and Creutzfeldt-Jakob disease. The latter two are easily distinguished from the other types of dementia, and are much less common. Down’s syndrome and AIDS also can result in dementia in the final stages of life, as can chronic alcohol abuse (Access-Economics, 2003).

Table 2.1 Dementia types, features and risk factors (Access-Economics, 2003)

<table>
<thead>
<tr>
<th>Dementia type</th>
<th>Neuropathology</th>
<th>Clinical</th>
<th>Risk factors</th>
<th>Percentage of all dementia cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alzheimer’s disease</td>
<td>Progressive atrophy of grey matter in brain from hippocampus through to the frontal cortex; neurofibrillary tangles inside neurons; amyloid plaques outside neurons; deficiency in acetylcholine production and transmission; elevated glutamate receptor activity</td>
<td>Progressive memory loss, language difficulties and various neuropsychiatric symptoms including personality changes;</td>
<td>Family history; Down’s syndrome; AIDS; age</td>
<td>50-70%</td>
</tr>
<tr>
<td>Vascular dementia</td>
<td>Narrowing of brain arteries resulting in chronic reduced oxygen supply; onset after major strokes or multiple transient ischaemic attacks causing lesions in the cortex; sometimes involving demyelination of subcortical white matter</td>
<td>Similar to Alzheimer’s disease, but onset triggered by stroke; often a stepped decline; weakening of specific part of body due to focal neurological damage; patchy loss of cognitive function.</td>
<td>Vascular risk factors</td>
<td>20-30%</td>
</tr>
<tr>
<td>Dementia with Lewy bodies</td>
<td>Presence of abnormal deposits of alpha-synuclein in neurons, particularly in frontal lobe and visual cortex; loss of neurons and dopamine production in the substantia nigra (hence Parkinson-type traits); some amyloid plaques like Alzheimer’s, but not neurofibrillary tangles.</td>
<td>Similar to Alzheimer’s disease, but some Parkinson-type motor traits, visual hallucinations, marked daily fluctuations in cognitive function and falls and fainting.</td>
<td>Family history</td>
<td>About 10%</td>
</tr>
<tr>
<td>Fronto-temporal lobe dementia (Pick’s disease)</td>
<td>Loss of neurons in fronto-temporal lobes; tangles similar to Alzheimer’s disease; abnormal accumulation of Pick bodies</td>
<td>More marked personality and behavioural changes, especially socially inappropriate disinhibition. Obsessive-compulsive type behaviours. Muteness in final stages</td>
<td>Family history</td>
<td>Rare - 1 in 5,000, more common in pre-senile dementia</td>
</tr>
</tbody>
</table>
2.1.1 Factors implicated in the genesis of dementia

While there is no single cause of dementia, the following factors increase the risk of dementia:

- increasing age;
- family history of dementia (particularly for pre-senile dementia);
- presence of ε4 allele at the apolipoprotein E gene on chromosome 19;
- Down’s syndrome; and
- vascular disease risk factors (e.g. high blood pressure, atherosclerosis, ischaemic heart disease, high cholesterol, diabetes, smoking, heart attacks and strokes) (Messier et al., 2004; Richards & Hendrie, 1999).

Certain endocrine and nutritional disorders can also result in dementia, such as hypothyroidism, and B-vitamin deficiency, particularly B12, folate and niacin. These disorders should be ruled out prior to a diagnosis of dementia (APA, 1994).

2.1.1.1 Neuropsychological effects of dementia

The deterioration of cognitive function includes:

- loss of procedural memory or the ability to perform previously learned skills, and
- loss of executive function or the ability to plan and organise complex activities (Richards & Hendrie, 1999).

Language and people recognition skills are usually affected, and as the dementia worsens, activities of daily living such as personal hygiene and feeding are impaired (Access-Economics, 2003).

2.1.1.2 Behavioural and psychological changes

The behavioural and psychological symptoms that often accompany dementia are perhaps even more disturbing than the loss of cognitive function. They are also referred to as neuropsychiatric symptoms, and were summarised into the following categories: apathy, depression and dysphoria, agitation, anxiety, irritability, aberrant motor behaviour, disinhibition, delusions, hallucinations and euphoria (Cummings, 1997).
According to Kaufer (2002), apathy or social withdrawal is the most prevalent symptom and occurs in about 60-70% of people with Alzheimer’s disease. Apathy could be due to lack of ability to focus attention on events, and a person’s withdrawal could be a coping mechanism in response to an overwhelming multiplicity of demands for attention.

Persistent apathy could lead to decreases in serotonergic transmission, thereby leading to dysphoria and later, depression. This is supported by the observation that in Alzheimer’s disease, both the locus coeruleus and dorsal raphe nuclei are affected by amyloid deposits, interrupting the normal release of both noradrenaline and serotonin in the hippocampus (Kar et al., 2004).

Agitation, anxiety and irritability are other key features in Alzheimer’s disease and other dementias (Cummings, 1997), and include agitated motor behaviours such as anxious pacing and rummaging without finding things (Cohen-Mansfield et al., 1990). Sleep-wake disturbances are also common in Alzheimer’s disease, and these may contribute to the memory deficits given the importance of sleep for memory consolidation (Kaufer, 2002). Aberrant motor behaviour, disinhibition, delusions and hallucinations are all associated with later stages of the disease, although not every person experiences these symptoms (APA, 1994).

### 2.1.2 Global ageing and dementia

Population ageing is a serious concern for many developed countries. In the Global Burden of Disease study in 1997, predictions were made about the likely causes of disability and death in developed and developing nations in 2020 (Murray & Lopez, 1997). By 2020 the population aged 65 and over in developed regions will have increased by 71% of its 1996 size, whereas the 15-44 age group is predicted to decline. In developing regions, the 44-59 age groups will increase by 140%.

Dementia is currently rated eighth out of the top ten conditions predicted to be the main causes of disability in developed regions in 2020. It is projected to comprise 0.4%-0.7% of the total global burden of disease in 2020 (Murray & Lopez, 1997, WHO, 2004) (see Table 2.2). In the developed world the 2020 main projected causes of disease are the non-communicable diseases, several of which are associated with ageing, for example osteoarthritis, dementia, ischaemic heart disease and cerebrovascular disease (Murray & Lopez, 1997).
Table 2.2 Ten projected leading causes of disease affected life years (DALYs) in 2020 derived from (Murray & Lopez, 1997).

<table>
<thead>
<tr>
<th>Developed world</th>
<th>Disease or injury</th>
<th>DALYs $(x10^6)$</th>
<th>%</th>
<th>Cumulative %</th>
<th>Disease or injury</th>
<th>DALYs $(x10^6)$</th>
<th>%</th>
<th>Cumulative %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Developed world</td>
<td>All causes</td>
<td>160.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Developed world</td>
<td>Ischaemic heart disease</td>
<td>18.0</td>
<td>11.2</td>
<td>11.2</td>
<td>Unipolar major depression</td>
<td>68.8</td>
<td>5.6</td>
<td>5.6</td>
</tr>
<tr>
<td>Developed world</td>
<td>Cerebrovascular disease</td>
<td>9.9</td>
<td>6.2</td>
<td>17.4</td>
<td>Road traffic accidents</td>
<td>64.4</td>
<td>5.2</td>
<td>10.8</td>
</tr>
<tr>
<td>Developed world</td>
<td>Unipolar depression</td>
<td>9.8</td>
<td>6.1</td>
<td>23.5</td>
<td>Ischaemic heart disease</td>
<td>64.3</td>
<td>5.3</td>
<td>16.1</td>
</tr>
<tr>
<td>Developed world</td>
<td>Respiratory system cancers</td>
<td>7.3</td>
<td>4.5</td>
<td>28.0</td>
<td>Chronic obstructive pulmonary disease</td>
<td>52.7</td>
<td>4.3</td>
<td>20.4</td>
</tr>
<tr>
<td>Developed world</td>
<td>Road traffic accidents</td>
<td>6.9</td>
<td>4.3</td>
<td>32.3</td>
<td>Cerebrovascular disease</td>
<td>51.5</td>
<td>4.2</td>
<td>24.6</td>
</tr>
<tr>
<td>Developed world</td>
<td>Alcohol use</td>
<td>6.1</td>
<td>3.8</td>
<td>36.1</td>
<td>Tuberculosis</td>
<td>42.4</td>
<td>3.4</td>
<td>28.0</td>
</tr>
<tr>
<td>Developed world</td>
<td>Osteoarthritis</td>
<td>5.6</td>
<td>3.4</td>
<td>39.5</td>
<td>Lower respiratory infections</td>
<td>41.1</td>
<td>3.4</td>
<td>31.4</td>
</tr>
<tr>
<td>Developed world</td>
<td>Dementia and other degenerative &amp; hereditary CNS disorders</td>
<td>5.5</td>
<td>3.5</td>
<td>43.0</td>
<td>War injuries</td>
<td>40.2</td>
<td>3.2</td>
<td>34.6</td>
</tr>
<tr>
<td>Developed world</td>
<td>Chronic obstructive pulmonary disease</td>
<td>4.9</td>
<td>3.0</td>
<td>46.0</td>
<td>Diarrhoeal diseases</td>
<td>37.0</td>
<td>3.0</td>
<td>37.6</td>
</tr>
<tr>
<td>Developed world</td>
<td>Self-inflicted injuries</td>
<td>3.9</td>
<td>2.4</td>
<td>48.4</td>
<td>HIV</td>
<td>34.0</td>
<td>2.8</td>
<td>40.4</td>
</tr>
</tbody>
</table>

The developed world includes established market economies and formerly socialist economies of Europe; DALY = Disability Adjusted Life Year.

2.1.2.1 Ageing and dementia in Australia

The incidence of dementia in Australia after the age of 65 is about 2%, and it doubles every 5 years, with 32% of people over 85 being likely to have some form of dementia. In Australia in 2004, the 65 and over age group had over 2.6 million people, that is, about 13% of the population (DOHA, 2005). The number of people over 65 is predicted to increase to about 5.7 million by 2041, which will be about 23% of the total population (Aged Care Ministerial Reference Group, 2000).

In 2002, dementia affected an estimated 162,300 Australians (0.8% of the population) and is estimated to increase to 500,000 (about 2%) by 2040 (Access-Economics, 2003). In 2002, 1.1% of all Australian women had dementia, and 0.6% of men. These figures did not include cases of undiagnosed mild to moderate disease, and are therefore likely to be underestimates.

An epidemic increase in the number of Australians with dementia is expected in the next 30 years due to the ageing of the population, although the incidence rate of the disease is not expected to change (DOHA, 2005). Perhaps the main societal concern about an epidemic increase of dementia is that people with dementia experience a gradual decline in the ability to...
live independently: 94% of people with dementia were classified as having profound restrictions to key independent-living abilities like communication, mobility and self-care (ABS, 1998). Not only does disability reduce enjoyment of life for the individual, but it adds responsibility, worry, stress and financial burden to relatives, and eventually to all tax-payers via the health care system (Aged Care Ministerial Reference Group, 2000).

2.1.2.2 Australian health costs of dementia

The Australian health expenditure budget in 2003-04 was estimated to be $78,369 million, which was about 9.7% of the gross domestic product (GDP) for that year (AIHW, 2005). In 2000-01 health expenditure was $61,635 million and of this, $2,228 million (or about 3.6%) was attributed to Alzheimer’s disease and other dementias (AIHW, 2005). However, the amount spent on high level residential care (nursing homes) was $4,985 million, about 6.4% of the health budget (AIHW, 2005).

While the exact percentage of aged care facility residents with dementia is difficult to calculate, it is estimated that more than 60% of all aged care residents have dementia (Rosewarne, 1997). Therefore, national dementia management strategies have focused on delaying institutionalisation, either by increasing support to care-givers or using pharmacotherapy to help prevent progression of the disease. Ideally, healthy life-style training ought to be implemented during high-school to prevent the development of the known dementia risk factors, such as cardiovascular disease.

2.1.3 Assessment of dementia

Initial memory difficulties that may develop into dementia are usually first noticed by close family and friends and by the at-risk person themselves. While memory function can slow down with normal ageing, mild cognitive impairment is thought to be a harbinger of dementia in later life (Morris et al., 2001). Formal diagnoses of mild cognitive impairment and dementia are usually carried out by clinical examination and cognitive testing, and can be confirmed by more detailed neuropsychological tests and brain imaging if required (Richards & Hendrie, 1999).

Several scales and tests have been devised to determine an individual’s cognitive status. These can be coupled with scales that measure daily living abilities, mood and behaviours to
get a full picture of the individual’s condition, either during initial diagnosis or over time, for example in a clinical trial. Clinicians also use simple global change scales to grade their impressions of overall change in dementia severity.

### 2.1.3.1 Cognitive function tests

Cognitive function tests can either test individual aspects of cognitive function, or several aspects at once. Two of the most commonly used screening tests are the cognitive subscale of the Alzheimer’s Disease Assessment Scale (ADAS-cog) (Rosen et al., 1984) and the Mini-Mental State Examination (MMSE) (Folstein et al., 1975, Molloy et al., 1991).

The ADAS-cog must be administered by a trained clinician and takes about 45 minutes (Burns et al., 1999). It has 74 questions, covering spoken language ability, recall of test instructions and ability to follow commands, word-finding difficulty, object naming, construction of drawings, sending oneself a letter (testing ideational praxis), spatial and temporal orientation, word recall and recognition. Test-retest reliability was found to be 0.93 for the ADAS-cog, indicating its usefulness in repeat screening (Weyer et al., 1997). The authors also suggested that a change of 7 points on the 74 point ADAS-cog was needed for a reliable indication of change, using reliability estimates.

The MMSE can be used by nursing staff and non-clinicians and takes about 10 minutes to administer (Walaszek & Snowden, 2004, van Gorp et al., 1998). It has 30 questions covering orientation (spatial and temporal), recall and retention of words, language (spelling, reading, writing and obeying orders) and a construction drawing task. MMSE test-retest reliability was reviewed and the authors suggested that the reliability coefficients between 0.80-0.95 were those most likely to be accurate (Tombaugh & McIntyre, 1992). An increase or decrease of 4 points over a sixteen week test period was found to be statistically significant (Sands et al., 1999), and is also likely to be clinically important (Burback et al., 1999). Studies of cognitive function over time need to take into account the approximate natural deterioration rate of 2.8 points per year or 1.4 points per six months as reported by Burback et al, (1999).

In several studies of the effects of cholinesterase inhibitors on cognitive function, both the ADAS-cog and MMSE have been used (Orgogozo et al., 2002, Requena et al., 2004, Sands et al., 1999, Doraiswamy et al., 1997). Although the ADAS-cog is considered the ‘gold standard’ for cognitive function trials, significant correlations between the ADAS-cog and
MMSE have been described (Doraiswamy et al., 1997). The MMSE can be used in place of the ADAS-cog in mild to moderate dementia (Ihl et al., 1992) provided that, in studies with multiple raters, there is a pre-determined agreement on how to score the most variable item of the test, the “Spell the word ‘WORLD’ backwards” question (Weyer et al., 1997).

2.1.3.2 Behavioural, mood and functional tests

There are many different tests for this area, either focusing on one particular aspect of behaviour or mood or daily living, or combining several aspects in the one test. For example, the Cohen-Mansfield Agitation Index (Cohen-Mansfield et al., 1990) focuses specifically on behaviours related to agitation, whereas the Neuropsychiatric Inventory (Cummings, 1997) includes apathy, anxiety, disinhibition, and aberrant motor behaviour amongst other neuropsychiatric states, as well as agitation.

The Nurses’ Observation Scale for Geriatric Residents (NOSGER) (Spiegel et al., 1991) is a 30 point scale with 6 pre-defined factors relating to mood, memory, activities of daily living, social behaviour, disturbed behaviour and independent activities of daily living. Data is collected by carers, making it useful for residential care situations. The test-retest reliability coefficient of the NOSGER was above 0.8 for all the sub-factors, although it was higher for the cognitive dimensions than the non-cognitive ones (Wahle et al., 1996).

2.1.3.3 Global scales

Global impressions of change scales are used to complement cognitive, functional, mood and behavioural scales. They are generally carried out by clinicians responsible for the individual, and can include interviews with carers and the individual in question. It is hard to validate scales like these, as they rely on the subjective impressions of the clinicians, but it is thought that any change on these scales is likely to be clinically significant, and they are therefore often used in dementia drug trials (Burns et al., 1999).

2.1.3.4 Olfactory testing in dementia

As age increases, olfactory perception declines (Schiffman, 1991, Hummel et al., 1998). In particular, a decrease in the ability to distinguish between odorants and a more rapid adaptation to odours characterises the decline in olfactory ability in ageing. While reduced
olfactory function has been associated with Alzheimer’s and Parkinson’s disease (Mesholam et al., 1998). Schiffman reports that chronic renal failure, vitamin B12 deficiency, hypothyroidism, diabetes mellitus and adrenal cortical insufficiency also reduce olfactory abilities (Schiffman, 1991). Some of these conditions are co-morbid with dementia (APA, 1994).

A meta-analysis of olfactory deficits in Alzheimer’s and Parkinson’s disease concluded that odour identification, recognition and detection thresholds in these diseases is significantly impaired or reduced compared to controls (Mesholam et al., 1998). Pathological changes occur in the olfactory bulb and the entorhinal cortex of Alzheimer patients (Kovacs et al., 2001) suggesting that early loss of olfactory ability may be an indicator of future cognitive decline.

In a study of 90 people with mild cognitive impairment, 19 of whom developed Alzheimer’s disease at 2 year follow-up, initial low olfaction scores on the University of Pennsylvania Smell Identification Test and lack of awareness of olfactory deficit were predictors of Alzheimer’s disease development (Devanand et al., 2000).

Brain-imaging of the olfactory process in people with Alzheimer’s disease can indicate which brain areas are compromised, and therefore whether olfactory reception and cognitive processing of olfactory input are impaired (Henkin, 2000). However, a decrease in olfactory ability does not necessarily predict the development of dementia, but can be due to infection of the nasopharyngeal cavity or the growth of nasal polyps (Henkin, 2000).

2.1.4 Pathophysiology of Alzheimer’s disease.

Although Alzheimer’s disease can only be confirmed on autopsy, it is the most common type of dementia, and several of its symptoms are common to the other dementias. For the purposes of this project, Alzheimer’s disease was the dementia-type targeted for review, and its pathophysiology, and current pharmacotherapy approaches are described.

2.1.4.1 Plaques and tangles

The main pathology in Alzheimer’s disease is the progressive death of neurons. The salient pathophysiological changes apart from dead neurons are: intraneuronally, the development of
neurofibrillary tangles and neuropil threads; extraneuronally, the development of so-called ‘senile plaques’ comprised of amyloid protein (Braak & Braak, 1996).

Neurofibrillary tangles and neuropil threads are made of abnormally phosphorylated tau protein. The neurofibrils are normally used to stabilise axonal microtubule formation, especially in neurons with long unmyelinated axons, like those that project from the limbic system to the cortex (Lovestone & McLoughlin, 2002). Neuropil threads occur in the distal ends of the dendrites. In Alzheimer’s disease, the tau proteins tangle up, rather than helping to support the cytoskeleton, though it has not yet been established whether the presence of tangles cause neuronal death, or whether it is loss of normal microtubule function that causes it (Lovestone & McLoughlin, 2002).

Senile plaques are comprised mainly of another protein known as beta-amyloid, which is an abnormal break-down product of the amyloid precursor protein controlled by secretase enzymes (Pepe & Curtiss, 1986). Although the plaques appear first in the pathogenesis of Alzheimer’s disease, and amyloid precursor protein may be implicated in development of tau pathology, there is still doubt about the extent to which senile plaques contribute to neuronal death (Lovestone & McLoughlin, 2002).

Apolipoprotein E is normally part of the immunoregulatory low-density lipoprotein (LDL-In) that controls transport of triglycerides, phospholipids and cholesterol in and out of neurons (Pepe & Curtiss, 1986). It is used in membrane remodelling and maintenance of synaptic plasticity (Lovestone & McLoughlin, 2002, Hashimoto et al., 2002). Mutations to the apolipoprotein E gene, in particular the Apoε4 variant, appear to exacerbate beta-amyloid deposition (Schmechel et al., 1993).

The plaques and tangles begin to form in areas of the brain associated with short-term memory formation, such as the entorhinal cortex and the hippocampus (Braak & Braak, 1996). The plaques also cause an inflammatory response from the surrounding glial cells, that in turn appears to accelerate neuronal death (Gao et al., 2002).

These pathophysiological changes are accompanied by impairments to most of the well-known neurotransmitter systems, as comprehensively reviewed by Gsell et al. (2004). The neurochemical changes also offer potential drug targets, so will be briefly discussed below prior to a discussion of Alzheimer’s disease pharmacotherapy.
2.1.4.2 Cholinergic impairment

Cholinergic neurons in the basal forebrain demonstrate pathological changes in the reduction of acetylcholine production, which in turn interfere with normal memory tasks in neurons in cortical association areas of the frontal, temporal and parietal lobes (Kaufer, 2002). The main impairment is a reduction in the activity of cholineacetyltransferase (ChAT), an essential intraneuronal enzyme for the synthesis of acetylcholine. The reduction in ChAT activity and acetylcholine levels occurs mainly in hippocampal and amygdalic neurons of the limbic system (Gsell et al., 2004). Nicotinic acetylcholine receptor densities also decrease, especially in the frontal cortex, although muscarinic acetylcholine receptors remain at near normal levels (Svensson et al., 1997).

Inadequate cholinergic supply means that the cortical neurons innervated by the impaired basal neurons function at less than their usual level, and eventually become susceptible to the neurofibrillary tangles and neuronal death (Kaufer, 2002).

2.1.4.3 Glutamatergic impairment

Alterations to the glutamatergic transmission in the hippocampus and cortical association areas also appear in Alzheimer’s disease (Kornbuber & Wiltfang, 1998). Reduction in the viability of transporter and re-uptake systems (Gsell et al., 2004) leads to excessive synaptic glutamate levels. Excessive levels of glutamate tend to cause over-excitation of neurons, leading to a host of mood and behavioural factors like anxiety and agitation, and eventually, neuronal death (Areosa Sastre et al., 2005).

2.1.4.4 GABA-ergic impairment

Gamma-amino butyric acid (GABA) is an inhibitory neurotransmitter that often counteracts the effects of glutamate and aspartate excitation. There are two types of CNS GABA receptor. GABA_A receptor agonists appear to be involved in reducing the synaptic transmission of acetylcholine, dopamine and serotonin production (Lanctôt et al., 2004). The GABA_A receptor site also has binding sites for benzodiazepines, barbiturates, steroids and ethanol that in turn modulate the GABA_A receptor function (Zorumski & Isenberg, 1991). GABA_B receptors on glutamate neurons inhibit release of glutamate and postsynaptically reduce neuronal excitability in hippocampal neurons (Lanctôt et al., 2004).
In Alzheimer’s disease, GABA-uptake is reduced, particularly in the limbic system and entorhinal cortex. There are significant decreases in GABA production and receptor density, though different subregions of the brain vary as to the receptor sub-type reduced (Gsell et al., 2004).

### 2.1.4.5 Noradrenergic impairment

The dorsal noradrenergic fibre bundle ascending from the locus coerulus appears to be the most impaired in Alzheimer’s disease (Gsell et al., 2004). The main targets are the amygdala, cholinergic nuclei in Broca’s area, the hippocampus and entorhinal cortex. Noradrenaline turnover is increased in these areas in Alzheimer’s disease, resulting in lowered synaptic noradrenaline levels. Decreases in receptor density are not found in these areas but in thalamic regions and the cerebellum (Gsell et al., 2004).

### 2.1.4.6 Dopaminergic impairment

As for noradrenergic impairment, dopaminergic impairment in Alzheimer’s disease is largely due to increased turnover by monoamine oxidase-B, rather than reduction in dopamine production or receptor density (Gsell et al., 2004).

Dopamine levels are decreased in the substantia nigra as in Parkinson’s disease, though unlike Parkinson’s disease, amygdala levels of dopamine increase in Alzheimer’s disease, compared to controls. These two impairments may account for the increased wandering and fidgeting in Alzheimer’s disease, and increased or sustained levels of emotional responsiveness, respectively (Gsell et al., 2004).

### 2.1.4.7 Serotonergic impairment

In Alzheimer’s disease, the ventral ascending serotonergic pathway is significantly impaired. This pathway has projections to the hypothalamus and thence to the amygdala, hippocampus and the cholinergic cells of Broca’s area and the septal nuclei. The losses are in both serotonin production and serotonin receptor expression, with an increase in the turnover rate of existing serotonin (Gsell et al., 2004). This results in an overall depletion of synaptic serotonin.

Deficits in the serotonin receptor 5HT₁ subtype were thought to be associated with increased anxiety, aggression and depression, though more recent studies have not been able to
substantiate earlier research (Lanctôt et al., 2001). Serotonin-producing neurons of the raphe nuclei are often vulnerable to neuronal death in Alzheimer’s disease, although the onset of symptoms often occurs after symptoms of acetylcholine deficiency arise (Zarros et al., 2005). More research is needed to unravel the complex interactions of serotonin and acetyl choline transmission in Alzheimer’s disease.

2.1.5 Cognitive and behavioural changes in Alzheimer’s disease

2.1.5.1 Cognitive changes

Alzheimer’s disease progresses slowly in most late-onset cases (that is, after age 65). Death from Alzheimer’s disease often occurs four to eight years after the initial diagnosis. In the early stages, the inability to remember recent past events and to switch attention from one task to another appear to be the most obvious changes (RJ Perry et al., 2000, Baddeley et al., 2001, Kaufer, 2002).

The ability to pay attention depends largely on the adequate functioning of cholinergic transmission (Perry & Hodges, 1999), although noradrenergic, serotonergic and dopaminergic systems all contribute to the full complexity of memory function (Scholey, 2002).

The major cholinergic activity arising from the basal forebrain and terminating in frontal and posterior parietal lobe and thalamic targets is crucial in focussing attention when the brain is involved in specific cognitive tasks (Scholey, 2002). The noradrenergic system appears to affect orientation of attention, enabling the brain to ignore novel distractions. The dopaminergic system appears to assist in shifting of attention, and serotonin probably assists with memory consolidation, given that serotonin depletion is shown to impair memory consolidation (Scholey, 2002).

While this is an extremely brief overview, it is sufficient to note that many neurotransmitter systems contribute to memory and attention. Disruptions to any of these systems may have consequences for memory function, either short-term (e.g. when drug-induced) or permanent (e.g. due to trauma or dementia).
2.1.6  Therapeutic targets for Alzheimer’s disease

As Alzheimer’s disease is a complex syndrome with multiple factors, there are several potential therapeutic targets. However, despite the varied target options, the Alzheimer’s disease pharmacotherapy research effort appears to have been largely focused on the cholinergic system since the mid-1970s.

More recently, agents that prevent amyloidogenesis have been investigated, along with NMDA-glutamate antagonists, antioxidants, anti-inflammatory agents, oestrogen and heavy metal chelation therapy (Kar et al., 2004).

2.1.6.1 Cholinergic drug targets

As the cholinergic function in basal forebrain cholinergic neurons is among the first known deficits in early Alzheimer’s disease, attempts have been made to boost cholinergic function. If cholinergic deterioration can be halted, or at least retarded, then the number of years a person spends with severe dementia is likely to be reduced to the increased likelihood of death from other age-related causes (Ibach & Haen, 2004). Retardation of the inevitable entry into institutionalised care is also preferable, both socially and financially.

Acetylcholinesterase inhibition (and more recently butyrylcholinesterase inhibition), acetylcholine agonism and trophic support for cholinergic neurons are all valid remedial targets, although cholinesterase inhibition has received the most attention (Auld et al., 2002).

2.1.6.1.1 Acetylcholinesterase inhibition

Inhibition of acetylcholinesterase artificially raises the levels of synaptic acetylcholine, thereby improving cholinergic transmission (Ibach & Haen, 2004, Grutzendler & Morris, 2001). Three cholinesterase inhibiting drugs are available, with modest clinical efficacy and varying side-effects: donepezil, galantamine and rivastigmine (Lanctôt et al., 2003). Improvement in cognitive function as measured by the MMSE (Folstein et al., 1975) and the ADAS-cog (Rosen et al., 1984) is often considered to be evidence of primary efficacy for cholinesterase inhibitors (Holden & Kelly, 2002).
Donepezil hydrochloride $C_{24}H_{29}NO_3$ is a water-soluble piperidine derivative. A systematic review revealed that most studies used daily doses of 5 mg and 10 mg of donepezil hydrochloride. Daily doses of 10 mg provided statistically significant improvements compared to placebo on the MMSE and ADAS-cog, as well as a series of other scales focusing on behavioural changes and activities of daily living (Birks & Harvey, 2006).

The meta-analysis for 12 week studies of 10 mg doses (6 studies) reported a maximum mean ADAS-cog change (sd) for donepezil of -2.77 (5.48), placebo 0.40 (5.42) (n = 139 ), with the smallest mean change being donepezil -0.15 (5.63), placebo 1.06 (6.28) (n = 19 ).

The meta-analysis for 12 week-studies of 10 mg doses (6 studies) using the MMSE reported a maximum mean (sd) change for donepezil of 3.52 (3.26), placebo -0.83 (3.33) (n = 5 ). The smallest mean change was donepezil 0.69 (2.89), placebo -0.18 (3.08) (n = 128 ).

Donepezil has the following side-effects:
- Insomnia 1 in 10
- Nausea and vomiting 1 in 10
- Diarrhoea 1 in 10
- Dizziness occurs 1 in 12.5

Galantamine $C_{17}H_{21}NO_3$ is derived from several bulb plants, including daffodils, and is water soluble. The drug form is presented as galantamine hydrobromide. In addition to inhibiting acetylcholinesterase, it modulates nicotinic acetylcholine receptors, making them more able to respond to acetylcholine (Olin & Schneider, 2001).

A systematic review revealed that daily doses of between 8 and 36 mg of galantamine hydrobromide provided statistically significant improvements compared to placebo on the ADAS-cog, as well as a series of other scales focusing on behavioural changes and activities of daily living (Loy & Schneider, 2006). The MMSE was not used as a cognitive test for galantamine.
The meta-analysis for 12 week studies of galantamine (7 studies) reported a maximum mean ADAS-cog change (sd) for galantamine (16-24 mg b.i.d.) of -2.60 (5.07) , placebo 0.00 (5.31) (n = 275 ), with the smallest mean change being galantamine (18 mg b.i.d.) -0.80 (6.23) , placebo 2.30 (6.55) (n = 53 ).

The most frequent side effects seen with galantamine are:

- Nausea 1 in 6
- diarrhoea 1 in 8
- vomiting 1 in 10
- anorexia (loss of appetite), and weight loss (not specified) (Saltiel, 2006).

These side effects generally occur during the beginning of treatment or when the dose is increased. These side effects typically are mild and temporary.

Rivastigmine tartrate C$_{18}$H$_{28}$N$_2$O$_8$ is a water-soluble arylcarbamate derivative, and a non-competitive reversible inhibitor of acetylcholinesterase. A systematic review revealed that only the higher daily doses of 6-12 mg of rivastigmine tartrate provided statistically significant improvements compared to placebo on the ADAS-cog and the MMSE, as well as a series of other scales focusing on behavioural changes and activities of daily living (Birks et al., 2000).

The meta-analysis for 12 week studies of rivastigmine (4 studies) reported a maximum mean ADAS-cog change (sd) for rivastigmine (6-12 mg daily) of -1.48 (5.60), placebo -0.13 (5.70) (n = 238 ), with the smallest mean change being rivastigmine (6-12 mg daily) 0.35 (4.60), placebo 0.80 (4.60) (n = 171 ).

The meta-analysis for 26 week-studies (in the absence of 12 week studies) of 6-12 mg doses (4 studies) using the MMSE reported a maximum mean (sd) change for rivastigmine of 0.60 (3.60), placebo 1.40 (3.60) (n = 220 ). The smallest mean change was rivastigmine -0.22 (3.50), placebo 0.50 (3.60) (n = 239 ). Interestingly, these MMSE results appear to indicate that people taking rivastigmine fared worse than those on placebo, as a positive change indicates an improvement.

The worst side-effects of rivastigmine are reported as:

- nausea 1 in 2
• diarrhoea 1 in 7
• vomiting 1 in 3
• anorexia 1 in 4.5
• dizziness 1 in 50 (Saltiel, 2003)

2.1.6.1.2 Comparison of efficacy of the three cholinesterase inhibitors

To compare the efficacy of the three cholinesterase inhibitors, the optimum dose weighted mean differences for studies at least six months in length were summarised from a systematic review by Birks (2006) (see Table 2.3).

Table 2.3 Largest weighted mean difference from meta-analyses of three different cholinesterase inhibitor drugs, donepezil, galantamine and rivastigmine, with optimum dose and at least 6 months study length (Birks, 2006).

<table>
<thead>
<tr>
<th>Drug</th>
<th>ADAS-cog N subjects</th>
<th>ADAS-cog Mean difference [95% CI]</th>
<th>MMSE N subjects</th>
<th>MMSE Mean difference [95% CI]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Donepezil</td>
<td>518</td>
<td>-2.92 [ -3.87, -1.97 ]</td>
<td>270</td>
<td>1.79 [ 0.84, 2.74 ]</td>
</tr>
<tr>
<td>Galantamine</td>
<td>409</td>
<td>-3.90 [ -5.03, -2.77 ]</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Rivastigmine</td>
<td>448</td>
<td>-1.60 [ -2.93, -0.27 ]</td>
<td>466</td>
<td>2.90 [ 2.36, 3.44 ]</td>
</tr>
</tbody>
</table>

ADAS-cog = Alzheimer’s Disease Assessment Scale (cognitive subscale); MMSE = Mini-Mental State Examination

While the cognitive improvements for the three cholinesterase inhibitors are statistically significant, the clinical significance of such changes is not clearly established. The most important benefit of cognitive improvement by cholinesterase inhibitors is the potential delay of admission into institutional care by up to twelve months (Holden & Kelly, 2002).

Donepezil has been in use for the longest period, and also appears to be preferred as only one daily dose and one titration period are required. Donepezil also appears to have fewer serious side-effects than either rivastigmine or galantamine (Thompson et al., 2004). There is not sufficient evidence from head-to-head trials to determine superiority of one drug over another, and although the changes in cognitive test scores are statistically significant, cognitive function of patients taking the medications only improves mildly from a clinical point of view (Grutzendler & Morris, 2001, Burbak et al., 1999, Winblad et al., 2001, Rockwood, 2004).
Although the three available drugs are most often prescribed for people with early or mild probable Alzheimer’s disease, a review of cholinesterase inhibitors showed that anticholinesterase drugs can help delay the decline in cognitive functioning in every stage of the disease, not just during the early stages as previously thought (Kurz et al., 2004).

Other reviews (Winblad et al., 2001, Trinh et al., 2003) found that cholinesterase inhibition therapy also has a beneficial effect on behavioural and psychological symptoms of dementia, though these benefits are quite small. The moderate usefulness of AChE inhibitors suggests the need for discovery and development of novel drugs with greater efficacy.

2.1.6.1.3 Other cholinergic drug targets

Other cholinergic system drug targets are the muscarinic and nicotinic ACh receptors. These have been studied using nicotine and other agonists, with the intention of improving cognitive function by mimicking acetylcholine (Johnson et al., 2002) (Mihailescu & Drucker-Colin, 2000, Auld et al., 2002). Several drugs are in development, but apart from nicotine, none are available commercially as yet.

Similarly, the role of nerve growth factor (NGF) in the preservation of cholinergic neurons is being investigated, with suggestions that it or NGF-mimetic compounds can nurture the cholinergic neurons and also provide cognitive improvements (Auld et al., 2002). However, direct infusion of NGF into the cerebrospinal fluid has unacceptable side-effects like severe back-pain.

2.1.6.2 Amyloid drug targets

As amyloid plaque formation appears to be one of the first pathological developments in Alzheimer’s disease, much research effort is being put into prevention of abnormal amyloid deposition, although no commercially available drug has been developed yet (Berg et al., 1998, Walsh & Selkoe, 2004). Phase two trials for a vaccine against beta-amyloid caused unacceptable levels of meningoencephalitis reactions (Morgan & Gitter, 2004) and until the non-pathological role(s) of beta-amyloid are elucidated, it may not be possible to develop a low-risk, effective vaccine.
Another way of preventing amyloid production is to prevent the abnormal cleavage of the apolipoprotein precursor protein. This would require inhibition of either of the secretase enzymes responsible for the cleavage. Preliminary animal studies show that secretase inhibition does indeed reduce the amount of beta-amyloid produced (Lovestone & McLoughlin, 2002). Additionally, the reduction of high levels of zinc, copper and iron in the brain could reduce amyloid deposition, as these metals appear to catalyse the formation of amyloid plaques (Zhang, 2005).

2.1.6.3 Relationship between cholinergic and amyloid neuropathology

A recent review of interactions between beta-amyloid deposits and central cholinergic neurons points out that excess beta-amyloid negatively affects many aspects of acetyl choline synthesis and release in vitro. The authors suggest that beta-amyloid production and normal cholinergic function may be in a dynamic balance, and that excess beta-amyloid can initiate the decline in cholinergic function, promoting the possibility of developing Alzheimer’s disease (Kar et al., 2004).

Other researchers have also come to similar conclusions, and suggest that targeting beta-amyloid is a preferable therapeutic strategy to targeting cholinergic symptoms (Walsh & Selkoe, 2004, Lemstra et al., 2003). However, until there is a safe and effective treatment for prevention of amyloid plaque formation, cholinesterase inhibitors are likely to prevail as the Alzheimer’s drug of choice.

2.1.6.4 Glutamate receptor inhibition

In the last few years, research has investigated the possibility of modifying the impaired glutamatergic system of people with more advanced Alzheimer’s disease. However, as under-stimulation of glutamate receptors leads to prevention of memory formation (Molinuero et al., 2005, Areosa Sastre et al., 2005), care must be taken in drug design not to block glutamate expression or reception too vigorously.

Antagonism of the NMDA glutamate receptor by low-affinity antagonists like memantine provides small but beneficial improvements in cognitive function and neuropsychiatric parameters in people with moderate to severe dementia (Koch et al., 2004). Memantine
C$_{12}$H$_{21}$N is a smaller molecule than the acetylcholinesterase inhibitors, readily soluble in water.

A systematic review of memantine for dementia revealed the following results for one six month study of cognitive improvement in mild to moderate dementia using the ADAS-cog (Areosa Sastre et al., 2005). The mean difference (sd) for memantine (n = 195) was 0.80 (7.81), placebo (n = 198) was -1.10 (7.87). Compared to the ADAS-cog results for the cholinesterase inhibitors (see Section 2.1.6.1.1), memantine provides slightly less benefit for cognitive function but the results were nevertheless statistically significant.

Occurrence of side effects was similar to placebo, the most prevalent being:

- hallucinations 1 in 50
- headache 1 in 59
- confusion 1 in 77
- tiredness 1 in 100
- dizziness 1 in 143 (Merz, 2004).

Less common side effects that were more frequent than placebo were anxiety, hypertonia, vomiting, cystitis and increased libido.

### 2.1.6.5 Other drug targets

Although the emphases in Alzheimer’s disease research are on the cholinergic, amyloid and glutamatergic pathological changes, several other drug targets are available. These include the reduction of high levels of oxidative damage and inflammation found in Alzheimer brains (Christen, 2000), and vitamin supplementation to boost low levels of vitamin B12 and folate (Meins et al., 2000, Malouf et al., 2003). In addition, other neurotransmitter systems impaired by Alzheimer’s disease may present opportunities for other drug targets (Gsell et al., 2004).

#### 2.1.6.5.1 Inflammation

Inflammation caused by extra-cellular amyloid plaques is also thought to contribute to neuron deterioration, and treatment with anti-inflammatory agents is another possible pharmacotherapeutic avenue (Combs et al., 2000, Koistinaho & Koistinaho, 2005). Inflammation and production of reactive oxygen species that cause oxidative stress often go
hand in hand (Christov et al., 2004). A recent review suggests that certain non-steroidal anti-inflammatory drugs (NSAIDs) like ibuprofen may also have anti-amyloidogenic properties, and that NSAID use in dementia is a worthy avenue of research (Townsend & Pratico, 2005).

2.1.6.5.2 Oxidative status

As Alzheimer’s disease is primarily an age-related disease, cumulative oxidation of brain lipids, proteins and DNA may contribute to the disease. Production of abnormal beta-amyloid is also known to generate oxidative stress in the brains of people with Alzheimer’s disease (Christen, 2000). A review of the neuroprotective effects of antioxidants (vitamin E, vitamin C, *Gingko biloba* extracts, red wine compounds and curcumin from turmeric) concluded that these antioxidants reduce neuronal oxidative damage *in vitro*, but that more research is needed to establish the value of antioxidants in improving cognitive function in humans, and whether antioxidants can ameliorate the disease (Esposito et al., 2002, Pham & Plakogiannis, 2005).

2.1.6.5.3 Vitamin B deficiencies

B12 and folate deficiencies are common in people with Alzheimer’s disease, and in reviews of the literature, supplementation appeared to improve cognitive ability and reduce behavioural dementia symptoms (Nilsson et al., 2001, Meins et al., 2000). The changes were statistically significant, but small. However, the correlation between low B12 levels and cognitive impairment indicates that participants’ baseline B vitamin levels need to be taken into account in clinical trials of products intended to improve cognitive function.

2.1.6.5.4 Other impaired neurotransmitter systems

The use of anti-psychotics, sedatives and anti-depressants is common in management of dementia (Richards & Hendrie, 1999). However, some sedatives and other drug types such as anti-arrhythmic drugs and anti-hypertensives (both very commonly prescribed in the elderly) have anti-cholinergic effects that can cause cognitive deficits very similar to ‘mild cognitive impairment’ (Ancelin et al., 2006).

From another point of view, anxiolytic drugs and anti-depressants may well enhance cognitive function by restoring balance to the impaired neurotransmitter systems. However, further research is needed, for example, to investigate which serotonin reuptake inhibitors are
effective in redressing the serotonin imbalances in Alzheimer’s disease (Bains et al., 2002, Lanctôt et al., 2001).

2.1.6.6 Combination therapy

Given the many pathological factors in Alzheimer’s disease, it may be appropriate to use multiple treatments, or drugs that affect multiple targets. This is being discussed in the literature as ‘combination therapy’ (Schmitt et al., 2004, Xiong & Doraishwamy, 2005), although there is a need for more well-designed trials to compare the benefits of single treatments with combinations.

2.1.6.6.1 Cholinesterase inhibition and glutamate antagonism

Results of trials combining different cholinesterase inhibitors and memantine, a glutamate antagonist have been reviewed (Standridge, 2004). Cholinesterase inhibitors used alone stabilized cognitive impairment and delayed deterioration by at least 3 months, and also improved performance of activities of daily living, reduced behavioural disturbances, decreased caregiver stress, and delayed onset of the first dementia-related nursing home placement. Trials combining cholinesterase inhibitors and memantine yielded greater benefit than cholinesterase inhibitors alone in all these areas.

2.1.6.6.2 Cholinesterase inhibition and monoamine oxidase inhibition

Zhang (2005) summarises research that has investigated compounds with multi-potent effects applicable in Alzheimer’s disease. Among these are compounds that have combined cholinesterase inhibition and irreversible monoamine oxidase-B inhibition. These compounds have been designed to address the lowered levels of acetylcholine and the monoamine neurotransmitters, serotonin and dopamine. Selegiline, a compound used for Parkinson’s disease is simultaneously a dopamine agonist and a monoamine oxidase-B inhibitor, but care must be taken when using it in Alzheimer’s disease, as it is also a pro-psychotic agent (Gsell et al., 2004).
2.1.6.6.3 Natural products with multipotent effects

Curcumin, a biphenolic molecule found in the Indian spice, turmeric (*Curcuma longa*) has three actions that have potential for Alzheimer's disease prevention and remediation:

- Blocking of beta-amyloid aggregation;
- Chelation of Cu$^{2+}$ ions;
- Anti-oxidant properties (scavenger for reactive oxygen species) (Zhang, 2005).

Other natural products that have anti-oxidant and metal chelating properties, and also prevent amyloid aggregation are flavonoids with catechol rings (phenols with two hydroxyl groups) (Zhang, 2005). Unfortunately however, catechol is a precursor in quinone synthesis, thereby presenting potential risks of neurotoxicity. Xanthones with only one catechol ring and a more stable formation are less likely to form quinones, and in addition have cholinesterase and monoamine oxidase inhibiting properties (Brühlmann et al., 2004). At the time of writing, no human clinical trials of these novel multi-potent drugs have been completed.

2.2 Dementia - management

As the available pharmacotherapeutic strategies for dementia do not provide a cure for the condition, management of dementia symptoms and alleviation of the accompanying distress and discomfort is important. Management strategies include home care assistance, admission to residential care, use of mood- and behaviour-modifying pharmacotherapy and complementary therapies, and socio-behavioural interventions.

In Australia, there is a trend for people with dementia to stay at home for as long as possible, and enter into residential care as a last resort (Aged Care Ministerial Reference Group, 2000). There is also a trend towards increasing numbers of people in low-care hostel accommodation, and decreased lengths of stay in the more costly high care facilities due to people entering at more advanced stages of dementia.

2.2.1 Care-giver burden

According to Jorm (2001), the Australian Bureau of Statistics estimated that in 1998, 201,000 people listed their role as primary carers for people over the age of 65. There is quite an
emphasis in the literature about the burden of care-giving experienced by people who care for elderly relatives. The burden of care-giving (care-giver burden) includes negative impacts on mental health such as increased depression, and illnesses relating to chronic stress (Savage & Bailey, 2004, Vitaliano et al., 2003).

2.2.1.1 Entry into residential care

As the costs of residential care for people with dementia are high, it is necessary to investigate when and why family care-givers of people with dementia decide to put them into residential care. In a review of medical management of advanced dementia, predictors of nursing home placement were summarised from the literature (Tariot, 2003). They included patient variables such as:

- behavioural problems;
- incontinence;
- aggressive behaviour, and
- severity of dementia.

Care-giver variables included:

- enjoyment of care-giver role;
- perception of burden;
- health status;
- symptoms of depression;
- income, and
- use of services.

In a survey of 109 caregivers, incontinence and social withdrawal were the key reasons for institutionalisation (Thomas et al., 2004). Maintenance of independence was considered to be one of the main goals of care-giving, with loss of independence leading to earlier institutionalisation (Livingston et al., 2004). As independence requires intact cognitive function, maintenance of cognitive function by whatever means possible is an appropriate management strategy.

Within aged care institutions, nurses and care-staff find dementia-related behaviours the most difficult behaviours to deal with. A survey of 253 staff from 12 Australian residential aged
care facilities (Brodaty et al., 2003a) found that the five attributes of dementia that staff found most difficult to cope with were:

- being aggressive or hostile;
- having little control over their difficult behaviour;
- being stubborn or resistive;
- being deliberately difficult, and
- being unpredictable.

Strategies to manage these aspects of dementia are discussed below.

### 2.2.2 Management strategies in residential care facilities

Dementia management strategies in residential care facilities need to be flexible enough to deal with varying symptoms of different stages of dementia. People with dementia are tending to enter institutions later and with more severe behavioural and psychological symptoms, so the care-burden due to dementia in residential facilities is increasing (Aged Care Ministerial Reference Group, 2000). A seven-tiered management model for the different stages of dementia is summarised in Table 2.4 (Brodaty et al., 2003b).

#### Table 2.4 Seven-tiered management model arranged by severity of behavioural and psychological symptoms of dementia (BPSD), proposed by Brodaty et al. (2003).

<table>
<thead>
<tr>
<th>Tier</th>
<th>Description and action to be taken</th>
<th>% of people with dementia in tier</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. No dementia</td>
<td>Normal aged-related memory impairment and slowing of cognitive functions. Life-style interventions to delay or prevent dementia onset: increased dietary protective agents like antioxidant, increased cognitive stimulation and physical exercise</td>
<td>0</td>
</tr>
<tr>
<td>2. Dementia with no BPSD</td>
<td>Memory and cognitive impairments severe enough to be assessed as dementia. Continue with approaches used in Tier 1, plus: introduce cholinesterase inhibitors to delay worsening of cognitive abilities introduce care-givers to training programs and support groups</td>
<td>40%</td>
</tr>
<tr>
<td>3. Dementia with mild BPSD</td>
<td>Mood changes and behaviours include: apathy, mild depression, repetitive questioning, following people around closely. Continue with Tier 1 &amp; 2 approaches, plus: train care-givers in behavioural management techniques such as using distracting pleasant activities and problem solving add relevant pharmacotherapy such as anti-depressants and/or anti-psychotics for aggressive, agitated or psychotic behaviours.</td>
<td>30%</td>
</tr>
<tr>
<td>4. Dementia with moderate BPSD</td>
<td>Mood changes and behaviours include: major depression, verbal aggression, non-dangerous physical aggression, psychosis, sexual disinhibition, and wandering. Continue with behavioural management and pharmacotherapy, but in consultation with a specialist custom-design a management program.</td>
<td>20%</td>
</tr>
<tr>
<td>Tier</td>
<td>Description and action to be taken</td>
<td>% of people with dementia in tier</td>
</tr>
<tr>
<td>------</td>
<td>-----------------------------------</td>
<td>----------------------------------</td>
</tr>
<tr>
<td>5. Dementia with severe BPSD</td>
<td>Mood changes and behaviours include: severe depression, severe agitation, psychosis, screaming. Care in a dementia-specific unit or case-managed by team of dementia specialists (psychiatrist, geriatrician, psychologist and social worker if still in community)</td>
<td>9-10%</td>
</tr>
<tr>
<td>6. Dementia with very severe BPSD</td>
<td>Mood changes and behaviours include: dangerous physical aggression, severe depression, suicidal tendencies, people would be admitted as psychiatric inpatients if they did not have dementia. As this is most likely due to an acute condition like delirium, temporary placement in a specialist psycho-geriatric unit is advised.</td>
<td>0-1%</td>
</tr>
</tbody>
</table>

BPSD = Behavioural and psychological symptoms of dementia.

Management approaches from Tiers 1-5 appropriate for use in residential aged care facilities include both pharmacological and non-pharmacological management approaches, and are discussed below.

### 2.2.2.1 Pharmacological management of dementia in residential aged care

Dementia drug targets and available pharmacological agents have been discussed in Section 2.1.3.4. Most people who have been institutionalised for dementia are in the moderate to severe stages of Alzheimer’s disease or other dementias (Brodaty et al., 2003b), so appropriate pharmacotherapy includes drugs to manage behavioural and psychological symptoms. Previously, anti-cholinesterase agents were thought to be of use only in the early stages of the disease, but recent research suggests that they also have impact on behavioural and psychological symptoms of more advanced Alzheimer’s disease (Wynn & Cummings, 2003, Ibach & Haen, 2004). As the current research project aims to investigate the effects of essential oils on cognitive function in people with dementia in aged care facilities, this recent research validates the decision to focus on the anti-cholinesterase properties of essential oils.

### 2.2.2.2 Non-pharmacological management of dementia in residential aged care

In residential aged care facilities, pharmacotherapy is often complemented by non-pharmacological or psychosocial approaches such as:

- changes to the physical environment to reduce confusion and prevent absconding;
- behavioural therapy;
- music therapy;
• bright light therapy, and
• aromatherapy.

Cochrane reviews of the literature concluded that most of these psychosocial interventions are not supported by a rigorous scientific evidence base (Opie et al., 1999, Forbes et al., 2004, et al., 2003). This suggests a need for further research, such as the current research project, to evaluate the efficacy of aromatherapy for improvement of cognitive function in dementia.

2.3 Conclusions: Dementia – impact and management

The background information on the impact and management of dementia described above confirmed the author’s opinion that additional research on non-pharmacological management strategies, such as aromatherapy, was worthwhile. The next chapter describes the practice of aromatherapy, reviews the existing literature on potential therapeutic effects of essential oils relevant to dementia, and critically reviews the studies of aromatherapy in aged care.
3 Background: Aromatherapy

Aromatherapy is a complementary therapy that aims to promote well-being via a holistic treatment using inhalation of essential oils, massage with essential oils, listening and life-style counselling (Battaglia, 1995, Price & Price, 1995, Worwood, 1990). Practitioners also treat specific ailments such as respiratory infections, fungal infections and arthritic pain. Although the word ‘aromatherapy’ implies that olfactory perception of aroma is the therapeutic principle, the current practice of aromatherapy could be better described as ‘essential oil therapy’, as essential oils are applied to the skin and inhaled with the intention of systemic absorption as well as olfactory stimulation.

The pleasantness of the olfactory experience is thought to be important, and authors often refer to the ‘art of blending’ which borders on perfumery (Battaglia, 1995). In Europe, essential oils are used as part of ‘phytotherapy’ (herbal medicine), and can be given orally or via suppository, depending on the diagnosis (Pênoël & Franchome, 1990). Oral and rectal administration of essential oils is not approved by the International Federation of Aromatherapists (Australia) (IFA, 2005), and neither method is used in main-stream Australian aromatherapy practice.

The sections on materials and application methods are largely taken from aromatherapy books, whereas the effects of aroma, and pharmacokinetic and pharmacodynamic effects of essential oils were derived from searches of the Medline database (most recent search was from 1966 to 23 October 2005) using ‘aroma therapy’, ‘essential oil’ and the relevant search term(s).

3.1.1 Materials used in aromatherapy

The ‘aroma’ referred to in the name ‘aromatherapy’ is a property of the plant extracts used in the therapy, known as essential oils. Essential oils are aromatic mixtures of terpenoid and phenylpropanoid compounds extracted from fresh or dried plant material by steam distillation, or by solvent extraction. The molecular weight of compounds found in essential oils ranges from 137-260, and most are lipophilic and minimally soluble in water. Terpenoid molecules may have one or more functional groups (alcohols, aldehydes, ketones, esters, methyl ethers,
cyclic ethers, and lactone or coumarin structures) and have various pharmacological properties and odours (Bowles, 2003).

The small size and lipophilic nature of most essential oil molecules means they are likely to cross the blood-brain barrier with ease (Guyton & Hall, 2000, Zetola et al., 2002). However, to date the concentrations of essential oil compounds reaching the brain in humans after aromatherapy treatments, are unknown.

3.1.2 Application Methods

Application methods have not been assessed for their comparative efficacy, although a common-sense approach prevails. For example, if you are treating a respiratory infection, the most logical application method is an inhalation of vaporised oil. Other application methods include massage, topical application, baths and ambient fragrancing, depending on the required outcome. Each application method uses a different dosage as outlined below, but these dosages have not been rigorously assessed for safety or efficacy.

3.1.2.1 Massage

Essential oils diluted in vegetable oil up to 5% (v/v) strength can be applied to the body via massage. Usually 3-5 essential oils comprise the blend to give the required range of effects, as determined from the consultation (Battaglia, 1995). A full body massage takes up to 1.5 hours which is inappropriate in most aged care facilities, whereas massage of a small area, such as hands or feet is often preferred. Massage of hands and feet is also less invasive and more likely to be well accepted by residents. In general aromatherapy practice, massage is often given for stress-related conditions, abdominal cramps, arthritic pain and musculo-skeletal tension (Battaglia, 1995).

3.1.2.2 Topical application

Two to five drops of a single essential oil can be applied directly to a small area to treat acute topical inflammation or pain, caused by insect bites, burns, boils and infectious conditions like tinea. Wound-healing gels have been formulated with up to 12% essential oil (w/v) for application to wounds (Kerr, 2002), and many over-the-counter arthritis joint-relief creams contain up to 22% essential oils and terpenoid compounds (e.g. Tiger Balm, Haw Par Health
Care Pty Ltd, Singapore). Topical applications are usually repeated as required until the condition subsides.

### 3.1.2.3 Inhalation

Two to three drops of an essential oil can be added to a bowl of hot water, and the resulting vapours inhaled. This method is of benefit when treating respiratory diseases, as the vapours and the warm steam are thus applied directly to the infected membranes.

### 3.1.2.4 Ambient fragrancing

Aromatherapy is often applied by indirect means, using a vaporiser or diffuser to introduce three to five drops of essential oils into the air of a room. This method gives the air a pleasant aroma and is used for mood modification, insomnia, and disinfection of the atmosphere. It is similar to inhalation in that it affects the respiratory system, but provides a smaller dose over a longer period of time.

### 3.1.2.5 Baths

Aromatherapy for stress-related conditions and infections especially of feet, groin and genital areas or haemorrhoids can effectively be applied in a warm bath. Three to five drops of essential oil (either single or a blend) are usually added to an emulsifier, or directly to a foot-bath or bath. For infections of the genital area, a shallower bath or ‘sitz’ bath can be used (Battaglia, 1995).

### 3.1.3 Effects of aroma

The therapeutic effects of aroma are difficult to determine objectively, although the link between perceived pleasantness of odours and improved mood has been established (Jellinek, 1997). Recent research showed aroma of Rosemary (Rosemarinus officinals) essential oil appeared to improve alertness and task-processing speeds in healthy normal adults (Moss et al., 2003, Diego et al., 1998), while Lavender (Lavandula angustifolia) oil reduced those parameters. Results for other odour components were not significantly different for controls (Ilmberger et al., 2001).
Unfortunately, none of the authors address whether the improvements would have clinical relevance in people with memory impairment. However, as people with dementia frequently suffer olfactory losses (Henkin, 2000), it is doubtful whether the mood- or cognitive-improving benefits of odour are likely to have significant impact in dementia.

3.1.4 Pharmacokinetics of essential oils in humans

While aromatherapy dosage and administration methods have not been rigorously assessed, essential oils are thought to be rapidly absorbed through the skin, lungs and gastrointestinal surfaces, and fairly rapidly eliminated. One English review on pharmacokinetics of essential oils was found. The review had 43 references, of which 13 were in German (Kohlert et al., 2000). Pharmacokinetic data in humans and animal models were summarised for the essential oil components alpha-pinene, beta-pinene, 1,8-cineole, menthol, camphor, borneol, isobornyl acetate, limonene and anethole. However, methodological details were not adequately reported in some cases, making it difficult to assess the results.

Dermal absorption of 2g of ointment (containing the essential oils components alpha-pinene, camphor, beta-pinene and limonene) was rapid in healthy, young volunteers (n=12), reaching peak plasma concentrations of up to 9.6 ng/mL in less than 10 minutes, although limonene had a second peak at 60 minutes (Schuster et al., 1986). Absorption rates could be faster in frail aged people as the epidermis tends to get thinner with age.

A similar biphasic pattern was observed for inhaled 1,8-cineole (4mL in a closed breathing circuit for 20 minutes) in 4 healthy human volunteers (Jäger et al., 1996). The initial half-life representing ‘distribution into the tissue’ ranged from 3.3-7.0 minutes, with plasma concentrations at the initial peak ranging from 459-1135 ng/mL. The second phase half-life representing the ‘elimination half-life’ ranged from 31.1 minutes to 2 hours 41.6 minutes.

This wide variation in the elimination half-life suggests variation in metabolic activity in individuals, and may represent distribution of the lipophilic 1,8-cineole into fat deposits as suggested by Jäger et al. (1996). Another paper suggested a third phase of at least 9 hours 15 minutes for the elimination of inhaled alpha-pinene (Falk et al., 1990), so perhaps pharmacokinetics of essential oils should be measured for more than one hour to observe a more accurate elimination pattern.
However, even if most essential oil compounds have tri-phasic elimination half-lives of 10 hours, at current aromatherapy dosages, it is unlikely that there would be a dose-loading effect with single daily applications. Further research would be necessary to confirm whether dose-loading is possible with essential oil components.

3.1.5 Pharmacodynamics of essential oils with possible relevance to dementia

In an aromatherapy treatment, essential oils are usually selected for the client’s presenting condition(s) based on their accepted therapeutic properties. Some of this anecdotal and historical knowledge is supported by scientific research on essential oils and their components. Research with possible application to the treatment of dementia is described below, with references being sourced from a text on essential oil chemistry (Bowles, 2003) or retrieved electronically from Medline databases starting from 1966 to date of the search (23 October 2005)

3.1.5.1 Antioxidant effects

Elevated oxidative stress accompanies amyloid deposition and neurofibrillary tangles in Alzheimer’s disease (Christen, 2000, Christov et al., 2004). Several oils exhibit in vitro antioxidant effects, for example, Thyme (*Thymus vulgaris*) and Oregano (*Origanum vulgare*) which have high percentages of the anti-oxidant phenolic compound thymol (Teissedre & Waterhouse, 2000). The most appropriate antioxidant assays would be measures of oxidation in human brain lipids or proteins, but the research is mostly confined to human plasma proteins.

Three papers in English have investigated the effects of some essential oils on lipoproteins or other molecules from the body (Teissedre & Waterhouse, 2000, Mantle et al., 1998, Grassmann et al., 2001). Grassmann et al. (2001) incubated human plasma with Lemon (*Citrus limonum*) oil and one of its components, gamma-terpinene, and found both to be protective against the oxidation of the low-density lipoproteins.

Teissedre and Waterhouse (2000) used a similar technique and found that 2µM concentrations of oils with high percentages of phenolic compounds (eugenol, thymol, and carvacrol) inhibited oxidation of low-density lipoproteins. However, Mantle et al. (1998) investigated the protective effects of essential oils on human brain alanyl aminopeptidase against oxidative
damage by reactive oxygen species, and found that none of the typically antioxidant essential oils had a protective effect, although the oils were active in two other *in vitro* antioxidant tests. Further research is required to elucidate the *in vivo* antioxidant benefits of essential oils in other brain lipids and proteins.

### 3.1.5.2 Anti-inflammatory effects

Inflammation is another by-product of amyloid plaques in brains of people diagnosed with Alzheimer’s disease (Koistinaho & Koistinaho, 2005, Combs et al., 2000). There are several reports on the *in vitro* anti-inflammatory effects of essential oils and their components. The anti-inflammatory mechanisms include:

- inhibition of cyclo-oxygenase by Pokeweed (*Minthostachys verticillata*), containing pulegone and methone (Gonzalez et al., 2003);
- inhibition of 5-lipoxygenase by a product containing menthol, alpha-pinene, 1,8-cineole (Beuscher et al., 1998) and extracts containing Frankincense (*Boswellia serrata*) triterpenoids (Safayhi et al., 2000);
- inhibition of nuclear factor-kappa-B by sesquiterpenoid lactones (found in *Arnica sp.*) and other plants (Lyß et al., 1998, Koch et al., 2001);
- inhibition of cytokine synthesis by 1,8-cineole (found in Eucalyptus oil) (Juergens et al., 2003);
- reduction of leukotriene production by the sesquiterpenoid chamazeulene found in Chamomile (*Matricaria recutita*) oil (Safayhi et al., 1994), and
- reduction of interleukin production (Juergens et al., 2004).

While anti-inflammatory effects of essential oils may be of benefit in Alzheimer’s disease, further clinical trials are necessary to determine clinically relevant outcomes, as the mechanisms by which non-steroidal anti-inflammatory compounds provide neuroprotection in ageing are not yet fully elucidated (Townsend & Pratico, 2005).

### 3.1.5.3 Calcium channel blockade and glutamate inhibition

Abnormally high levels of synaptic glutamate cause excitotoxicity in Alzheimer’s disease and vascular dementia (Kornbuber & Wiltfang, 1998). Blockade of calcium ion influx in Alzheimer’s disease and vascular dementia prevents excitotoxicity as calcium influx initiates the cascade towards glutamate release (Stewart, 2002). The dementia drug memantine is a
low-affinity antagonist for the NMDA-glutamate receptor, blocking excessive levels of glutamate production, while allowing normal glutamate function for memory formation (Lipton, 2004).

The compounds cadinol, anethole, estragole and eugenol caused temporary anti-spasmodic effects in smooth muscle (animal trials) (Albuquerque et al., 1995, Zygmunt et al., 1993), as did Peppermint (*Mentha x piperita*) oil and menthol in human and animal models (Pittler & Ernst, 1998). The anti-spasmodic mechanism in each case was purported to be the prevention of calcium ion flow into sensory neurons. Whether these essential oil compounds can function as calcium-channel modifiers in brain tissue is yet to be elucidated.

The monoterpenol linalool has been shown to interfere with $K^+$-mediated glutamate release (Brum et al., 2001) which may account for its sedative properties (Buchbauer et al., 1991) and indicate possible usefulness in dementia therapy. Relatively high doses of linalool (1mg/mL) prevented glutamate-induced neurotoxicity in rat pups (Buyukokuroglu et al., 2003).

**3.1.5.4 Cholinesterase inhibition**

As cholinesterase inhibition is the main pharmacotherapeutic approach in mild and moderate Alzheimer’s disease, and offers both cognitive and behavioural improvements (Standridge, 2004, Ibach & Haen, 2004), cholinesterase inhibition by essential oils was explored in greater detail. The next sections summarise the available cholinesterase inhibition assays, and present a review of the literature on cholinesterase inhibition by essential oils.

An electronic search of the following databases: Medline, AlliedMed and ISI Current Contents, was carried out on 30 March 2004 and again on 23 October 2005. Both searches searched all the available entries from either 1966 (Medline) or the start date of the databases if later than that, to the date of the search. The second search was intended to identify any research that had been reported during the experimental phase of the project that could have an impact on the conclusions drawn from the experimental phase. ‘Acetylcholinesterase’, ‘cholinesterase’ ‘essential oil’, ‘volatile oil’ were the primary search-terms combined with the following terms: ‘dementia’, ‘Alzheimer’, and ‘review’. No published reviews were found at the time of searching. References were managed on EndNote (v7) reference management software.
3.1.5.4.1 Available cholinesterase inhibition assays

The most-utilised assay is a spectrophotometric method known as the Ellman method (Ellman et al., 1961). The Ellman method depends on the hydrolysis of acetylthiocholine iodide (ATCI) by acetylcholinesterase (AChE). The hydrolysed thiocholine iodide moiety reacts with the colour-change reagent 5,5'-dithiobis-(2-nitrobenzoic acid) (DTNB) to create a yellow anion with maximum absorbance at 412nm. Different enzyme and substrate concentrations yield different rates of enzyme activity, with substrate concentrations of greater than $10^{-3}$M inhibiting enzyme activity.

The method was initially developed for a single well manual spectrophotometer (Ellman et al., 1961). Subsequent modifications have adapted it for use with 96-well plate automatic spectrophotometers (Nostrandt et al., 1993, Savelev et al., 2003). The Ellman method has also been adapted for thin layer chromatography (Rhee et al., 2001), while another variant uses 1-naphthyl acetate instead of acetylthiocholine iodide as the enzyme substrate, and Fast Blue B salt instead of DTNB to visualise the inhibition zones (Marston et al., 2002). However, the usefulness of the thin layer chromatography method for testing AChE inhibition by essential oils is likely to be limited by the low solubility of essential oil compounds in the aqueous enzyme solution.

3.1.5.4.2 In vitro inhibition of human brain enzyme

Selection of essential oils by one of the early papers reporting anti-cholinesterase activity of essential oils was done by screening plants with known insecticidal activity. The hypothesis was that insecticidal activity could be due to cholinesterase inhibition by the essential oils in the plants (Perry et al., 1996). Homogenates of normal young human autopsied brain tissue were used as a source of cholinesterase, and measured using the Ellman method (though not specified whether it was in a single cell spectrophotometer, or in a 96-well plate machine). Essential oils were diluted in 80% ethanol, pre-incubated with samples for 30 minutes, and then 0.33mM ATCI and 10mM DTNB were added and the samples incubated at 37° C for 5-30 minutes. The reaction was terminated with physostigmine, and then optical density was measured at 412nm after a further 30 minutes incubation at 4° C.

There are several limitations to the report which make it difficult to compare the results with other reports. Final cell concentrations of reagents were not specified, nor was there report of
enzyme activity rates. Other reports use standardised enzyme units as a measure of enzyme activity. Furthermore, the step of adding physostigmine and then incubating for 30 minutes at 4° C was not explained, and is not included in the original Ellman method. However, the results are worth noting, as they are the first indications that essential oils may have therapeutic benefit as cholinesterase inhibitors in human brain tissue.

Melissa (*Melissa officinalis*), Lime (*Citrus latifolia*), Lavender Sage (*Salvia lavandulaefolia*) and Sage (*Salvia officinalis*) oils significantly inhibited human brain enzyme at 0.1µL/mL (inhibition >60% of the control). Clary Sage (*Salvia sclarea*), Rosemary (*Rosmarinus officinalis*), Hyssop (*Hyssopus officinalis*) and Angelica (*Angelica archangelica*) oils also showed some inhibition at 0.1µL/mL, but <25% of the control.

### 3.1.5.4.3 Inhibition of human erythrocyte enzyme

Another paper (NS Perry et al., 2000) reported the inhibitory effects of Lavender Sage (*Salvia lavandulaefolia*) oil and its major constituents on human erythrocyte enzyme (1U hydrolyses 1µmole of acetylcholine/min at 37° C). It is widely held that erythrocyte acetylcholinesterase has similar activity to brain acetylcholinesterase (Al-Jafari & Kamal, 1996), although no papers have reported comparisons of brain and erythrocyte enzyme with essential oils under the same experimental conditions.

Perry et al. (2000) reported that essential oil constituents were serially diluted in 96% ethanol to yield dilution series ranging from 0.09-9.4mM, and essential oil from 0.01-0.5µL/mL. Constituents and oil were pre-incubated with 50µL of 0.39U/mL enzyme (final well concentration 0.009U/mL) in 2.0mL of buffer (not 3.0mL as in the Ellman method) for 30 minutes at 4° C, and then run in a spectrophotometer with 20µL each (also different than the Ellman method) of 10mM DTNB and substrate (at varying concentrations 0.03-0.5mM) for 6 minutes at 25° C. Absorption was measured at 412nm over the 6 minute period, but it was not clear whether the percentage inhibition was calculated from the final value or from the maximum slope of the Absorbance/time graph.

Dose-dependent inhibition in order of potency was observed for Lavender Sage (*S. lavandulaefolia*) oil > alpha-pinene > 1,8-cineole > camphor. The IC$_{50}$ values for these materials were approximately 0.17mM, 0.63mM, 0.67mM and >10mM respectively. Beta-
pinene, borneol and limonene did not display dose-dependent inhibition of the human erythrocyte enzyme.

3.1.5.4.4 Inhibition of bovine erythrocyte enzyme

There are four reports on the inhibition of bovine erythrocyte enzyme by essential oil compounds (Miyazawa & Yamafuji, 2005, Miyazawa et al., 1998, Miyazawa et al., 1997, Savelev et al., 2003, NS Perry et al., 2000). As human erythrocyte enzyme is expensive, bovine enzyme is often used for preliminary investigations, and it appears to have become the accepted standard.

The papers by Miyazawa et al. all use a variation of the Ellman method, with 0.5mL of 0.04U/mL of enzyme (final well concentration of 0.0065U/mL) added to 2.4mL of buffer, 100µL of DTNB and 50µL of sample (mint oils and terpenoid constituents), pre-incubated at 25° C for 5 minutes, and then 40µL of substrate added to the cell and incubated for 20 minutes at 25° C. All assays contained final concentrations of 5% ethanol, including controls. From the volumes used, it appears that they used a single cell spectrophotometer.

The departure from the Ellman method concentrations is not explained, but the authors do mention the need to measure non-enzymic hydrolysis and subtract this from the results prior to calculation of the IC$_{50}$ values, which was not mentioned in either of the papers by Perry et al. The results by Miyazawa et al. do not have error estimates, and it is not stated whether the values are calculated from maximum slopes or from the 20 minute end-point readings. The four most potent compounds in order of potency were viridiflorol, elemol, (+)-3-carene and 1,8-cineole, with 50% inhibitory concentration (IC$_{50}$) values of 0.11mM, 0.15mM, 0.20mM and 0.26mM (final well concentration).

The paper by Savelev et al. (2003) uses yet another variant of the Ellman method, the microplate assay developed by Nostrandt et al. (1993). Here the final well concentration of bovine erythrocyte enzyme was 0.008U/mL (10-fold less than the Ellman method), added to samples (Lavender Sage (S. lavandulaefolia) oil and terpenoid compounds) diluted in 86% ethanol (final well concentration of 2% ethanol did not inhibit the enzyme), buffer and DTNB (final well concentration 0.3mM), and incubated for 30 minutes at 30° C. Then substrate was added (final well concentration 0.5mM) and the absorbance measured at 412nm for a period of six minutes at 30° C. Non-enzymic hydrolysis which contributes significantly to measurements
taken for longer than 5 minutes (Ellman et al., 1961) was accounted for automatically by the spectrophotometer software, presumably by the use of blank subtraction.

The results are reported in terms of % inhibition of controls calculated from dose-response curves, although it is not clear whether the data used were maximum slopes or 6 minute end-point data. The most active samples in order of potency were Lavender Sage (*S. lavandulaefolia*) oil, 1,8-cineole, alpha-pinene and beta-pinene (0.38mM, 0.66mM and 1.47mM).

3.1.5.4.5 Inhibition of electric eel enzyme

One paper reported results for Tea Tree (*Melaleuca alternifolia*) oil and its major compounds in electric eel acetylcholinesterase (Mills et al., 2004). Final well enzyme concentration was 0.0035 U/mL, about 3 times lower than that used by Savelev et al. and it was carried out in large wells (3.1mL total volume), so presumably single cell spectrophotometer. The very low enzyme concentration most likely accounts for the reported sensitivity of the enzyme to 1,8-cineole (IC$_{50}$ was 0.04mM, ten-fold less than other researchers found using bovine and human enzyme).

Terpinen-4-ol (the major component of Tea tree oil) on the other hand was found to be a very weak inhibitor of the enzyme (IC$_{50}$ = 10.3 mM). Several samples of tea tree oil with varying amounts of 1,8-cineole and terpinen-4-ol had IC$_{50}$ values ranging from 0.26mM to 0.61mM, although curiously the two most potent samples also had the lowest levels of 1,8-cineole, suggesting the presence of other inhibiting compounds, and perhaps synergistic interactions.

One paper also used electric eel enzyme, but did not use the Ellman method (Gracza, 1985). They report a cascade of IC$_{50}$ values: 1,8-cineole < carvacrol < (+)-carvone < fenchone in the 0.1-10 mM range, and thymol did not reach 50% inhibition. This is a similar order and level of potency to that observed by Miyazawa et al. (1998).

3.1.5.4.6 Comparison of results from different reports

Comparison of results between papers is problematic, as assay methods vary considerably, and often no error estimates are reported. However, IC$_{50}$ values for 1,8-cineole in both human and bovine enzyme were within the same order of magnitude 0.26-0.67mM. Also, where the
same compounds have been assayed by different researchers, the general order of potency is the same. For example, the order of potency is 1,8-cineole > alpha-pinene > beta-pinene > camphor.

3.1.5.4.7 Review of Lavender Sage oil AChE inhibition

A review on the use of Lavender Sage (Salvia lavandulaefolia) for acetylcholinesterase inhibition (Perry et al., 2003) summarises three in vitro studies of Lavender Sage oil on acetylcholinesterase enzymes from different sources using different incubation temperatures and time frames. The differences between the Lavender Sage results are similar to the differences found for 1,8-cineole and alpha-pinene.

Comparison was made with the IC\textsubscript{50} values of other known cholinesterase inhibitors reported by Perry et al. (1996): tacrine, galantamine and physostigmine (2.0x10\textsuperscript{-8}M, 5.0x10\textsuperscript{-7}M and 4.5x10\textsuperscript{-8}M respectively). The approximate molar IC\textsubscript{50} values for Lavender Sage (being a largely monoterpenoid oil, average mw for monoterpenoids =154) range from 0.2-0.4 mM. However, the known cholinesterase inhibitors were not tested in Perry et al. (2000) or by Savelev et al. (2003), and may have provided a range of values. Known inhibitors are often used as positive controls in experimental designs, to allow comparison between studies from different research groups. It is generally acknowledged to be good laboratory practice to include such reference materials if possible.

3.1.5.4.8 Synergy and antagonism between compounds

Miyazawa et al. (1998), Mills (2004) and Perry et al. (2000) note that essential oils are often more potent inhibitors than any of their individual components. A careful study of pair-wise interactions between the major compounds of Lavender Sage oil showed both synergistic and antagonistic interactions (Savelev et al., 2003). Mixtures of 1,8-cineole and (+)-alpha-pinene, or 1,8-cineole and caryophyllene oxide showed synergy, whereas mixtures of 1,8-cineole and (+)-camphor showed less activity than either compound on its own. The authors suggest that essential oils with high 1,8-cineole and low camphor are likely to be more effective cholinesterase inhibitors than ones with a higher cineole:camphor ratio.
In one study of essential oil cholinesterase inhibition in a live rat model, two groups of 6 rats were gavaged with either 20µL or 50µL of Lavender Sage (*Salvia lavandulaefolia*) essential oil diluted 1:2 in a sunflower oil base for 5 days prior to being sacrificed and their brain tissue examined for cholinesterase function (Perry et al., 2002). A control group of 6 rats received plain sunflower oil. The results showed that 20µL doses inhibited striatal acetylcholinesterase, and that 50µL doses inhibited both striatal and hippocampal acetylcholinesterase (though less than 50% of controls). Neither dosage inhibited cortical acetylcholinesterase activity.

While these results support the hypothesis that Lavender Sage oil may have significant anticholinesterase activity in living organisms, the final essential oil concentration in the blood-stream and brain tissue after treatment was not quantified, so it is not possible to compare these results with the *in vitro* \( \text{IC}_{50} \) reports. Measuring concentrations of compounds and/or metabolites present in the brain tissue would have contributed to understanding the pharmacokinetics of the oil in a living organism, and where possible should be carried out in future research.

Future research should use an essential oil compound with known inhibitory activity as a standard as well as galantamine or one of the other known inhibitors. Additionally, until a comparison of essential oil inhibition of acetylcholinesterase from different animal sources under the same assay conditions has been completed, the usefulness of absolute \( \text{IC}_{50} \) values remains questionable.

### Essential oils as possible multi-potent agents for dementia

Certain essential oils and their components inhibit cholinesterase, reduce inflammation and oxidation and modify glutamate transmission *in vitro*. The compound 1,8-cineole is an example of one compound with two activities, acetylcholinesterase inhibition and anti-inflammatory action via cytokine inhibition.

Further research could investigate the potential for multiple *in vitro* activities of essential oil components and also establish clinically relevant effects of whole essential oils containing components with known *in vitro* efficacy for various Alzheimer’s disease drug targets.
3.1.6  A review of aromatherapy in dementia care

3.1.6.1  Search strategy

An electronic search of the following databases: Medline, AlliedMed, Ovid Nursing Full-text, CINAHL, PsychInfo and ISI Current Contents, was carried out on 30 March 2004 and again on 23 October 2005. Both searches searched all the available entries from either 1966 (Medline) or the start date of the databases if later than that, to the date of the search. The second search was intended to identify any research that had been reported during the experimental phase of the project that could have an impact on the conclusions drawn from the experimental phase. ‘Aromatherapy’, ‘aroma therapy’ and ‘essential oil’ were the primary search-terms combined with the following terms: dementia, residential care, Alzheimer, cholinesterase, and review. Published reviews were obtained first, and the reference lists searched for further studies. References were managed on EndNote (v7) reference management software.

3.1.6.2  Study selection criteria

Studies on aromatherapy for dementia in aged care facilities or hospitals were included if they also reported:

- intention of the study;
- participant characteristics;
- trial design;
- dosage and application methods;
- quantitative primary outcome measures.

Studies of aromatherapy for conditions other than dementia were not included, nor were studies of aromatherapy for dementia in the community.

3.1.6.2.1  Types of intervention

All doses, application methods, frequencies of dosage and types of essential oils used were included, as well as all trial designs.
3.1.6.2.2 *Types of outcome measures*

All types of outcome were considered, including cognitive function, behavioural and psychological symptoms of dementia, and sleep enhancement, but only studies reporting quantitative outcome measures were selected.

3.1.6.3 *Review methods*

3.1.6.3.1 *Study selection*

Using the above search strategy nine reviews were selected, only one of which was specifically about aromatherapy and dementia (Thorgrimsen et al., 2003). From these reviews and the database searches, twenty-two papers were initially selected. Seventeen of these studies met the selection criteria.

Fourteen studies reported the effects of aromatherapy *via* inhalation, massage or bathing. Agitation, difficult behaviours, resistive behaviours and sleeplessness were the main symptoms treated.

Three studies reported the effects of ingestion of essential oils or ethanolic extracts containing essential oil components on cognitive function. While ingestion of essential oils is not normally advised by Australian aromatherapy practitioners, the findings are discussed as they support the hypothesis that essential oils may improve cognitive function in dementia.

3.1.6.3.2 *Quality assessment*

Clinical trial reviews commonly assess the methodological quality of clinical trials using checklists (Jadad et al., 1996, CONSORT, 1996). According to these two references, clinical trials are considered to be good quality if they report at least:

- Details of randomisation;
- Appropriate controls;
- Details of blinding of assessors and participants;
- Description of trial design that would allow replication;
- Appropriate outcome measures;
- Adequate result data (means, error estimates, range, and appropriate statistics);
• Details of participation (e.g. number of drop-outs);
• Discussion of study limitations.

Only one of the selected aromatherapy and dementia studies was adequately randomised, controlled and blinded (Ballard et al., 2002). However, the intention of reviewing the literature was to gather ideas for an aromatherapy clinical trial protocol, not primarily to assess the rigour of studies of aromatherapy for dementia. Therefore all of the available studies were reviewed, not just the one that complies with the criteria for good quality clinical trials.

3.1.6.3.3 Aromatherapy for agitation

Five studies investigated aromatherapy for management of agitation in dementia (Ballard et al., 2002, Brooker et al., 1997, Holmes et al., 2002, Smallwood et al., 2001, Snow et al., 2004).

3.1.6.3.3.1 Ballard et al.

The study by Ballard et al. (2002) was the only study meeting sufficient quality criteria to be accepted as a randomised, controlled trial in a Cochrane Database systematic review (Thorgrimsen et al., 2003). Seventy-two elderly people from 8 different aged care facilities in England started the trial (mean age (sd) = 78.5 (8.1) years), with 71 completing (one died of unrelated causes). Facilities were matched for resident similarity and then randomly allocated into either the treatment group or placebo group to avoid cross-contamination and unblinding of staff as to the purpose of treatment. Participants were enrolled if they had moderate to severe agitation on the Neuropsychiatric Inventory (NPI) (Cummings, 1997) and a Clinical Dementia Rating of 3, indicating severe dementia (Hughes et al., 1982).

After baseline measurements, the four week treatment period involved application of 3 pumps of lotion, twice daily, applied to hands, arms and face of participants, containing either sunflower oil (placebo) or sunflower oil with 10% (w/w) of Lemon Balm (*Melissa officinalis*) essential oil. This delivered 200 mg of essential oil per day to the treatment group. Measures were taken at the end of every week.
The primary outcome measure was change in total score on the Cohen-Mansfield Agitation Index (CMAI) (Cohen-Mansfield et al., 1990) between baseline and the end of trial. Secondary outcome measures included sub-scores on the CMAI, NPI and changes in quality of life parameters.

Both treatment and placebo groups reduced agitation significantly on the Cohen-Mansfield Agitation Index (CMAI), the treatment group by 33%, and the placebo group by 11%. The authors suggest that a change on the CMAI of >30% would be clinically significant, and found that 21/36 people in the active group achieved this, whereas only 5/36 in the placebo group achieved this over the 4 weeks of the trial. Quality of life data suggested that aromatherapy significantly reduced social withdrawal and increased participation in social activities. There were no adverse events like skin irritation or other signs of toxicity, though one person experienced 2 days of diarrhoea during the study. Linear regression analysis did not show any significant difference between facilities, but there was a trend towards the treatment group having a higher agitation score at baseline compared to the placebo group which could have biased the reported decrease in agitation in the treatment group.

While this study shows a positive outcome for aromatherapy, there are some concerns about the protocol which preclude using it as a direct model for a future clinical trial. The first is the use of 10% concentration of essential oils. Although Lemon Balm (*Melissa officinalis*) oil may be safe to use in this way, other oils may be more skin-irritating and more toxic. Safety-data are not currently available for use of 10% concentrations of most oils, so it is probably safer to use accepted aromatherapy dosage levels (1-5%) in a vulnerable aged population (Tisserand & Balacs, 1995). Furthermore, a 10% dilution of some essential oils would be a strong smell, and could cause difficulties in staff and residents with respiratory allergies.

The allocation to treatment by facility, rather than by individual may bias the results due to systemic differences between facilities. As people with dementia often have reduced olfactory ability (Mesholam et al., 1998), it is unlikely to cause unblinding of participants. Blinding of the assessor is more feasible than blinding of staff, although staff blinding could be achieved by use of two different smelling blends or different dosages of the same blend as well as a placebo.

Four other trials also focussed on reducing agitation, but used Lavender (*Lavandula angustifolia*) oil, not Lemon Balm oil either with massage or by inhalation (Holmes et al.,
These trials did not report drop-outs, and were not blinded or the randomisation method was not described. Sample sizes were also small: The study by Brooker had n=4; that of Holmes n=15; that of Smallwood n=21; and that of Snow n=7. All four studies were on people with severe dementia from one institution, whether long-stay hospital ward or aged care facility.

3.1.6.3.2  Brooker et al.

The four severe dementia cases evaluated by Brooker et al. (1997) were selected on the basis that they appeared to be able to register odours and that they demonstrated some behaviour considered by staff to be ‘distressed’. The aim of the study was to evaluate an existing aromatherapy program to see if it helped residents feel more relaxed and less distressed. Age ranged from 74-91 years old, 3 had Alzheimer-type dementia, and one had frontal lobe dementia. No formal assessment of dementia severity was reported.

A single case study design was used with three 30 minute treatments and one ‘no treatment’ option. Each person received 8-12 sessions of each treatment (and the ‘no treatment’ option) over a 3 month period. All sessions involved sitting in a small sitting room on the ward with either: fan-diffused Lavender oil (dosage not specified), hand and arm massage with unscented body oil, combination of diffused Lavender and massage, or just sitting in the room at the same time as someone else was receiving massage. The sessions were carried out in the early afternoon when it was usual for participants to be sitting in that room.

The primary outcome measure was change on a 6 point individualised ‘distressed behaviour’ scale developed by staff for each person prior to the trial. Observations were made every minute for the hour following the treatment, and there was no ‘wearing off’ observed when the data was summarised into 15 minute intervals.

The best-fit slope of median scores in the no-treatment and treatment conditions was compared. Statistically significant results were only observed in one person, where both aroma alone and massage alone decreased behaviours more than the ‘no treatment’ condition, but aroma and massage combined did not reach significance.

The small number of observations may have veiled the true significance of the results, due to fluctuations in dementia behaviours (Walker et al., 1999). The study could have been
improved by choosing only the treatment they were evaluating (presumably fan diffusion of Lavender oil) and comparing that with the ‘no treatment’ condition, thereby doubling the number of observations for each condition. The study had other flaws, such as not reporting the dosage of Lavender oil used, but the single case study design could be helpful in controlling for individual responses to aromas and touch.

3.1.6.3.3 Holmes et al.

The study by Holmes et al. (2002) aimed to evaluate the efficacy of Lavender oil in treating agitation in 15 patients (mean age (sd) = 79.0 (6.3) years) with severe dementia (according to the WHO ICD-10 criteria) living in a long-stay unit for patients with behavioural problems. Probable type of dementia was determined using established scales like the NINCDS-ADRDA (McKhann et al., 1984), and included Alzheimer’s disease, vascular dementia, dementia with Lewy bodies and fronto-temporal dementia. All participants had daily scores > 3 on the 16 point Pittsburgh Agitation Scale (PAS) (Rosen et al., 1984) in one week prior to the study.

Participants were exposed to 10 alternate days (over 2 weeks) of diffused lavender oil (treatment) or diffused water (placebo) in the communal area of the unit, between 4 and 6 pm. Over the second hour, an observer wearing a nose-peg, and blinded to the study design scored the individual behaviour of each participant using the PAS. Median scores for treatment and placebo conditions of each individual were compared with the Wilcoxon Signed Ranks test.

Comparison of the group median PAS scores in treatment (median = 3, range 1-7) and placebo conditions (median = 4, range 3-7), showed a statistically significant difference (p = 0.016). Nine patients’ behaviour improved by 1-3 points, five stayed the same, and one worsened by 2 points.

The clinical significance of score changes on the PAS was not discussed, although the reported improvement was called ‘modest’ by the authors. The number of subjects was too small to provide a normally distributed set of data, and no means or error estimates were reported. This made it difficult to gauge the variability within individual responses to the treatment and placebo conditions. Another consideration is that the effects of aromatherapy may have carried over into the placebo days if the essential oils were absorbed into the
bloodstream. A repeat of this study with the placebo and lavender conditions given one week at a time may yield better results.

3.1.6.3.4 Smallwood et al.

Smallwood et al. (2001) report a randomized controlled trial of lavender oil for control of motor behaviour disturbances in 21 hospital inpatients (mean age (sd) = 66.8 (11.5) years) with severe dementia (as rated by a psychiatrist). Dementia type was not specified, and one person was excluded during the study due to deteriorating health.

Participants were randomly allocated to one of three treatment groups: aromatherapy massage with unspecified quantity of lavender oil; massage with plain oil; and conversation with therapist and diffused lavender oil (unspecified quantity). Treatments were twice weekly, but duration of each treatment was not reported, or body part massaged, nor total length of the trial. Daily baseline records were taken for two weeks prior to treatment, for 4 periods of 15 minutes each between 10 am and 4 pm. Scores were rated from video records by raters blinded to condition.

The primary outcome measure was changes in frequency of disturbed motor behaviours (Bowie & Mountain, 1993) after treatment at different times of day.

One way analysis of variance (ANOVA) showed no significant differences between frequency of behaviours between baseline and post-treatment for any of the treatment groups. Analysis of the data for effects due to time of day, there was one significant result. During the 3 pm to 4 pm time period, the ‘aromatherapy massage’ group displayed significantly fewer behaviours than the ‘conversation and aromatherapy’ group (p=0.05).

Baseline means and standard deviations are reported for the three treatment groups with no accounting for time of day. Post-treatment changes in behaviour are graphed by treatment group (without error-bars) with time of day as the variable, but post-treatment scores are not reported separately. It would have been easier to interpret the data displayed with time of day on the x-axis, and the variables being the treatment groups.

As the authors point out, a simpler design using only one treatment and a placebo group may have shown better results. In addition, the baseline data could have suggested which time of
day motor behaviours occurred with greatest frequency, thus reducing complexity of the analysis.

### 3.1.6.3.3.5 Snow et al.

Snow et al. (2004) compared the effects of Lavender, Thyme and unscented grape seed oil (a vegetable oil) on the agitated behaviour of seven residents of a nursing home with severe dementia (Severe Impairment Rating Scale scores of less than 18) and a score of >24 on the Cohen-Mansfield Agitation Index (CMAI) indicating moderate agitation.

Baseline measures were taken for 4 weeks, and then a 10 week treatment period followed, with a 2 week washout. During the treatment period, participants had a small fabric sachet pinned to the collar on which 2 drops of the treatment was placed, 3 times a day. The treatments were given in two week blocks, in the following order: Lavender, Thyme, grape seed, Thyme, Lavender. The expected outcome was that agitation would only be reduced during the lavender condition. Measures were collected by interviewing staff every other day with a modified CMAI (i.e. 7 data points for each treatment). Assessment of olfactory function was carried out prior to commencement of the study.

The primary outcome measure was comparison of best-fit slopes of median data for the grape seed oil treatment (placebo) with the slopes of the two essential oil treatments on a case-by-case basis. This method is justified by the authors as appropriate for small sample sizes, and less influenced by large variability.

No overall significant differences were noted between any of the conditions, even in those participants with some evidence of olfactory ability. One individual showed a significant increase in agitation in the first Lavender treatment phase following baseline observations (p<0.05).

This study uses the same data analysis method as that of Brooker et al. (1997). Both studies would have benefited from simpler study design, and longer treatment periods, as the behaviour fluctuations for several of the subjects was so wide from day to day that more observations would have provided greater robustness.
3.1.6.3.6 Conclusions about aromatherapy for agitation in severe dementia

One of the main findings of these studies is that mere ability to smell the odours of the essential oils does not appear to influence agitated behaviours of severely demented people. Perhaps odours perceived as comforting would have more effect. The research on olfactory hedonics suggests that odours perceived as pleasant generate positive mood states in normal healthy adults (Jellinek, 1997).

Furthermore, inclusion of massage appears to be important in achieving a positive outcome for people with severe dementia, and it may be necessary to increase the dosages used in aromatherapy to achieve clinically significant outcomes. Twice-daily massage treatments for at least one month appear to be the most effective as in Ballard et al. (2002).

3.1.6.3.4 Aromatherapy for resistive behaviours

Five papers report effects of aromatherapy on resistive behaviours (Beshara & Giddings, 2002, Bowles et al., 2002, Gray & Clair, 2002, Mitchell, 1993, Walker et al., 1994). None of these were considered randomised controlled trials by Thorgrimsen et al. (2003), but they have some helpful methodological considerations.

3.1.6.3.4.1 Beshara et al.

Beshara et al. (2002) investigated the effects of diffused essential oil blends on 10 people with varying types of dementia (Alzheimer’s, vascular, and Korsakov’s) living in an aged care facility. Individuals were selected due to their extreme behaviour severity, and a list of ‘problem’ behaviours particular to each individual were identified using the ‘Minimum Data Set’, an American assessment tool used to monitor psychotropic drug administration (reference not cited in paper).

A list of individual behaviours was identified for each participant. Baseline frequencies for each behaviour were recorded for one month prior to treatment. Treatment blends containing 5 or more essential oils were diffused into participants’ rooms or communal areas for a six month period. No dosages or frequencies of treatment were reported. None of the blends contained Lavender (*Lavandula angustifolia*), Lemon Balm (*Melissa officinalis*), or other oils thought to be sedative, for example, German Chamomile (*Matricaria recutita*) or Sweet
Marjoram (*Origanum marjorana*). Oils present in the blends included Tangerine (*Citrus deliciosa*), Orange (*Citrus sinensis*), Ylang-ylang (*Cananga odorata*), Patchouli (*Pogostemon cablin*) and Blue Tansy (*Tanacetum annum*). The primary outcome measure was changes in monthly frequency of behaviours at 1 month, 3 months and 6 months.

Behaviours in six out of ten people reduced by 50% or more compared to baseline by the sixth month. However, only two individuals had baseline behaviours occurring at frequencies of greater than 90 per month (i.e. greater than 3 per day). Three individuals experienced an increase in certain behaviours by the sixth month, but not by more than 7 occurrences per month. No statistical analyses were carried out.

The conclusions made by the authors that aromatherapy can be used to control these behaviours are unfortunately flawed by the study design. Firstly, no record of concurrent neuroleptic medication is provided. Without this, it is impossible to determine whether the effects are due to aromatherapy or not. Secondly, if the incidence of behaviours is initially as low as 2 per month, it is difficult to say whether a reduction to 0 per month is significant.

### 3.1.6.3.4.2 Bowles et al.

The study by Bowles et al. (2002) investigated the effects of aromatherapy massage on the resistive behaviours of 56 people with moderate to severe dementia in a nursing home, both during nursing care interventions and at other times. Complete data sets for thirty-six participants were reported. Reasons for drop-outs were not provided.

Two groups matched by sex, mobility, type and severity of dementia received an unscented placebo lotion for 2 weeks followed by two months of either the placebo lotion or essential oil lotion in a staggered cross-over design, with a final 2 week washout with placebo lotion. The 3.5% essential oil lotion contained Lavender (*Lavandula angustifolia*), Sweet Marjoram (*Origanum marjorana*), Patchouli (*Pogostemon cablin*) and Vetiver (*Vetivera zizanoides*) oils and 5mL of lotion was applied with a 5 minute massage 5 times a day during the treatment month, delivering a daily dose of approximately 0.9mL of essential oil blend. Each participant’s behaviours were scored every shift by nursing staff responsible for them on that shift for the entire study.
The primary outcome measure was change in behaviour scores compared between baseline, placebo and treatment, using a ‘frequency x severity’ nurses’ observation scale derived from the Cohen-Mansfield Agitation Index (CMAI) and Neuropsychiatric Inventory (NPI). Two factors were drawn out, summarising behaviours caused by resistance to nursing care and disturbed behaviours at other times. Cognitive function was measured using the Mini-Mental State Examination (MMSE) before and after essential oil treatment.

The two treatment groups responded differently to the essential oil treatment. Mean scores for Group A significantly increased in resistance to nursing care (p=0.003), and decreased in behaviours at other times (p=0.04). Mean scores for Group B were not significantly different for resistance to nursing care, but were significant between the baseline and treatment phase (p=0.003). Seven out of eight people who could complete the MMSE test significantly improved on their baseline scores by an average of 3.1 (sd >5.9) after essential oil treatment (p=0.015).

Unfortunately the group behaviour scores were significantly different at baseline, and there were no inter or intra-rater reliability measures, which weaken the reliability of the scoring. However, this weakness may have been overcome by the number of scores for each participant.

While 5 applications daily appears to make a significant difference to resistive behaviours, it is unlikely that most nursing homes will have the commitment or staff resources to carry out such a regime for all residents with dementia. It is interesting that behaviours due to resistance to nursing care were more frequent than behaviours at other times for both groups.

3.1.6.3.4.3 Gray & Clair

Gray & Clair (2002) reported effects of vaporised essential oils on resistive behaviours in 13 people with dementia in two aged care facilities. Lavender (Lavandula angustifolia), Tea Tree (Melaleuca alternifolia) and Sweet Orange (Citrus sinensis) oils were put onto cotton balls by inverting the essential oil bottle for not more than 20 seconds and the cotton balls were taped to the clothing of participants well before they received their morning medications, as participants were resistant to receiving medication. Each person received each of the treatments 4 times (a total of 16 treatments including control cotton balls with no odour) and the medication attempts were videotaped and rated by two design-’blind’ raters.
The primary outcome was changes in frequency of resistant behaviours and duration of medication attempt for each participant. Each 10 second segment of the video-tape in which behaviours occurred was scored as a ‘plus’, and the total number of plusses per medication attempt was used as the resistive behaviour score. Total time of medication attempt was also recorded, and these sets of data were compared by one-way ANOVA. Unfortunately, variations between facilities were not taken into account during the analyses, nor were the ranges given with the means, making comparisons less robust.

No statistically significant difference was found between treatments or the placebo, though the Sweet Orange treatment displayed a possible trend towards reducing behaviours. This is not surprising on two accounts, namely the reduction in olfactory function in people with dementia, and the attempt to use aromatherapy like a ‘knock-out’ drug to block the resistive behaviours. As the authors commented, the time-frame of observation of behaviours was possibly too short, as there were changes to behaviours noted by staff after the medication attempts were completed. The dosage method was also very imprecise and could have biased results.

### 3.1.6.3.4.4 Mitchell

Mitchell (1993) investigated the effects of aromatherapy on 3 functional and 3 behavioural criteria for 12 people (age range 64-91) with dementia in a residential respite unit. Severity of dementia was not indicated.

Participants were randomly assigned to two groups. Baseline measures were established for one week, then participants received either the treatment or control oils for two weeks, followed by one week washout, then cross-over of treatment/control for two weeks. The treatment consisted of a composite aromatherapy regime including application of Lavender and Melissa oils in a wash (6 drops daily) and as a cologne respectively (about 1 drop), followed by nightly inhalation of Lavender oil (3 drops on pillow). Grape seed oil was applied similarly during the control condition.

The primary outcome measure was comparisons between baseline, treatment and control conditions on a 3 point nurses’ satisfaction scale (Very poor, Poor, and Satisfactory) for each of the six criteria:
• communication;
• independence;
• feeding and toileting;
• resistance;
• day-time wandering;
• night restlessness and night-time agitation.

Group A mean scores appeared to improve in functional criteria, and remain improved during the following control condition. Behaviours appeared to get worse during treatment, but return towards baseline during washout and placebo. Group B mean behaviour scores appeared to improve in behaviours during treatment (this was after the placebo treatment), but little change was noticed in the functional criteria. The two groups were quite different in their initial functional scores, but similar in behaviour scores. The author suggested that these findings appear to contradict the expected sedative effects of Lavender and Melissa oils.

However, statistical significance of results was not reported, only mean scores without error estimates or ranges. This makes it difficult to assess whether the changes observed could be used to make such a statement. The graphical representation of the data did not make clear which bars represented washout or baseline scores, and had no error bars.

3.1.6.3.4.5 Walker et al.

Walker et al. (1994) investigated the effects of lavender oil for reduction of aggressive and resistive behaviours, wandering, agitation and sleep patterns in 5 female residents of a dementia unit.

Using an A-B-A-B design, baseline behaviours were recorded hourly for 7 days for each participant followed by one week of aromatherapy treatment with continued hourly observations. During the third week participants received no treatment, and during the fourth week treatment was resumed. The treatment consisted of Lavender (Lavandula angustifolia) oil added in various concentrations to bath oil, body wash and fragrant water, and Lavender oil added to pot-pourri (2 drops a day), curtains (2 drops a day) and pillows (1 drop at night) in each person’s room. Frequency of dermal application was not reported, and no estimate of daily dosage made.
The primary outcome measure was reduction in observed behaviours between the start and end of the trial.

The authors reported a significant improvement in negative behaviours by the end of the trial and improved participant cooperation in activities of daily living, but failed to present any numerical data to support their observations. Details about response differences for the five participants were not reported. Staff liked the use of aromatherapy, and reduction in the number of back injuries was noted.

A-B-A-B (A = treatment, B = washout) trial designs are useful in determining whether there are dose-dependent drug effects, but while the pharmacokinetics of aromatherapy treatments are unclear, the precise length of each trial period remains guess-work.

3.1.6.3.4.6 Conclusions about aromatherapy for resistive behaviours

Two out of the five studies did not report sufficient information to be useful in determining whether aromatherapy is useful for resistive behaviours (Beshara et al. 2002, Walker et al. 1994). Aromatherapy massage may be helpful for reducing spontaneously-occurring resistive behaviours, but it appears as though resistance during nursing care is not amenable to change by the aromatherapy methods used in these studies.

3.1.6.3.5 Aromatherapy for sleeplessness

There were three studies on aromatherapy for sleeplessness (Connell et al., 2001, Hardy, 1993, Wolfe & Herzberg, 1996).

3.1.6.3.5.1 Connell et al.

A two week trial of aromatherapy to improve sleep in 58 elderly hospital patients resulted in a statistically significant improvement in number of hours spent asleep by the end of the trial (Connell et al., 2001).

The patients came from the dementia assessment unit, psychiatric units, and elderly continuing- and acute-care wards. Twenty-two out of fifty-eight had some diagnosis of
dementia, though the severity was not noted. Two patients per ward were tested at a time to fit in with available staff time, meaning that the trial was run in a staggered design. 43 subjects completed the trial, due to withdrawals (8), death (5) and administrative error (2).

Half-hourly baseline observations were recorded daily for one week between the hours of 9:00pm-7:00am by night staff. During the treatment week, two drops of Roman Chamomile (Anthemis nobilis) oil were placed on the patient’s pillow and the half-hourly observations continued. Other illnesses or ailments likely to interfere with sleep e.g. pain, and medications were recorded.

The primary outcome measure was a change in the mean numbers of hours sleep per week. Three classifications were made on the half-hourly observation, whether ‘asleep’ or ‘awake and calm’, or ‘awake and restless’.

Subjects spent a statistically significant greater numbers of hours in ‘sleep’ (p=0.004) during the treatment week than the baseline week. The mean difference was an increase of 0.8 hours a night. They also had significantly fewer hours ‘awake and calm’ (p=0.02), and a trend towards fewer hours ‘awake and restless’ during the treatment week, suggesting increased quality of sleep also. Patients with dementia did not sleep significantly more during the treatment week though there was a trend towards increased sleep.

The authors suggest that the major flaw of this trial was that the 22 participants from the acute wards were entered into the trial immediately they entered hospital. Increased sleep during the second week of the trial could therefore be due to a decrease in distress due to increasing familiarity with their new surroundings, rather than due to the aromatherapy intervention. Participants who just had sleep observations taken for two weeks without aromatherapy would have provided an acceptable control group.

It also appears that people with dementia were less responsive to the aromatherapy, and again this may be due to the impaired olfactory ability. As the studies on resistive behaviours and agitation showed, massage application of aromatherapy may be more helpful for people with dementia.
3.1.6.3.5.2 Hardy

Hardy (1993) reported a case study of aromatherapy for improved sleep in 4 male residents (age range 67-88) of a nursing home. They were reported as having ‘various stages of dementia’, and three were on some form of sleeping medication. All four men were in the same dormitory of the facility. The aim of the study was to investigate whether there was a detrimental effect if Lavender (*Lavandula angustifolia*) oil was used in place of sleeping medications.

Twenty-four hour sleep patterns were observed every half-hour for two weeks with all participants on their normal medication. This was followed by two weeks without medication, and then a further two weeks with vaporised Lavender oil in their dormitory.

The primary outcome measure was changes in mean hours of sleep per 24-hour period. Mean sleep hours were also calculated for day and night periods.

The results were presented graphically, and for each person, a reduction in sleep hours during the no-treatment fortnight was followed by a subsequent increase in total sleep hours during the Lavender treatment weeks. The mean number of hours sleep during Lavender treatment was almost the same as with medication. No statistical tests were used to compare the case studies, but the results appear to be significant, with the mean changes being about 1.5 hours more sleep per night with Lavender or medication.

In three residents, day-time sleep length during Lavender treatment was reduced compared to the medication baseline, suggesting that their night-time sleep was more restful during the Lavender treatment.

While the number of cases is small, this trial design appeared to show that vaporised Lavender oil is at least as effective as night-time sleeping medication, but it did not specify the severity of dementia or the volume of lavender oil used. Nor did it have inter-rater reliability tests, although with a simple sleep scale, it probably is not necessary.
3.1.6.3.5.3 Wolfe and Herzberg

Wolfe and Herzberg (1996) investigated the effects of aromatherapy on sleep times of two people with dementia (1 multi-infarct, 1 Alzheimer’s type) and aggressive behaviours in a residential care unit (ages 76 and 88).

Night-time sleep and frequency of eight behaviour types were recorded at half hourly intervals, starting with two weeks of baseline measurements. During the subsequent treatment weeks, two drops of essential oil were placed on the sheets near the face of the participants. The essential oils were tested in the following pattern: Lavender (Lavandula angustifolia), Roman Chamomile (Anthemis nobilis), and a mixture of Lavender and Roman Chamomile. Each oil or blend was applied for one week in this order, and then the whole lot repeated.

The primary outcome measure was changes in mean number of hours sleep per week, with changes in behaviours as secondary measures.

One person experienced a mean increase of sleep hours of 5.3 per week, and fewer hours of being ‘awake and distressed’, particularly during the Roman Chamomile treatments. The other person’s results were compromised by changes to their night-time medication, but the authors suggest that the essential oils appeared to be almost as effective as the night-time medication once the Roman Chamomile started again in the two final weeks of the trial. Although this is only a case study, it is interesting to note that Roman Chamomile was also used by Connell et al., (2001) with similar results.

3.1.6.3.5.4 Conclusions about aromatherapy for sleeplessness

Inhaled aromatherapy appears to increase sleep times for people with dementia. However, the mean increases range from about 0.8-1.5 hours per night, and no variance measures were reported, either for baseline or treatment periods, so it is difficult to say whether these increases were in fact significant. A larger scale trial with a control arm would assist in validation of these findings.
Ingestion of essential oils and ethanolic extracts for cognitive enhancement in dementia

Three papers report on the ingestion of essential oils and extracts containing essential oil compounds for cognitive enhancement of people with dementia in residential aged care facilities (Akhondzadeh et al., 2003a, Akhondzadeh et al., 2003b, Perry et al., 2003).

3.1.6.3.6.1 Akhondzadeh et al.

Akhondzadeh et al. (2003a and 2003b) investigated the effects of ingested ethanolic herbal extracts on 42 residents (age 65-80) from three aged care facilities in a randomised placebo-controlled trial. It appears as though the same facilities and possibly some of the same residents were involved in both trials, so the reports were reviewed in the light of this.

In both trials, the participants had histories of at least 6 months progressive cognitive decline, diagnosis of probably Alzheimer’s (NINCDS/ADRDA), >12 on ADAS-cog and <2 on the clinical dementia rating scale (CDR). These criteria identified people as having mild to moderate dementia.

Of the 42 people randomised to treatment in each trial, 35 completed the Melissa (Melissa officinalis) (also known as Lemon Balm) trial, (withdrawals: 6 from the placebo group, 1 from the treatment group) and 30 people completed the subsequent Sage (Salvia officinalis) trial (withdrawals: 5 from the placebo group, 4 from the treatment group).

Participants were randomised to receive 16 weeks of daily dosages of 60 drops of herbal ethanolic extract or 60 drops of an unspecified placebo. Each extract had standardised quantities of essential oil compounds. The method of administration was not described, but presumably the drops were given in capsules. Baseline measures were taken in the first week, and then assessed every two weeks, yielding nine repeated measures.

The primary outcome measures were changes in mean scores on the cognitive subscale of the Alzheimer’s Disease Assessment Scale (ADAS-cog) and Clinical Dementia Rating (CDR) ‘sum of boxes’. Each scale was administered and assessed by a neurologist. For both measures, a decrease between initial and final scores indicates an improvement in cognitive functioning (Burns et al., 1999).
Effects of both Melissa and Sage extracts on ADAS-cog scores were similar, yielding a statistically significant decrease in scores (i.e. an improvement) for the treatment group compared to the placebo group. As the Melissa paper was published first, perhaps the trial was also completed first. In any case, the initial mean ADAS-cog scores for the treatment and control group in the Melissa trial were about 27 at baseline, and not significantly different.

At end-point, the placebo group ADAS-cog scores had increased by 5.60 (sd 1.40), and the Melissa treatment group scores had changed by -6.40 (sd 1.66). The difference between treatments was highly significant, p<0.0001. Significant difference at p=0.05 was reached by week 4. ADAS-cog changes of 2-5 points are commonly observed for cholinesterase inhibitors (Doody et al., 2001), so it appears that Melissa extract is at least as effective as currently available cholinesterase inhibitors. Improvements on the ‘CDR-sum of boxes’ scores in the Melissa trial were also statistically significant for the treatment group, reaching significance at 8 weeks.

Initial mean ADAS-cog scores for the treatment and placebo group in the Sage trial were about 25.5 at baseline, and not significantly different. At end-point, the placebo group ADAS-cog scores had increased by 5.53 (sd 1.12), and the Melissa treatment group scores had changed by -6.60 (sd 1.63). The difference between treatment and placebo scores was highly significant, p<0.0001. Significant difference at p=0.05 was reached by week 4. Improvements on the ‘CDR-sum of boxes’ scores in the Sage trial were also statistically significant for the treatment group, reaching significance at 8 weeks. A possible flaw in the repeated measures design is that with only two weeks between measures, participants would likely experience a ‘learning effect’. The possibility of learning effects was not discussed by the authors.

Interestingly, Melissa extract significantly reduced agitation, compared to the placebo group (p=0.03). This may be an instance of improved cognition leading to a reduction in behavioural symptoms of dementia (Trinh et al., 2003). It also reinforces the observations of Ballard et al. (2002) reviewed above, who found aromatherapy massage with 10% Melissa oil effectively reduced agitation in dementia compared to placebo. Judging by the increase in the Melissa trial placebo group scores to above the baseline in the Sage placebo group, it is after all unlikely that the participants were the same in each group.

As the studies do not report a full chemical analysis for either extract, it is difficult to surmise whether the extracts are likely to be impacting the same neurochemical targets in the subjects.
Ethanolic extracts are likely to contain similar compounds to essential oils as the terpenoid constituents are largely ethanol-soluble, but there may be some water-soluble compounds in the extract with greater potency than any compound found in the essential oils. Sage essential oil usually contains alpha-pinene, 1,8-cineole and camphor with varying amounts of thujone. Melissa essential oil usually contains neral and geranial and varying amounts of coumarin derivatives. Miyazawa et al., (1997 and 1998) demonstrated that alpha-pinene and 1,8-cineole have in vitro inhibitory effects on acetylcholinesterase, but neral and geranial were not assessed by these authors.

3.1.6.3.6.2 Perry et al.

Perry et al. (2003) assessed the tolerability of different oral dosages of Lavender Sage (*Salvia lavandulaefolia*) oil in eleven people with dementia (age 76-95), nine in residential care, two still at home. Patients had MMSE scores between 10 and 26 and NPI scores of zero for items 3 and 9 (agitation and irritability).

The trial was a phase II open label trial, with no control, aiming to establish the potential efficacy and appropriate dosage range of Lavender Sage oil for subsequent clinical trials. In the first week of the trial, participants received one capsule a day containing 50µL of essential oil and 50µL of sunflower oil. Two capsules a day were received in the next two weeks, and three capsules a day in the final 3 weeks of the trial.

Primary outcome measures included blood samples and both physical and neurological examinations. The measures used in the neurological examinations were changes on the MMSE between baseline and the end of the trial, changes on the Cognitive Drug Research computerised battery of tests, and the NPI.

The mean MMSE had a slight increase (statistically not significant) between baseline and 6 weeks (+ 0.48, but with sd >3), and the vigilance task in the Cognitive Drug battery was significantly improved (p=0.014), although there were also more false alarms made. The NPI scores were significantly reduced by 3 points (sd >5) (p=0.024), indicating a reduction in behavioural and psychological symptoms of dementia. There was a mean increase in blood pressure (diastolic 15.09, systolic 6.19, p=0.049) due mainly to two patients with pre-existing hypertension whose blood pressure rose sharply during the highest dosage weeks.
These results suggest improved cholinergic function, and inhibition of blood cholinesterases was noted (though not reported as statistically significant). It would be interesting to compare ingestion of Lavender Sage oil with aromatherapy massage as in Ballard et al. (2002), although establishment of an equivalent dermal dose may prove problematic.

3.1.6.3.6.3 Conclusions about essential oils and plant extracts for cognitive enhancement

It appears as though ingestion of Melissa (*Melissa officinalis*) and Sage (*Salvia officinalis*) extracts and Lavender Sage (*Salvia lavandulaefolia*) essential oil significantly improve cognitive function in aged care residents with mild to moderate dementia, though a longer clinical trial of the essential oil is necessary. It is interesting to note that the participants in these trials probably had less severe dementia than those in the other aromatherapy studies reviewed above.

3.1.7 Conclusions about the clinical use of aromatherapy for dementia

The reviewed studies aimed to evaluate existing aromatherapy practices or assess the usefulness of aromatherapy as a replacement for existing pharmacotherapy. While the available research is not methodologically robust, aromatherapy appears to have possible beneficial effects on agitated behaviours and sleeplessness in dementia, and it would be worth carrying out further research in these areas as well.

Most of the studies investigated the effects of very small doses of inhaled essential oils over short periods in residents with severe dementia, and most showed equivocal results on the selected outcome measures. The one study that was adequately designed and also showed significant effects on agitation in severe dementia used higher doses of essential oils in a massage application and had a 4 week treatment phase (Ballard et al. 2002).

Effects of aromatherapy on cognitive function of people with dementia have not yet been adequately researched, although essential oils with anti-cholinesterase function appear to show promise as therapeutic agents for dementia.

From the reviewed literature, it is evident that the best methodology for aromatherapy use in dementia is yet to be identified, although dermal application or ingestion may be the most appropriate routes. Effective dosage regimens have not yet been determined, even for those
oils with a substantial anecdotal basis like Lavender (*Lavandula angustifolia*) and Roman Chamomile (*Anthemis nobilis*).

Although some of the case studies showed aromatherapy to be effective for some people, the studies were not carried out for long enough to establish whether these effects were part of the normal fluctuation of dementia, or effects due to treatment. Placebo controls, matched ‘no treatment’ groups and larger sample sizes would add to the experimental rigour of research.

Use of standard outcome measures is also important, to compare the usefulness of aromatherapy with other drugs. Where possible, both cognitive and behavioural outcome measures should be used, as essential oils may have multiple effects due to their many different compounds.
4 Background investigation: a survey of aromatherapy practices in aged care facilities in northern NSW, Australia

4.1 Introduction

The literature review of trials of aromatherapy for dementia revealed the use of different treatment regimes and essential oils for dementia. There is currently no published professional consensus among aromatherapists as to what constitutes an effective aromatherapy treatment regime for aged care residents with dementia.

The aim of this phase of the research project was to investigate the existing aromatherapy practices in the aged care facilities in the area local to Southern Cross University using a survey. The investigation was intended to establish a rationale for the subsequent clinical trial design based on a quantitative evaluation of existing practices, rather than on anecdote. The survey was also intended to introduce the author to the aged care facility staff who would be integral to the subsequent clinical trial.

The published peer-reviewed paper reporting on this project is reproduced in Appendix 1 (Bowles et al., 2005). Survey design rationale and key findings pertinent to the subsequent laboratory work and follow-up clinical trial are discussed. It should be noted that not all the survey responses are reported in the body of the thesis, as some related to background information not pertinent to the laboratory work or clinical trial.

4.2 Survey development

Typical survey development tasks are itemised in several survey methodology texts. The list by Fink (2003) was used as a guide, and contained the following steps:

1) Identify the survey’s objectives
2) Design the survey
3) Prepare the survey instrument
4) Pilot-test the instrument
5) Administer the survey
6) Organize the data
7) Analyse the data
8) Report the results

These steps were followed with more or less the detail listed by Fink (2003), and the survey development process is described below.

4.2.1 Identification of survey objectives

The objectives of the survey were developed in discussion with the supervisors. The objectives were to:

- Describe the variations in aromatherapy practices in aged care facilities in northern NSW.
- Explore staff perceptions about the efficacy of aromatherapy for dementia.
- Determine if any aromatherapy practices correlate significantly with perceptions of efficacy for dementia.
- Identify the essential oils most used in aged care, and which conditions are being treated with essential oils.
- Identify appropriate treatment regime elements for the subsequent clinical trial protocol.

4.2.2 Survey design

According to Fink (2003), survey design is concerned with defining the environment, time frame and participants that are to be surveyed. As this survey was intended to be a data collecting exercise for the subsequent laboratory work and clinical trial, it was decided to do a descriptive survey of aged care facilities that were within two or three hours drive from the university in the Northern Rivers area of NSW. It was decided, based on expediency, to contact all the facilities in the same geographical area, rather than randomly sample from all NSW facilities. Fink (2003) suggests that non-random sampling is appropriate in pilot situations that have not been previously surveyed, which was the case here. This reduces the generalisability of the results, but was considered appropriate, as the subsequent clinical trial was going to be conducted in the same facilities. The survey was therefore used to establish contact and rapport with staff members in facilities who would be called on to assist with the clinical trial.
4.2.3 Preparation of the survey instrument

Fink (2003) recommends the adaptation of previous survey instruments of similar design, so as to reduce the preparation time. However, the author thought it would save time to write her own list of questions to fit them to the aims of the survey, as no previous survey of aromatherapy in aged care facilities had been carried out, and she had specific data-gathering objectives likely to be unique to this survey.

The author then convened a meeting of the Northern Rivers Aged Care Aromatherapy Network (NRACAN) as a focus group to assist with further development of the questions and to fine tune the survey design. NRACAN is a monthly forum for nurses working with aromatherapy in aged care to share their ideas, receive support and solve problems within a peer-group setting. Six NRACAN members attended the meeting, representing aged care facilities in the local area that were likely to be part of the survey sample. The author asked the NRACAN members’ advice about the following issues:

- Who would be the best people to interview to get the most accurate picture of aromatherapy practices and its perceived efficacy?
- Which survey format would be most acceptable to these people?
- Are there issues that might affect people’s willingness to do the survey?
- Are the questions comprehensive enough in their scope and are there any items missing that should be included?

The advice was that each facility using aromatherapy as a therapeutic intervention would have a particular person who would be the aromatherapy planner in the facility, and that person would be the best person to interview about the aromatherapy practice questions. However, it was advised that the directors of care would be the best people to interview for the background questions about the facilities.

Everyone in the NRACAN meeting agreed that a mail survey would be unlikely to receive good responses because aged care nurses are already so busy with paperwork. Telephone surveys were also rejected, because phone calls are normally kept to a minimum in facilities. A face-to-face interview was thought to be the best format, because a set appointment time could be made, and the interview could take place in a quiet area with minimum disturbance. Face-to-face interviews also would allow the author to visit the facilities likely to participate in the subsequent clinical trial. The point was made that the survey should be as short as
possible, and that box-ticking and rating scale questions were easier to answer than open-ended questions.

Discussion during the meeting revealed that the practice of aromatherapy in aged care facilities within the local area is highly variable. The six group members present said that they designed residents’ aromatherapy care plans around management of physical, emotional and behavioural symptoms, regardless of a resident’s dementia status. They advised that questions should be asked about the use of aromatherapy for these symptoms, rather than for the dementia, as there was the supposition that residents with dementia in residential care are “too far gone” for their cognitive function to be improved by aromatherapy. During the meeting we brain-stormed the items for all the questions in the aromatherapy planner’s survey, including the list of essential oils likely to be used.

Using the comments from the NRACAN meeting, and subsequent phone discussions with a couple of the members, a two-part fact-finding survey questionnaire was created to record responses of directors of care and aromatherapy planners in face-to-face interviews.

4.2.3.1 Question design rationale

The question design was discussed with the research supervisors who had previous experience in question design for complementary therapies. A variety of question formats were used to elicit the required responses. Closed-ended questions with either a yes/no or tick-box list format were used to gain quantitative data about items that had been determined by the author. Closed-ended questions designed to gauge opinions had multiple visual analog rating scales. Open-ended questions were used to gain new information, and the responses were subjected to thematic analysis.

Closed-ended question types were chosen for the majority of the survey questions because it was a fact-finding survey, and respondents would have limited time to complete the surveys. According to de Vaus (1995), closed-ended questions are appropriate for questions that have concrete answers, or for collecting quantitative data about pre-determined items. They are also easier for respondents to answer quickly, and responses are more easily analysed than open-ended questions. Responses required either a tick against ‘yes’ or ‘no’, or the writing of a number in the space provided.
The closed-ended questions designed to gauge respondents’ opinions used rating scales to detect the range of opinion intensities (de Vaus, 1995). Furthermore, as opinions about a complex concept can be more closely investigated by asking respondents to rate several components of the concept using multiple rating scales (de Vaus, 1995), most of the opinion-gauging questions in the survey had multiple rating scale responses.

The scales used in the survey were diagrammatic or visual analog rating scales, which are straight horizontal lines with least and most intense responses labelling either end of the line. Respondents mark their response along the line according to where they feel their opinion fits. For example, if the question was “Which aspects of aromatherapy do you think are helpful for dementia?” the two descriptor phrases at either end of the scale (for example, for items such as ‘touch’ or ‘nurturing’) were “Not at all helpful” and “Extremely helpful”. If a respondent felt ‘touch’ was extremely helpful, they would make a mark closest to that end of the scale. The advantage of visual analog scales over Likert-type scales (with defined intensity points from 1 to 5) is that the gradings of opinion can be distinguished even further, with the score measured in millimetres from one end of the line. Visual analog scales were also preferred over a Likert-type scale by elders responding to a satisfaction survey (Castle and Engberg, 2004), and subsequently by nursing home administrators responding to a job-satisfaction survey (Castle, 2006).

Multiple scale responses for the same question, can be subjected to factor analysis, a statistical procedure yielding clusters of linked responses (factors), allowing a more in-depth understanding of people’s opinions about the complex issue (de Vaus, 1995). For example, the question in the survey asking which conditions people thought aromatherapy was effective for has several responses, some of which are dementia-related, others which are not. Factor analysis could reveal a cluster of conditions that aromatherapy is thought to be most effective for, thereby eliciting more information than a single scale response to the question “How effective is aromatherapy for improving cognitive function in dementia?”.

Open-ended questions are best used for those items in a survey where the researcher does not want to influence the responses (Fink, 2003). Open-ended questions were used to find out what were the main uses of the different essential oils and commercial blends, adverse reactions and what types of drugs were reduced by aromatherapy use. A disadvantage of open-ended questions is that they require subjective thematic analysis that can be more open to bias and error than closed-ended questions (de Vaus, 1995), but the information gained can
open up new perspectives not previously imagined by the researcher. Some questions in the survey were partially closed-ended, in that table headings guided the type of responses required, but the responses were open-ended. These responses also needed to be coded, but it was easier to create codes for the different responses than to create themes.

Part one of the survey (see Appendix 1), completed by the directors of care, had eleven questions (see Table 4.1). Questions 1-9 were intended to gather data about the characteristics of the facilities and staff that might affect aromatherapy practices. Questions 10 and 11 were asked to gauge whether aromatherapy is perceived to reduce pharmaceutical use, and for which conditions. As the data collected in this part of the survey was largely quantitative, closed-ended questions were considered appropriate (Nardi, 2003). Question 11 was open-ended as the number of pharmaceuticals used in aged care is extensive, and the author did not want to limit the responses.

Table 4.1 Questions asked in Directors of Care survey.

<table>
<thead>
<tr>
<th>Number</th>
<th>Question</th>
<th>Response type</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>How many staff members in your facility apart from the aromatherapist have aromatherapy qualifications</td>
<td>Ordinal</td>
</tr>
<tr>
<td>2</td>
<td>Please list the qualification details of staff members below:</td>
<td>Open-ended, space for six responses (qualification type, number of staff with qual., institution, length of course, year obtained)</td>
</tr>
<tr>
<td>3</td>
<td>Is the facility government owned, non-government, other?</td>
<td>Cardinal</td>
</tr>
<tr>
<td>4</td>
<td>Do you have a dementia-specific unit/ward?</td>
<td>Closed-ended, yes/no</td>
</tr>
<tr>
<td>5</td>
<td>How many residents do you currently have?</td>
<td>Ordinal</td>
</tr>
<tr>
<td>6</td>
<td>How many have a geriatrician’s diagnosis of dementia?</td>
<td>Ordinal</td>
</tr>
<tr>
<td>7</td>
<td>Who funds aromatherapy in this facility?</td>
<td>Closed ended, yes/no. Options: facility; residents/family; other</td>
</tr>
<tr>
<td>8</td>
<td>If funded by the facility, what is the average monthly cost of aromatherapy in this facility?</td>
<td>Ordinal</td>
</tr>
<tr>
<td>9</td>
<td>Who administers aromatherapy to residents in this facility?</td>
<td>Closed ended, yes/no (12 responses)</td>
</tr>
<tr>
<td>10</td>
<td>To what extent does the use of aromatherapy reduce the use of other pharmaceutical preparations for residents that receive aromatherapy?</td>
<td>Single visual analog</td>
</tr>
<tr>
<td>11</td>
<td>If aromatherapy reduces use of other pharmaceuticals in your facility, which ones does it reduce?</td>
<td>Open-ended</td>
</tr>
</tbody>
</table>

Part two of the survey completed by the aromatherapy care planners had ten information questions and four background questions (see Table 4.2). The rationale for the different questions is also outlined.
<table>
<thead>
<tr>
<th>Number</th>
<th>Question</th>
<th>Response type</th>
<th>Rationale</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>To what extent do you think the following are major causes of challenging behaviours of residents in this facility?</td>
<td>Multiple visual analog (16 responses)</td>
<td>Determine whether dementia is considered to be major cause of challenging behaviours</td>
</tr>
<tr>
<td>2</td>
<td>To what extent do you think the following environmental factors affect challenging behaviours of residents in this facility?</td>
<td>Multiple visual analog (12 responses)</td>
<td>Determine whether unpleasant ambient odour is thought to be a major cause of challenging behaviours compared to other environmental factors</td>
</tr>
<tr>
<td>3</td>
<td>How helpful do you think aromatherapy is in improving the following in this facility?</td>
<td>Multiple visual analog (20 responses)</td>
<td>Determine which behaviours and symptoms of dementia and other old age ailments are thought to be helped by aromatherapy. If the dementia symptoms factor together, new variable can be created to correlate with other independent variables.</td>
</tr>
<tr>
<td>4</td>
<td>Which aspects of aromatherapy help residents with dementia?</td>
<td>Multiple visual analog (8)</td>
<td>See if aromatherapy is thought to aid cognitive function.</td>
</tr>
<tr>
<td>5</td>
<td>How many residents in this facility receive aromatherapy, and how often?</td>
<td>Closed ended, ordinal</td>
<td>Compare aromatherapy practice variations between facilities</td>
</tr>
<tr>
<td>6</td>
<td>Of those who receive daily aromatherapy treatments, what is the average number of treatments per day?</td>
<td>Closed ended, ordinal</td>
<td>Compare aromatherapy practice variations between facilities</td>
</tr>
<tr>
<td>7</td>
<td>Which aromatherapy treatments are used in this facility?</td>
<td>Closed ended, ordinal (18) with option for ‘other’ response.</td>
<td>Compare aromatherapy practice variations between facilities</td>
</tr>
<tr>
<td>8</td>
<td>Which conditions are these essential oils mostly used for in this facility, and how widely?</td>
<td>Open-ended, list of oils provided, space for verbal response (22 oils, three options for ‘other’)</td>
<td>Investigate which conditions are most treated with aromatherapy, and whether any of the oils is used for cognitive function.</td>
</tr>
<tr>
<td>9</td>
<td>If you regularly use commercial oil blends please list blend name and company name below and the conditions they are most used for.</td>
<td>Open-ended, structure provided for 6 responses (blend name, company, condition most used for)</td>
<td>Investigate which conditions are most treated with blends, and whether any blends are used for cognitive function.</td>
</tr>
<tr>
<td>10</td>
<td>List any adverse effects of aromatherapy that have occurred in your facility.</td>
<td>Open-ended, structure provided for two responses (adverse effect, essential oil used, application method, your explanation)</td>
<td>Find out if any adverse events relate to cognitive function effects, to be contraindicated in subsequent clinical trial.</td>
</tr>
<tr>
<td></td>
<td><strong>Background information 1-4</strong></td>
<td>Open-ended, structure provided for 6 responses (blend name, company, condition most used for)</td>
<td>Gauge level of aromatherapy training – possibly affects aromatherapy practice?</td>
</tr>
</tbody>
</table>

In questions 1-4 where opinions were sought, multiple visual analog scales were used. In question 1, the list of possible causes of challenging behaviours was derived from the NRACAN discussion. It was thought that asking the first question about challenging behaviours would reveal to what extent staff perceive dementia to be a cause of challenging behaviours. In question 2, the list of environmental factors thought to affect challenging behaviours was derived from the NRACAN discussion. As aromatherapy does modify the environment by its odour, it was thought that it would be important to consider the extent to
which staff thought pleasant odour affected behaviours in comparison with all the other environmental factors.

In question 3, the list contains challenging behaviours caused by dementia taken from the Cohen-Mansfield Agitation Index (Cohen-Mansfield et al., 1990), and also other ailments and conditions that were thought to cause difficult behaviours arising from the NRACAN discussion. This list was intended to be as comprehensive as possible, to gauge which conditions the staff thought aromatherapy was most effective for. In question 4, the different aspects of aromatherapy thought to be effective for dementia were derived during the NRACAN discussion.

Questions 5-7 were closed-ended questions to gather information about the different types of application method, and different types of essential oil used and the frequency of use. The closed-ended lists of questions also included an option titled “other” with a space for respondents to write any answer that was not covered by the list of closed-ended questions. The frequency categories in question 5 and 6 and the application methods in question 7 were derived during the NRACAN meeting, based on knowledge and experience of NRACAN members.

Structured open-ended questions were used for questions 8-11 where the author wished to find out the practices and experiences of the aromatherapy planners. For example, Question 10 required responses to be written in a table with column headings such as “adverse effect”, “essential oil used”, “application method” and “your explanation”. The table guided the responses so that while still freely generated, the content of each respondent’s answers would cover the same four points, allowing for easier data coding and analysis.

4.2.4 Pilot testing of the survey

Once the questions had been written and the format drawn up, the survey was given to a member of the NRACAN group to check and see if she thought it would be appropriate for the target audience. She was a director of care and the aromatherapy care planner for her facility, so it was thought that she would be a good person to check it. It was also given to the university statistician to see if there would be any major problems with data coding or analysis that the author had missed. Comments from both these people were incorporated into the survey, and it was then considered ready for use. In retrospect, it would have been better
to do a formal pilot study with several respondents, and run the data analyses on their results, as suggested by Fink (2003), but at the time it was not considered necessary.

4.2.5 Participating facilities

A list of residential aged care facilities in the area (postcodes 2443-2487) was obtained from the Australian Department of Health and Ageing. Forty-eight facilities out of seventy-one facilities in the area responded to phone contact within the two weeks allotted for initial contact. It was considered that facilities that did not respond to telephone messages were unlikely to participate in the survey. During the first phone call it was determined whether facilities met the following inclusion criteria:

- Residents with dementia;
- An existing formal aromatherapy care plan;
- A person responsible for developing and evaluating the aromatherapy care plans.

Twenty-eight of the forty-eight facilities contacted met all the inclusion criteria and all had appropriate staff who wished to participate in the survey. Three of the excluded facilities used essential oil vaporisers in common rooms, but were included as they did not have a formal aromatherapy care plan. For reasons of privacy, the names and locations of facilities are not to be reported (see Patient Information Sheet in Appendix 1).

Surveyed facilities were mainly auspiced by religious organizations and ex-services organizations. Because this was a fact-finding survey about how aromatherapy is used in aged care, and not a survey to find out what factors influence whether a facility uses aromatherapy or not, the survey did not collect information about the facilities such as ‘number of years established’, and to protect privacy, the auspicing body was described as ‘government’ or ‘non-government’. All of the facilities studied were ‘non-government’. Data about the numbers of residents with and without dementia, and the number of beds in the facilities is found in the results section 4.3.1.

4.2.6 Survey administration

After ascertaining that a facility met the inclusion criteria, the Director of Care and the person responsible for designing the aromatherapy care plans were contacted by phone to ask if they would like to participate, and appointments were arranged for interviews. During the
interviews they first read information sheets about the survey process and signed consent forms and then filled in the surveys whilst sitting with the author (see end of the thesis for the information sheets, consent forms and surveys). It was considered acceptable by the Ethics Committee to have them read the information sheets and sign the consent forms during the interview, as the survey process had been explained during the initial phone conversation. Interviews with the Directors of Care took about 10 minutes, and interviews with the aromatherapy care planners took about half an hour. Interviews were carried out within a two month period, and surveys were filled in without any identifying marks, thus ensuring privacy and confidentiality of information. Ethics approval was obtained from the Human Research Ethics Committee at Southern Cross University (approval number ECN-03-71).

4.2.7 Data analysis methods

Responses to open-ended questions were grouped into themes for ease of analysis. The themes were developed during the initial observation of the responses, rather than being designed a priori. For example, in question 8, the themes were either considered to be behavioural or physical, each of these comprising several sub-themes. For the behavioural group, the themes were: agitated behaviours; depressed mood states; cognitive malfunction and; insomnia. For the physical group, the themes were: musculoskeletal; infections; skin integrity; circulatory problems; digestive problems and; ‘other ailments’. Once grouped into thematic categories, the number of responses in each category could be counted, giving a clearer picture of what types of condition were being treated by essential oils and which conditions were being most treated with aromatherapy.

Yes/no questions were coded 1/0, and responses on visual analogue scales were measured to ±0.1 cm. Thematic categories from the open-ended questions were also coded with ordinal codes. Data were entered into a spreadsheet and about 10% of entries were randomly double-checked for accuracy. Descriptive statistics for most of the raw data were generated, and the means and ranges reported.

Question three in the aromatherapy planner’s section had multiple visual analog rating scale responses to the question “How effective is aromatherapy for the following conditions in your facility?”. Factor analysis was used to discover if any of the responses clustered together, and could therefore be made into a new variable corresponding to the respondents’ perceptions of the efficacy of aromatherapy. The factor analysis method chosen was a principal components
extraction with varimax rotation. If the factor analysis yielded such a factor of ‘perceived efficacy’, it was planned to carry out Pearson’s correlations with other independent variables (such as numbers of people with dementia in the facility) to gauge if any other variables were affecting people’s perceptions of the efficacy of aromatherapy. Question four in the same section also was subjected to factor analysis to see whether the responses about cognitive function clustered together. The statistical package used for all of the statistical analyses was SPSS for Windows v11.01 (Statistical Package for Social Sciences, LEAD Technologies, Inc).

4.3 Results

The results from the two parts of the survey have been combined and presented together to answer the aims of the survey. Staff qualifications levels, ‘funding for aromatherapy’ and ‘average cost of aromatherapy materials’ are not reported here because the results did not contribute to the development of the subsequent laboratory work or clinical trial. It was decided to keep the presentation of the results related to items that contributed to the furthering of the project, rather than present all the material. Similarly, results for questions about causes of, and environmental effects on, challenging behaviours from the aromatherapy care planners’ section are not reported as they did not pertain directly to that aim.

4.3.1 Numbers and percentages of residents receiving treatment

The total number of residents in the participating facilities was 1767 comprising 827 (47%) with dementia and 940 (53%) without dementia. A total of 1032 (59%) residents received aromatherapy including 468 residents with dementia which was 57% of the total number of residents with dementia and 564 residents without dementia which was 60% of the total number of residents without dementia. The mean number of residents per facility was 63.0 (sd 34.3), minimum 12, maximum 183 people.

The facilities varied as to whether they used aromatherapy as a treatment for all residents, or only for a few residents. Only five facilities (18%) give all of their residents aromatherapy treatments. 14 facilities (50% of the facilities surveyed) give all of their dementia residents aromatherapy (though not necessarily daily), whereas only nine facilities (32% surveyed) give all of their non-dementia residents aromatherapy.
4.3.2 Perceptions of efficacy

Perceptions of efficacy of aromatherapy for various behaviours and symptoms were collected in Question 3 on the aromatherapy care planners’ survey. Perceptions were measured on 10 cm visual analogue scales. The two highest mean scores were ‘reduction in arthritic pain’ (8.3, SD = 1.4, range 4.7–10); and ‘increase in relaxation’ (8.0, SD = 1.6, range 3.3–10). However, most of the data were bi-modal with opinions clustered in two groups, either near the top or the bottom end of the range. As it is not statistically valid to compare the means of bi-modally distributed variables, factor analysis was used to group highly correlated items into new variables (see Table 4.3 and Table 4.4). Components with initial eigenvalues over 1 were considered to be significant factors in explaining the variance in responses.

<table>
<thead>
<tr>
<th>Component</th>
<th>Initial Eigenvalues</th>
<th>Extraction Loadings</th>
<th>Sums of Squared Loadings</th>
<th>Rotation Loadings</th>
<th>Sums of Squared Loadings</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
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<td>58.277</td>
<td>58.277</td>
<td>11.655</td>
<td>58.277</td>
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<tr>
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<td>2.641</td>
<td>13.205</td>
<td>71.481</td>
<td>2.641</td>
<td>13.205</td>
</tr>
<tr>
<td>3</td>
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<td>9.103</td>
<td>80.584</td>
<td>1.821</td>
<td>9.103</td>
</tr>
<tr>
<td>4</td>
<td>1.407</td>
<td>7.034</td>
<td>87.618</td>
<td>1.407</td>
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<td>99.616</td>
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<td>12</td>
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<td></td>
<td></td>
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</table>

Extraction Method: Principal Component Analysis.
Table 4.4 Rotated component matrix for factor analysis of perceptions of efficacy of aromatherapy.

<table>
<thead>
<tr>
<th>Component (a)</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
</tr>
</thead>
<tbody>
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<td>.256</td>
<td>1.829E-02</td>
<td>.109</td>
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<tr>
<td>verbal aggression</td>
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<td>.255</td>
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<td>.679</td>
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<td>6.260E-02</td>
<td>.124</td>
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<tr>
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<td>8.928E-02</td>
<td>.464E-02</td>
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<td>confusion</td>
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<td>.216</td>
<td>5.415E-02</td>
<td>.257</td>
<td>-.170</td>
</tr>
<tr>
<td>restlessness</td>
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<td>.449</td>
<td>-.183</td>
<td>.369</td>
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<tr>
<td>absconding</td>
<td>.155</td>
<td>.357</td>
<td>.141</td>
<td>.880</td>
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<tr>
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<td>-.171</td>
<td>.580</td>
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<tr>
<td>reduce pain</td>
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<td>.109</td>
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<tr>
<td>social withdrawal</td>
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<td>.355</td>
<td>.115</td>
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Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.

a Rotation converged in 9 iterations.
Shaded cells for component 1 comprise the items in the new variable.

The first component derived from factor analysis accounted for 58% of the variation, and was composed of the following highly correlated items: reduction of agitation, anxiety, muscle tension, pain, restlessness, confusion, depression, insomnia, and increase of relaxation. As agitation, anxiety, restlessness, confusion and insomnia are all symptoms of dementia (Cummings, 1997) the above-mentioned grouping from the factor analysis was treated as a single variable indicating how useful aromatherapy was perceived to be for dementia. The new variable is referred to in the text as the ‘perceptions of efficacy’ variable for dementia symptoms. Correlations were then examined between this new variable and other independent variables to explore whether any characteristics of the facilities or the survey respondents affected how people perceived the effectiveness of aromatherapy for dementia.
4.3.3 Aromatherapy treatment protocols

In the survey, responses to the question “How often do residents receive aromatherapy treatments?” were either: daily; three or more times a week; 1–2 times a week; or less than once a week, to fit with the established care plan protocols. Seventy-five percent (75%) of dementia residents receiving aromatherapy were treated daily or \( \geq 3 \) times per week, whereas 63% of non-dementia residents receiving aromatherapy were treated daily or \( \geq 3 \) times per week as shown in Table 4.5.

<table>
<thead>
<tr>
<th>Group</th>
<th>Daily</th>
<th>( \geq 3 ) per week</th>
<th>1–2 per week</th>
<th>(&lt; 1 ) per week</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dementia (n = 468)</td>
<td>201 (43%)</td>
<td>150 (32%)</td>
<td>70 (15%)</td>
<td>46 (10%)</td>
</tr>
<tr>
<td>Non-dementia (n = 564)</td>
<td>282 (50%)</td>
<td>73 (13%)</td>
<td>113 (20%)</td>
<td>96 (17%)</td>
</tr>
</tbody>
</table>

The numbers of residents in each group (dementia or non-dementia) are the sum of residents in each group receiving aromatherapy in the 28 facilities.

4.3.3.1 Daily treatments and perceptions of efficacy

Eleven facilities gave 100% of their treated dementia residents daily treatments, and nine facilities gave 100% of their treated non-dementia residents daily treatments. However, there were no statistically significant correlations between numbers of residents with or without dementia receiving daily treatments and the ‘perceptions of efficacy’ variable for dementia symptoms.

4.3.3.2 Number of aromatherapy applications per day

The aromatherapy care planners were asked how many applications of aromatherapy were carried out per day. The mean number of treatments given per day was 1.5 (sd 1.934, range 1-9, median and mode both = 1). There was no statistically significant correlation between the number of aromatherapy applications per day and ‘perceptions of efficacy’ variable for dementia symptoms.

4.3.3.3 Application methods

The occurrence of seventeen different application methods was surveyed. Twenty-six out of twenty-eight facilities used footbaths and foot-massages, 25/28 used neck-and-shoulder and hand massages, 24/28 used limb or joint massage, 21 used drops on tissue or pillow, 19 used...
abdominal massage, and 18 used vaporisers, after-shower lotions and skin integrity creams and 14 used aromatherapy spritzers (face sprays). The least prevalent applications methods were: steam inhalation, compresses, wound-dressings, full body massage and baths (<10 facilities). Facilities used a mean of 10.0 different application methods (sd = 3.39, range 2-15).

Use of aromatherapy spritzers was the only application method that positively correlated with the ‘perception of efficacy’ variable for dementia symptoms (two-tailed Pearson’s correlation coefficient = 0.516, p = 0.008, n = 25).

### 4.3.3.4 Most useful aspects of an aromatherapy treatment

Survey respondents were asked which of the following components of aromatherapy treatments they thought were most useful for dementia: touch; one-to-one contact; memory stimulation; reminiscence due to smell; relaxing effect; calming effect; and a sense of nurturing. Factor analysis (using the same method as that used to derive the ‘perceptions of efficacy’ variable) yielded two components with eigenvalues over 1, the first factor comprising touch, one-to-one contact and nurturing. This factor represented 51% of the variance, indicating that respondents felt these aspects of aromatherapy were more important than the secondary factor containing mental stimulation and reminiscence due to the smell of the oils.

### 4.3.3.5 Essential oils used

The list of oils in the survey was generated at the initial NRACAN meeting of aged care nursing staff as being those most likely to be used in aged care. Respondents were also asked to list any other oils they used that were not on the list. The only oil that was used by every facility was Lavender. Tea tree oil was next most widely used (75% of facilities). Table 4.6 lists the oils used and the percentage of facilities using that aromatherapy oil. Only common names for the aromatherapy oils were used in the survey, so botanical names have not been included in this section of the thesis.
Table 4.6 Percentage of facilities using different essential oils.

<table>
<thead>
<tr>
<th></th>
<th>≥ 50%</th>
<th>25–49%</th>
<th>&lt;25%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lavender (100%)</td>
<td>Cypress (39%)</td>
<td>Orange 21%</td>
<td></td>
</tr>
<tr>
<td>Tea tree (75%)</td>
<td>Roman chamomile (39%)</td>
<td>Myrrh 18%</td>
<td></td>
</tr>
<tr>
<td>Bergamot (64%)</td>
<td>Basil (36%)</td>
<td>Peppermint 18%</td>
<td></td>
</tr>
<tr>
<td>Eucalyptus (64%)</td>
<td>Frankincense (36%)</td>
<td>Lemongrass 14%</td>
<td></td>
</tr>
<tr>
<td>Geranium (64%)</td>
<td>German Chamomile (32%)</td>
<td>Melissa 14%</td>
<td></td>
</tr>
<tr>
<td>Ginger (61%)</td>
<td>Lemon (32%)</td>
<td>Ylang-ylang 14%</td>
<td></td>
</tr>
<tr>
<td>Rosemary (57%)</td>
<td>Lime (32%)</td>
<td>Cedarwood 11%</td>
<td></td>
</tr>
<tr>
<td>Clary Sage (54%)</td>
<td>Sandalwood (29%)</td>
<td>Rosewood 11%</td>
<td></td>
</tr>
<tr>
<td>Sweet Marjoram (54%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Black Pepper (50%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Juniper (50%)</td>
<td>Mandarin 7%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Black Pepper (50%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rosemary (55%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Basil (54%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bergamot (54%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Eucalyptus (54%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Geranium (54%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ginger (54%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rosemary (57%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Clary Sage (54%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Black Pepper (50%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Juniper (50%)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

4.3.3.6 Oils used specifically to improve cognitive function

Very few comments (13/463 = 2.8%) suggested that essential oils were being chosen specifically for cognitive function improvement (see Table 4.7). Rosemary, Basil, Clary Sage, Lime, Roman Chamomile, Sage and Sandalwood were thought by at least one survey respondent to be useful for improvement of some aspect of cognitive function.

Table 4.7 Oils recommended for cognitive function improvement

<table>
<thead>
<tr>
<th>Oil</th>
<th>Cognitive function stated</th>
<th>Number of statements</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rosemary</td>
<td>Alertness</td>
<td>1</td>
</tr>
<tr>
<td>Basil</td>
<td>Concentration</td>
<td>1</td>
</tr>
<tr>
<td>Clary Sage</td>
<td>Confusion</td>
<td>1</td>
</tr>
<tr>
<td>Lime</td>
<td>Mental stimulation</td>
<td>1</td>
</tr>
<tr>
<td>Roman Chamomile</td>
<td>Confusion</td>
<td>1</td>
</tr>
<tr>
<td>Sage</td>
<td>Confusion</td>
<td>1</td>
</tr>
<tr>
<td>Sandalwood</td>
<td>Confusion</td>
<td>1</td>
</tr>
</tbody>
</table>

4.3.3.7 Adverse effects of essential oils

Survey respondents were asked to report on the nature and frequency of any adverse effects they had noticed during their practice in the facilities. Twelve adverse effects were reported,
seven for residents and five for staff members. Lavender oil was reported as the cause in 7
incidents, blends containing Lavender caused 2 incidents, Ylang-ylang oil caused 1 incident,
and the oil or blend was not specified in two incidents.

The most severe resident reactions were two skin irritation reactions to massage oil containing
Lavender oil, one of which was 13 drops of Lavender in 30mLs of macadamia oil. The other
reactions were increase in agitation (3), headache (1) and one comment that the resident didn’t
like the smell. Four of these reactions were due to either Lavender oil or a blend containing
Lavender, and one resident increased in agitation when any essential oil was used, including
Lavender oil.

Two staff members reported headaches during vaporisation of oils, one to Ylang-ylang, and
the other to Lavender oil. One staff member with emphysema reported dyspnoea while
administering a foot-bath, but didn’t know which oil it was. One staff member reported hay-
fever when massaging feet with Lavender oil, and turned out to have an allergy to Lavender
oil. One staff member increased in agitation during vaporisation of Lavender oil.

4.3.3.8 Commercial blends used

Fifteen of the 28 respondents 15/28 (54%) stated that their facilities used commercial
aromatherapy blends. In each case, they used two or more different blends. There was no
significant correlation between the number of oils used and the number of blends used. The
facility with the lowest usage of individual oils (only Lavender oil) used two blends, a
calming and a joint relief blend. Blend types were categorised as used for: calming; sleep
promoting; arthritis management; mood uplifting or nurturing; for respiratory ailments; for
oedema management.

4.3.3.9 Categories of staff giving aromatherapy treatments

Aromatherapy was applied by assistants-in-nursing (AINs) in 96% (27/28) of the facilities
and Activities Officers (AOs), in 68% of facilities. Registered nurses (RNs) applied
aromatherapy in 60% of facilities, and diversional therapists did so in 40% of facilities.
Physiotherapy aides applied aromatherapy in 32% of facilities. Only four facilities employed
the services of a visiting aromatherapist. In two cases, this aromatherapist was not the
aromatherapy care planner who responded to the survey but a person employed by the residents on an individual basis.

Occupational therapists and physiotherapists were least represented among health professionals applying aromatherapy and were only identified in one facility. Volunteers and family members were involved in aromatherapy application in 43% and 39% of facilities, respectively.

4.3.4 Health conditions for which aromatherapy is used

Respondents were also asked to indicate the primary conditions for which they used the oils. As shown in Table 4.8, of the 463 responses from the 28 surveys, 46% of responses related to behavioural and psychological symptoms, of which agitation and anxiety type behaviours were the most prevalent.

Management of arthritis symptoms and musculoskeletal discomfort were the next most prevalent conditions, followed by management of infections, mainly respiratory diseases and topical fungal infections. Although the listed behavioural and psychological conditions correspond well with symptoms commonly found in residents with dementia, survey respondents did not usually distinguish whether they thought the symptom was due to dementia.

<table>
<thead>
<tr>
<th>Conditions that oils are used for</th>
<th>% of responses</th>
</tr>
</thead>
<tbody>
<tr>
<td>Behavioural and psychological symptoms</td>
<td></td>
</tr>
<tr>
<td>Agitation, anxiety, restlessness, aggression, tension, stress, lack of calmness</td>
<td>25</td>
</tr>
<tr>
<td>Depression, withdrawal, apathy, grief, moodiness</td>
<td>12</td>
</tr>
<tr>
<td>Confusion, concentration, poor memory, ‘mind and spirit’</td>
<td>5</td>
</tr>
<tr>
<td>Insomnia</td>
<td>3</td>
</tr>
<tr>
<td>Total</td>
<td>46%</td>
</tr>
<tr>
<td>Physical ailments</td>
<td></td>
</tr>
<tr>
<td>Arthritis, rheumatism, joints, muscles, aches, pains, stiffness</td>
<td>21</td>
</tr>
<tr>
<td>Infections-respiratory, fungal infections, viral infections, wound healing</td>
<td>16</td>
</tr>
<tr>
<td>Skin-integrity, dryness, itch, care</td>
<td>6</td>
</tr>
<tr>
<td>Poor circulation, oedema, fluid retention, altering blood pressure</td>
<td>4</td>
</tr>
<tr>
<td>Digestive system-constipation, indigestion</td>
<td>4</td>
</tr>
<tr>
<td>Other ailments-emphysema, diabetes, hormonal imbalance, palliation</td>
<td>3</td>
</tr>
<tr>
<td>Total</td>
<td>54%</td>
</tr>
</tbody>
</table>
Most of the oils listed were recommended for several different conditions, including behavioural or psychological symptoms and physical symptoms. Only seven oils were recommended solely for physical conditions: Black Pepper, Eucalyptus, Ginger, Juniper, Lemongrass, Peppermint and Tea Tree. All the other oils had at least one recommendation for use with a behavioural or psychological symptom.

Conditions for which commercial blends were used are shown in Table 4.9. Blends were used more for behavioural or psychological symptoms, rather than physical symptoms, although the reverse was true for the single essential oils. Some blends were used for more than one condition. As with the essential oils, calming and relaxing blends were considered to be different than blends for sleep promotion.

The groups of symptoms and ailments used in Table 4.9 were clustered by thematic similarity of the symptoms or ailments. Each condition could have been listed separately, but in the mind of the author it appeared more useful to cluster them into themes as shown. Insomnia was included as a separate item as it occurred so frequently.

Table 4.9 Conditions that blends were used for.

<table>
<thead>
<tr>
<th>Conditions that blends are used for</th>
<th>% of responses</th>
</tr>
</thead>
<tbody>
<tr>
<td>Behavioural or psychological</td>
<td></td>
</tr>
<tr>
<td>symptoms</td>
<td></td>
</tr>
<tr>
<td>Agitation, anxiety, restlessness,</td>
<td>23</td>
</tr>
<tr>
<td>aggression, tension, lack of</td>
<td></td>
</tr>
<tr>
<td>calmness</td>
<td></td>
</tr>
<tr>
<td>Depression, grief, emotional</td>
<td>16</td>
</tr>
<tr>
<td>imbalance</td>
<td></td>
</tr>
<tr>
<td>Confusion, concentration, poor</td>
<td>–</td>
</tr>
<tr>
<td>memory,</td>
<td></td>
</tr>
<tr>
<td>Insomnia</td>
<td>22</td>
</tr>
<tr>
<td>Total</td>
<td>61%</td>
</tr>
<tr>
<td>Physical ailments</td>
<td></td>
</tr>
<tr>
<td>Arthritis, rheumatism, joints,</td>
<td>25</td>
</tr>
<tr>
<td>muscles, aches, pains, stiffness</td>
<td></td>
</tr>
<tr>
<td>healing</td>
<td></td>
</tr>
<tr>
<td>Infections-respiratory, fungal</td>
<td></td>
</tr>
<tr>
<td>infections, viral infections,</td>
<td></td>
</tr>
<tr>
<td>wound</td>
<td></td>
</tr>
<tr>
<td>Skin-integrity, dryness, itch, care</td>
<td>3</td>
</tr>
<tr>
<td>Poor circulation, oedema, fluid</td>
<td>4</td>
</tr>
<tr>
<td>retention, altering blood pressure</td>
<td></td>
</tr>
<tr>
<td>Digestive system-constipation,</td>
<td>–</td>
</tr>
<tr>
<td>indigestion</td>
<td></td>
</tr>
<tr>
<td>Other ailments-emphysema, diabetes,</td>
<td></td>
</tr>
<tr>
<td>hormonal imbalance, –</td>
<td></td>
</tr>
<tr>
<td>palliation</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>38%</td>
</tr>
</tbody>
</table>

Total number of facilities that use blends = 15, total number of responses = 69. The missing 1% is due to a blend used for ‘therapist balance’.

4.3.5 Perceptions about reduction of pharmaceutical use

In 22 facilities, directors of care perceived the use of sedatives to be reduced due to residents receiving aromatherapy treatments (see Table 4.10). This correlated significantly with perceptions of overall reduction of pharmaceutical use (Pearson’s = 0.491, two tailed
The 11 other classes of drugs perceived by directors of care to be reduced by aromatherapy use were: analgesics, benzodiazepines, antipsychotics, aperients, anti-depressants, arthritis medication, diuretics, respiratory drugs, anti-hypertensives, skin care creams and anti-fungal preparations.

Table 4.10 Types of drug usage reduced by aromatherapy.

<table>
<thead>
<tr>
<th>Type of drug</th>
<th>Facilities reporting % (n)</th>
<th>Residents receiving aromatherapy % (n)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sedatives</td>
<td>79% (22)</td>
<td>63% (860)</td>
</tr>
<tr>
<td>Analgesics</td>
<td>39% (11)</td>
<td>79% (486)</td>
</tr>
<tr>
<td>Benzodiazepines</td>
<td>21% (6)</td>
<td>84% (229)</td>
</tr>
<tr>
<td>Anti-psychotics</td>
<td>21% (6)</td>
<td>42% (168)</td>
</tr>
<tr>
<td>Aperients</td>
<td>14% (4)</td>
<td>69% (158)</td>
</tr>
</tbody>
</table>

Types of drug perceived by the Directors of Care (n = 28 facilities) to be reduced by aromatherapy. The percentage of residents receiving aromatherapy is calculated as a percentage of the total number of residents represented by the reporting facilities. Types of medication listed as being used by only one or two facilities are not reported in the table.

4.4 Discussion

4.4.1 Survey quality

The survey quality could have been improved by proper pilot testing, including the data analysis stage. This would have enabled the author to pre-plan the data analysis better, and perhaps find more meaningful ways of talking about ‘perceptions of efficacy’ of aromatherapy than using the factor analysis method. Future surveys done by the author will be pilot tested. Pilot testing would also allow for the validity and reliability of the survey to be checked.

For instance, in a pilot test, validity of the survey questions could be checked by writing additional questions with different words but similar meanings, and comparing similarity of responses. Objective measures of pharmaceutical usage, aromatherapy dosages and associated costs could also be obtained by checking facility records. These could then be compared with the opinions of survey respondents. Such comparisons would increase the validity and reliability of the survey.
The survey was not tested for reliability which reduces the usefulness of the results, especially the opinion questions, as these are most likely to fluctuate over time (de Vaus, 1995). Unfortunately many survey respondents were unavailable to repeat the survey to gauge test-retest reliability due to requirements of government accreditation procedures during this phase of the research. However, as the survey was intended as a data-gathering tool for the subsequent laboratory and clinical trial work, it was not considered essential for this project. If the survey is used again, researchers should incorporate test-retest reliability measures.

4.4.2 Survey results

The survey set out to gather information about current aromatherapy practices for dementia in aged care facilities. The stated aims were to:

- Describe the variations in aromatherapy practices in aged care facilities in northern NSW.
- Identify how effective aromatherapy is perceived to be for dementia.
- Determine if any aromatherapy practices correlate significantly with perceptions of efficacy for dementia.
- Identify the essential oils most used in aged care, and in particular those thought to be effective for dementia or cognitive impairment.
- Identify elements for an appropriate treatment regime for the subsequent clinical trial protocol.

The results of the survey will be discussed in this order, ending with consideration of how the results contribute to the development of an appropriate treatment regime for the subsequent clinical trial protocol.

4.4.3 Aromatherapy practices in aged care facilities

The essential oils used and the conditions for which they were used confirm that aromatherapy is being used for a mixture of behavioural, psychological and physical symptoms. Aromatherapy is perceived to be most effective for management of agitation, anxiety, nervous tension, depression and sleeplessness. It is also used widely for management of physical pain and infections. Resolving pain and infection may help reduce behavioural and psychological symptoms of dementia (Paton et al., 2004).
Aromatherapy is also perceived as a more useful treatment for residents with dementia than those without – 14/28 facilities (50%) give aromatherapy to 100% of their residents with dementia, whereas only 9/28 facilities (32%) give aromatherapy to 100% of their residents without dementia.

The finding that directors of care perceived aromatherapy to reduce pharmaceutical usage suggests that quantitative assessment of actual pharmaceutical use reduction while using aromatherapy would be a useful follow-up study. In particular, use of sedatives, benzodiazepines and anti-psychotic medications was perceived to be reduced while using aromatherapy. This was corroborated by the survey findings that showed aromatherapy was also being used for sleeplessness and depression, both of which often accompany dementia (Draper et al., 2001). Sedatives, benzodiazepines and anti-psychotic medications are often used for management of anxiety, aggression and agitation in the later stages of dementia (Draper et al., 2001, Borson & Raskind, 1997). The survey findings suggest that aromatherapy is perhaps more useful in dementia management than the survey participants perceived, and that it is used extensively in northern NSW aged care facilities to manage behaviours and symptoms that occur in dementia, as well as in non-demented people.

### 4.4.4 Perceived efficacy of aromatherapy for dementia

While the survey revealed that aromatherapy was perceived to be effective for many of the psychological and behavioural symptoms of dementia, it was not perceived as effective for improving reminiscence or memory.

The data relating to perceptions of efficacy of aromatherapy had mostly bi-modal distribution. This implies that there are care planners who perceive aromatherapy to be very effective for various symptoms of dementia, and others who perceive it to be less effective, with not much of a middle ground. This was unexpected, especially as there were no significant correlations of perceived efficacy of aromatherapy with the percentage of residents being treated or percentage of residents with dementia. The propensity to perceive aromatherapy as effective may correlate with optimistic or enthusiastic character traits of the care planners.

Using factor analysis to derive new variables is a standard statistical procedure. However, the fact that the ‘perceptions of efficacy’ variable generated by factor analysis only correlated significantly with one item (use of sprays), suggests that either the new variable was not an
appropriate measure of efficacy, or that respondents’ perceptions of aromatherapy’s efficacy are more closely linked to some other factor not appearing in the survey. For many respondents, it was the first time they had been asked to quantify their opinions about aromatherapy in such a formal way, and perhaps this also contributed to the variation in perceptions.

However, the items that were factored into the new ‘perceptions of efficacy’ variable were: reduction of agitation, anxiety, muscle tension, pain, night restlessness, confusion and insomnia, and increase of relaxation. The inclusion of these items in the same factor suggests that these are things perceived as most affected by aromatherapy. Agitation, anxiety, night restlessness and confusion are all symptoms of dementia (Cummings, 1997).

4.4.5 Application methods

Foot-baths, and foot and hand massages were used in 26 out of 28 facilities. The popularity of footbaths and foot massage is possibly due to the ease of administration, and the fact that several facilities also had an established podiatry regime into which aromatherapy could be easily incorporated (discovered from informal discussion with respondents). Some of the least used methods would take too long or be impractical for regular use in an institutional setting (for example steam inhalation and full body massage). More pertinent perhaps, is the issue that the most effective delivery route for maximisation of the pharmacological effects of essential oils has not yet been established, and may well vary according to the condition being treated.

Use of an aromatherapy spray was the only aromatherapy practice that positively correlated with the ‘perception of efficacy’ variable for dementia symptoms, suggesting that perhaps aromatherapy sprays could be a useful application method, but they were only used by half the facilities. Use of sprays for people exhibiting agitation, anxiety, aggression and restlessness could be researched further in comparison with dermal application methods.

Only half the facilities used essential oils in a vaporiser in the public areas of the facility. This could be due to a number of reasons. These could range from the possibility that aromas can be offensive to some people through to the rationale that if the essential oils are being used as therapeutic agents, it may be unethical to have them present in the air for people for whom they have not been prescribed, including staff and visitors.
4.4.6 The most-used essential oils

The essential oils most used in aged care were among those listed on the survey form, rather than those generated by respondents. The fact that some form of Lavender oil was used in every facility is not surprising, as it has a reputation for being one of the safest essential oils in the industry (Cavanagh & Wilkinson, 2002). Lavender oil is also thought to be one of the most versatile oils, with a reputation for reducing nervous tension and inducing sleep as well as being a topical anti-inflammatory agent (Battaglia, 1995).

The wide-spread usage of Tea Tree oil for fungal infections, particularly of the feet, suggests that aged care aromatherapists are as interested in remediation of physical ailments as they are in mood-enhancement. It may also be that the fungal effects of Tea Tree oil are among the more well-researched effects of essential oils (Martin & Ernst, 2004), so nurses feel more confident in using Tea Tree.

Bergamot, Geranium and Clary Sage oils are all used for uplifting depressed mood and reducing stress and anxiety (Battaglia, 1995). It is understandable therefore why they would be in the list of most-used essential oils in aged care facilities with people with dementia. Sweet Marjoram oil has a reputation as a sleeping aid and for calming aggression and anxiety (Battaglia, 1995).

Eucalyptus, Rosemary, Black Pepper, Ginger and Juniper oils are all used for management of physical aches and pains and improving circulation (Battaglia, 1995). Arthritis was noted as a major reason for use of essential oils in the survey, so it is not surprising that these oils are among the most-used in the facilities surveyed.

Commercial blends appeared to be useful for agitation and anxiety, sleeplessness, depression and for arthritic pain. In a busy aged care facility, pre-blended essential oils offer an easier treatment option than the preparation of an individualised blend for each person. Further research comparing commercial blends with individualised blends could demonstrate whether aromatherapy needs to be individualised to be effective.

The oils used for stress reduction contain high percentages of monoterpenoid alcohols. The compound (-)-linalool is in high proportions in Lavender, Bergamot and Clary Sage oils, while Sweet Marjoram has terpinene-4-ol and Geranium has geraniol. (-)-Linalool is most
likely an active component in the stress-reduction and analgesic properties, given its anti-convulsive interaction with glutamate receptors in vitro (Brum et al., 2001) and its analgesic and anti-nociceptive properties in animal models (Peana et al., 2004). No research has been done on the stress-reducing properties of geraniol or terpinen-4-ol, though both have antibacterial and anti-fungal properties (Pattnaik et al., 1997, Cox et al., 2000).

The compound 1,8-cineole found in high proportions in Eucalyptus and Rosemary oils shows potential as an anti-inflammatory and analgesic component (Santos & Rao, 2000), which might contribute to the use of these oils for muscular aches and pains.

The research on analgesic and anti-inflammatory properties of Ginger relates to the pungent gingerol and shogaol compounds, which are only present in trace amounts in steam-distilled Ginger essential oil. The main compounds of Ginger essential oil, zingiberene and curcumene do not have well-established anti-inflammatory properties in humans, although animal studies suggest they do have anti-inflammatory properties (Chrubasik et al, 2005).

Similarly, the main components of Juniper are not known for their anti-inflammatory or analgesic properties. However, beta-caryophyllene, one of the components in Black Pepper oil, is reported to have in vitro local anaesthetic (Ghelardini et al., 2001) and anti-inflammatory effects, (Tambe et al., 1996).

4.4.7 Essential oils used for dementia

None of the respondents stated that any particular oils were primarily used for dementia. Instead respondents stated which oils were primarily used to enhance alertness, memory and concentration, and to reduce confusion. Only 5% of responses related to these items, and Rosemary, Basil, Clary Sage, Lime, Roman Chamomile, Sage and Sandalwood were the oils listed. Of these, only Rosemary was used by more than 50% of the facilities. These findings reflect the discussion held with the NRACAN members at the start of the project that aromatherapy is not used in aged care facilities to treat dementia per se, but to treat symptoms and behaviours caused by dementia. All these oils were included in the list of oils to be tested in the laboratory.
4.4.8 Implications for the clinical trial

4.4.8.1 Regional suitability for aromatherapy research

The results suggest that aromatherapy is widely accepted as a worthwhile therapy for residents of aged care facilities in the Northern Rivers region of NSW, and that it would be a suitable region to carry out a clinical trial of aromatherapy.

4.4.8.2 Dosing regimen

One of the issues facing aromatherapy as a profession is the issue of effective dosage. The survey did not collect data about the concentration of oils used in the various application methods, because there is no established evidence for effective essential oil dosage for different conditions, and the survey would have become cumbersome trying to cover all conditions and dosage possibilities. The discovery that 47\% of residents receive between 1-2 daily aromatherapy treatments suggests that aromatherapy may be being used in a similar way to other types of medication, aiming to maintain blood-levels of the drugs within a therapeutic range. However, no significant correlations between perceptions of efficacy and dosage frequency were found, so there was no emergent rationale for dosage frequency arising from the survey.

A large proportion of residents with dementia (75\%) received 3 or more aromatherapy treatments per week. This suggests that aromatherapy was perceived to be most effective at this frequency. Further investigation would be required to establish the most effective dosage frequency, though the fluctuating nature of dementia may pose some difficulties.

4.4.8.3 Most useful components of aromatherapy treatments

Survey respondents were asked which components of aromatherapy treatments they thought were most useful for dementia. After factor analysis, respondents felt that touch, one-to-one contact and nurture were more important than mental stimulation and reminiscence due to the smell of the oils. These results corroborated the findings of the aromatherapy literature review, so it was decided that the clinical trial should use a touch-based application method rather than inhalation alone. The results also suggested the use of a control arm receiving touch but no essential oils.
4.4.8.4 Type of people administering aromatherapy

In order to determine who would be the most appropriate people to administer aromatherapy treatments during the clinical trial phase, each facility was asked what type of people in the facility were involved in the application of aromatherapy. Assistants-in-nursing (AINs) were involved in 27 of the 28 facilities. This was followed by Activities Officers in 19/28 facilities, and registered nurses in 16/28 facilities.

These results indicated that AINs are considered able to administer the therapy. It was decided that for the clinical trial, it would be appropriate and probably the least disruption to normal nursing routine to ask AINs to assist with the treatment application. The choice of AINs was a logistical matter, designed to reduce stress on the already busy nursing staff.

4.4.8.5 Application methods used

The survey did not have a section asking about the frequency of use of each application method, but in discussion with some of the staff during the surveys, it was ascertained that the foot-baths and foot-massages were often done accompanying a podiatry or foot-hygiene session, which would only be about once a week or once a fortnight. The daily treatments were more likely to be massage, drops on the pillow, vaporisers and after-shower lotions.

Although the aromatherapy spray correlated significantly with the ‘perception of efficacy’ variable, it was felt that a pump-pack lotion bottle would deliver a more easily measured dose than a spray, and would also be less likely to deteriorate.

Therefore, it was decided for the clinical trial protocol to apply an aromatherapy lotion to the neck and shoulders as most facilities already used this technique and it would also be quicker to carry out than a foot-bath or foot massage. Speed of application was considered important, as care staff in the aged care facilities told researchers informally that they had very little time in their routine for doing extra treatments like aromatherapy, or for participating in extra activities like research.

4.4.8.6 Oils used specifically to improve cognitive function

The small number of comments about essential oils being chosen for cognitive function improvement suggested that nurses are not primarily using aromatherapy for cognitive
function improvement in dementia. This is possibly due to the paucity of research on aromatherapy and dementia, or due to the fact that nobody has previously thought of trying to stimulate cognitive function in dementia with aromatherapy, perhaps considering aged care facility residents to be “too far gone” to respond to treatment.

Each of Rosemary, Basil and Peppermint oils would be anticipated choices for memory and alertness (Price, 1998). In spite of anecdote, Peppermint oil was not recommended by any facility for improving alertness or mental stimulation. Sage oil is not usually used in mainstream aromatherapy due to its neuro-toxic thujone content (Battaglia, 1995), although this precaution may be unnecessary given the tiny doses used in aromatherapy.

Although it appeared from the survey that aromatherapists in aged care facilities did not target cognitive improvement with specific essential oils, the cholinesterase-inhibiting possibilities of essential oils were still intriguing enough to pursue the cholinesterase-inhibiting hypothesis. It was decided to screen the oils that were used in more than 25% of aged care facilities for \textit{in vitro} cholinesterase inhibiting properties.

### 4.4.8.7 Adverse effects of essential oils

The small number of adverse reactions reported in the survey compared to the total number of residents receiving aromatherapy appears to indicate that aromatherapy is a relatively safe therapy, although extra care should be taken with the use of Lavender oil.

It was decided that for the clinical trial, essential oils used would be patch-tested on participants prior to the trial to reduce the possibility of skin irritation, and participants would be asked if they like the smell of the oils to be used in the trial before application, thereby reducing the risk of increasing agitation or headache. If skin irritation was to occur, the base oil or carrier lotion should also be patch-tested, as some people have allergies to preservatives used in commercial bases or to nut oils.

### 4.5 Summary

The survey showed that aromatherapy was thought to be effective for the management of psycho-emotional issues like anxiety, agitation, sleeplessness, and the management of
musculo-skeletal discomfort and pain. Nursing staff were more likely to consider aromatherapy effective for various symptoms and conditions rather than underlying disease states such as dementia.

As no oils were specifically suggested to be good for memory in more than 4 facilities, it was decided that all the oils used in 25% or more of the facilities should be tested for \textit{in vitro} acetylcholinesterase inhibition, and the clinical trial blends selected according to laboratory results.
5 Research hypothesis and plan

From the literature review and the survey, it appears that aromatherapy is not considered useful for the improvement of cognitive function in aged care facility residents with dementia. Whether this is because the residents with dementia are considered to be too far advanced in the disease for cognitive enhancement agents to make an effect, or whether aromatherapy has been found to be ineffective, was uncertain from the literature.

Evidence of usefulness of aromatherapy for improving in vivo cognitive function was slight, in spite of evidence of numerous essential oil compounds with in vitro anti-cholinesterase properties. Very few of the essential oils identified in the aromatherapy survey had been tested for acetylcholinesterase (AChE) inhibiting properties. This suggested that investigation of the AChE-inhibition of essential oils used in aged care aromatherapy could add novel information to the research area, and potentially discover some novel AChE-inhibitors.

5.1 Research hypothesis

The overall research hypothesis developed out of the investigative phase of the research was that:

‘A blend of essential oils with demonstrated in vitro anti-cholinesterase activity would improve the cognitive function of people with mild to moderate dementia more than either a blend of oils with no in vitro activity or a non-scented placebo control lotion.’

5.2 Research plan

The research plan was therefore split into two parts:

1. Assessment of AChE-inhibition by essential oils identified in the aromatherapy survey
2. Testing of the cognitive effects of the most and least active oils in a clinical trial in residents of aged care facilities with dementia.
5.2.1 Part one: laboratory assessment of essential oils

The first step towards testing the hypothesis was to screen essential oils identified in the survey for *in vitro* cholinesterase inhibiting properties using a modified Ellman method (Savelev et al., 2003). The screening aimed to select oils with highest activity and oils with lowest activity to develop an ‘active’ and an ‘inactive’ blend for the clinical trial. The choice to use blends rather than single essential oils was because blends would not remind participants of particular odours from their past. Odours which remind people of negative experiences could possibly interfere with participants’ acceptance of the treatment, and result in more withdrawals from the trial than if blends with non-identifiable odours were used. The argument that single oils should be used to reduce complexity of the experiment was considered, but as this research was not going to include the next logical steps to identify individual active compounds, it was considered preferable to aim for highest participation rates in the trial.

5.2.2 Part two: clinical trial

It was decided that the two blends of essential oils to be developed during the laboratory testing phase would be tested in a randomised, controlled clinical trial. This trial would be designed to investigate the effect of aromatherapy on the cognitive function of aged care residents with dementia. Considerations arising from the literature review and the survey that informed the design of the clinical trial protocol are discussed below.

5.2.2.1 Preliminary dosage regimen trial

If possible, a dosage regimen trial should be conducted to establish an effective dosage and frequency of application for improvement of cognitive function, but this may not be feasible within the constraints of the research funds or study population. Safety of dermal dosages higher than 5% (v/v) needs to be established (Maddocks-Jennings & Wilkinson, 2004), as skin irritation of some pure essential oils is a known risk factor (Battaglia, 1995). If the safety of doses higher than 5% (v/v) cannot be established, then the trial should use an aromatherapy dosage regimen known to be safe from clinical experience.
For the clinical trial it was decided that aromatherapy lotion should be applied once a day to neck and shoulders area by assistants-in-nursing or personal carers, and that the aromatherapy lotions should be patch-tested on residents prior to the trial to avoid skin irritation.

5.2.2.2 Inclusion and exclusion criteria

Inclusion and exclusion criteria should be considered so as to eliminate possible sources of bias that may affect cognitive function (such as epilepsy, diabetes, recent acute illness and vitamin B12 deficiency). Where possible, participants should be selected according to established guidelines such as diagnosis of dementia (DSM IV or ICD-10) and a baseline measure of cognitive function. If the Mini-Mental State Examination (MMSE) is used, scores should fall within the range of 18-23, although a wider range can be selected if results are sought for a broader range of dementia severities (Tombaugh & McIntyre, 1992). For the clinical trial, it was decided to use the range of 10-26 as this would increase the number of people available to do the trial.

5.2.2.3 Olfactory testing

Olfactory function tests should be designed so that responses of a person with dementia can be unequivocally interpreted (Mesholam et al., 1998). However, it may be unadvisable to exclude people from the trial if they report inability to smell on the day of testing, as there may be other mitigating factors like nasal congestion. Participants should also be asked if they like the smell of the oils before inclusion in the trial, so that risk of non-compliance due to their dislike of the odour is reduced. For the clinical trial it was decided to test olfactory function and whether participants liked the smell of oils.

5.2.2.4 Treatment groups

Each centre in a multi-centre trial should have equal numbers of participants in each treatment group to control for differing environmental factors between facilities. For the clinical trial it was decided that this would be attempted as far as possible, but acknowledged that it might be difficult to get exactly equal numbers of participants from each facility.
5.2.2.5  Blinding

Adequate blinding during dermal or inhaled aromatherapy trials is difficult to achieve, due to the necessary odour of the treatments. Concentrations of dermally-applied essential oils should not be so strong as to be perceptible at normal inter-personal distances unless all participants are on the same treatment.

In multi-centre trials where all trial arms are present in each facility, blinding can possibly be achieved by using ineffective dosages of the same essential oil blend. Using an alternative essential oil blend or synthetic fragrance risks confounding the trial, as most odours cause an hedonic response that may affect mood and consequently release of neurotransmitters (Jellinek, 1997).

Blinding of the researchers carrying out the memory testing can be achieved by an adequate time lapse between dermal application and assessment, so the odour is no longer perceptible at normal interpersonal distances. Alternatively researchers can wear nose-pegs. For the clinical trial it was decided that the researchers should only carry out the memory testing after one hour had elapsed between application of the treatment lotions and testing time.

5.2.2.6  Dosage

Daily massage should be used, as inhalation doses are likely to be too small when vaporised into the air, or placed on the bedclothes, as outlined in the studies. The dosage for dermal application in a lotion should be no more than 3% essential oil (w/v) according to aromatherapy guidelines (Battaglia, 1995). The rationale for this seemingly low dose is to prevent possible dermal irritation by the essential oils. As the trial was to be carried out in aged care facilities with care policies specifying the 3% dosage, it was decided that it would be a good test of aromatherapy as a treatment to keep the dosage within the guidelines. The risk that this dose would be too low for a discernable effect on memory function was outweighed by the concern to minimise risk of dermal irritation in participants, as recommended by the Ethics Committee.
5.2.2.7 Essential oils

Choice of essential oils should be guided by *in vitro* results of acetylcholinesterase inhibition. If time allows, assessment of anti-inflammatory or anti-oxidant properties of the selected oils would be of additional benefit.

5.2.2.8 Outcome measures

Good research practice (as advised by the supervisor of this project) requires that ‘gold-standard’ quantitative primary outcome measures should be selected where possible. Clinical trials for Alzheimer’s disease use the ‘gold-standard’ Alzheimer’s disease Assessment Scale (ADAS-cog) and the Mini-Mental State Examination (MMSE) to measure cognitive function. Behavioural and functional ability scales are often used as secondary measures (Doraiswamy et al., 1997).

5.2.2.9 Treatment design

Treatment design requires attention to avoid unintentional confounding of a study (Schulz et al., 1995). Several of the reviewed studies employed cross-over designs using patients as their own controls. While this has merit in short case studies, it is not appropriate for larger longer-term trials due to the idiopathic nature of degeneration in dementia. A parallel control group of sufficient size would be better for comparing average deterioration over time than a cross-over design.

Participants should all be treated with placebo treatments during a ‘wash-out’ baseline phase, to control for the effects of changes to normal routine, or the ‘Hawthorne’ effect where people improve due to the ‘specialness’ of being in a trial (Opie et al., 2002).

While a repeated measures design requires more complex statistical analysis, repeated measures provide more information about the effects of the treatment during the trial. Ideally, measurements should be taken:

- on entry to the trial;
- after an initial ‘wash-out’ period;
- at appropriate intervals throughout the treatment phase;
- at the end of the treatment phase, and
after a post-treatment ‘wash-out’ phase to observe possible return to baseline as the treatment leaves the system.

Care should be taken in selection of the measurement tool to ensure that the repeated measures are separated far enough apart to avoid any practice effects (Kaufman, 1994). Alternatively, a simpler design of baseline and end-point measures can be undertaken.

5.2.2.10 Randomisation

Randomisation is required to eliminate bias, but care must be taken to randomise the groups after the baseline measurements so as to ensure that the experimental groups are not significantly different at baseline, particularly when assessing cognitive function.

5.2.2.11 Sample size estimates

Sample size estimates should be based on the expected effect size of the treatment. If a small change is expected, larger samples will be required to detect a statistically significant change. It is better to use established and validated outcome measures than invent a new one unless it can be validated prior to the trial. Also, it is beneficial to use at least one, if not two cognitive scales and a behavioural and functional ability assessment tool.
6 Essential oils and cholinesterase inhibition

6.1 Introduction

Aromatherapy use was shown to be wide-spread in the survey of aged care facilities. Although the survey results were equivocal as to whether aromatherapy was perceived to be useful for dementia, it was being used to ameliorate behaviours and symptoms characteristic of dementia. This suggested it would be worth screening the essential oils for activity in tests used for dementia drugs. As cholinesterase inhibition was a common target for dementia drugs, and the literature review identified some essential oils with anti-cholinesterase activity it was chosen as a screening test for the oils to be used in the clinical trial. The twenty-five most used essential oils identified in the survey were screened for their inhibition of acetylcholinesterase prior to selection of the most and least active oils for a trial of aromatherapy for cognitive function improvement in dementia.

6.2 Acetylcholinesterase inhibition assay

The most-utilised method of assessing cholinesterase inhibition for in vitro assays is the Ellman method (Ellman et al., 1961). This method depends on the hydrolysis of acetylthiocholine iodide (ATCI) by acetylcholinesterase (AChE). The hydrolysed thiocholine iodide moiety reacts with the colour-change reagent 5,5'-dithiobis-(2-nitrobenzoic acid) (DTNB) to create a yellow anion with maximum spectrographic absorbance at 412nm. Enzyme inhibitors prevent substrate hydrolysis, thereby preventing formation of the yellow anion. Absorbance values of samples and controls can be compared to determine the extent of enzyme inhibition by the samples.

6.2.1 Method selection

In an attempt to obtain a rapid visual screening test for cholinesterase inhibition, a thin layer chromatography method using 1-naphthyl acetate instead of acetylthiocholine iodide as the enzyme substrate, and Fast Blue B salt instead of DTNB to visualise the inhibition zones (Marston et al., 2002) was modified for use with essential oils. Preliminary assays provided
equivocal results thought to be due to the repulsion of the aqueous enzyme media by the lipophilic essential oils, so the method was not pursued further.

The modified 96-well microplate Ellman method used by Savelev et al. (2003) was then selected as it was the most thoroughly described method, and appeared to produce results with low variability.

However, some concerns arose about the methodological robustness of various aspects of the Ellman assay. The first of these was about the use of solvents for essential oils in the assay. The second was whether freezing of the enzyme would significantly alter results over different testing occasions.

Preliminary methodological assessment to resolve these concerns was carried out prior to screening of the essential oils. The results are discussed below.

6.2.2 Preliminary methodological assessment

6.2.2.1 Solvent selection

Ethanol was used as a solvent for essential oils in previous assays reported by other researchers. Savelev et al. (2003) reported that 86% ethanol was used to prepare inhibitor stock solutions, and had a final well concentration of 2% including controls and blanks. 2% ethanol did not inhibit the enzyme. Miyazawa et al. (1997 and 1998) reported 5% (v/v) ethanol final well concentrations, but did not indicate the extent of enzyme inhibition by ethanol. Other researchers used a polysorbate emulsifier (Gracza, 1985). However, none of the papers address whether the solvent was able to continue holding the essential oils in solution when serially diluted in the aqueous enzyme media, or whether all types of essential oil are dissolved equally well by ethanol. Prior experience with different types of essential oils suggested that the more lipophilic oils may require higher concentrations of ethanol to be held in solution in an aqueous medium.

To address this issue, preliminary solubility trials of six essential oils were carried out. The oils were selected due to their relatively high percentages of different types of compounds. Clove (Syzygium caryophyllatum) contained phenolics, Geranium (Pelargonium graveolens)
contained monoterpenols, Patchouli (*Pogostemon cablin*) contained sesquiterpenoids, Lemongrass (*Cymbopogon citratus*) contained aldehydes, Roman Chamomile (*Anthemis nobilis*) contained esters and Orange (*Citrus sinensis*) contained monoterpenes. The different chemical groups were expected to show differences in solubility, with different concentrations of ethanol.

Different concentrations of ethanol used in the assay method were prepared in 0.1M phosphate buffer at 8.0 pH. The concentrations of ethanol were: 95%, 90%, 80%, and 40%. A 95% concentration means 95 mL EtOH in 5 mL 0.1M phosphate buffer. None of the samples remained completely solubilised in the phosphate buffer pH 8 below 90% ethanol when examined visually.

Serial dilutions were performed on a large scale in 25mL clear glass test-tubes to observe the effects of reducing the ethanol concentration on solubility of the oils. Samples were dissolved in 95% EtOH/buffer, (10µL oil per 990µL 95% EtOH/buffer), and diluted into plain buffer in 10-fold steps. The 1:10 dilutions resulted in essential oils coming out of solution forming globules, forming a coloured layer or sticking to the side of the test-tubes.

Further concerns about the use of ethanol as a solvent arose when the first 96-well plate assays were attempted. Due to the evaporation of ethanol, well volumes did not appear to be consistent, and pipetting during the preparation of dilution series appeared to be inaccurate due to evaporation.

Triton-X100, a non-volatile surfactant was selected in place of ethanol, and by similar visual assessment, it was established that a 5% (w/v) solution of Triton solubilised 5mg/mL of essential oils and kept them in solution after a 1:10 dilution step. The blank-corrected uninhibited enzyme produced a mean maximum slope of $4.34 \times 10^{-4}$ (sd $0.25 \times 10^{-4}$, $n=14$ occasions). The blank-corrected enzyme with 5% Triton produced a mean maximum slope of $37.4 \times 10^{-4}$ (sd $4.5 \times 10^{-4}$, $n=14$ occasions). This indicated that 5% Triton inhibits the enzyme by $13.7\% \pm 11.3\%$, and that inhibition due to Triton must be taken into account when calculating IC$_{50}$ values for samples.
6.2.2.2  **Effects of freezing the enzyme on assay variability**

Minimization of variability associated with preparation of the enzyme was considered to be as important as taking care with pipetting technique. The low enzyme concentrations recommended by Savelev’s method (2003) require very precise measurements of tiny amounts of enzyme for each plate if it is made up fresh every time. To reduce measurement error, 10mL volumes of enzyme solution were made as required and frozen in 1mL aliquots at -18°C, each containing 3.52U/mL of active enzyme.

To determine whether freezing affected activity of the enzyme, activities of fresh, 5 day-old, and 15 day-old enzyme were compared, the latter two having been frozen in 1mL aliquots. For the test, one aliquot of enzyme from each batch was thawed and all the samples of enzyme were diluted in buffer prior to addition to the test plates, yielding the final well concentration of 0.008U/mL as per the report of Savelev et al. (2003). Each plate had 4 replicates of each batch of enzyme, and 3 replicate plates were run within 1 hour of each other using the same enzyme preparations on each plate. Six Sage oil dilutions (1:2) were used to show the effect of the different-aged enzyme batches on a known essential oil inhibitor.

6.2.2.2.1  **Results**

Results for each plate were averaged, and the dilution series of Sage oil gave the dose-dependent curves shown in Figure 6.1.

*Figure 6.1 Dose-dependent enzyme inhibition by Sage oil using 3 different-aged batches of enzyme.*

![% inhibition vs mg/mL](image)

**Error bars = standard error of the mean.**
A two-way ‘repeated measures’ ANOVA carried out using GraphPad 4.0 showed that Enzyme Batch was the main source of variation between the replicates (see Table 6.1). The interaction between Enzyme Batch and Plate # also contributed to the variation, but not as much as Enzyme Batch. There were no significant differences between Plate Number or Replicates.

Table 6.1 Two-way ‘repeated measures’ ANOVA comparing fresh, 5 day old and 15 day old enzyme batches.

<table>
<thead>
<tr>
<th>Source of Variation</th>
<th>Df</th>
<th>Sum-of-squares</th>
<th>Mean square</th>
<th>F</th>
<th>% of total variation</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Interaction (Batch x Plate)</td>
<td>4</td>
<td>5.279E-09</td>
<td>1.32E-09</td>
<td>2.951</td>
<td>21.5</td>
<td>0.0488</td>
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<tr>
<td>Enzyme Batch</td>
<td>2</td>
<td>8.03E-09</td>
<td>4.02E-09</td>
<td>19.3</td>
<td>32.7</td>
<td>0.0006</td>
</tr>
<tr>
<td>Plate #</td>
<td>2</td>
<td>1.324E-09</td>
<td>6.62E-10</td>
<td>1.481</td>
<td>5.4</td>
<td>0.2539</td>
</tr>
<tr>
<td>Replicates (matching)</td>
<td>9</td>
<td>1.872E-09</td>
<td>2.08E-10</td>
<td>0.4651</td>
<td>7.6</td>
<td>0.8793</td>
</tr>
</tbody>
</table>

Bonferroni post-tests for pairs of data were carried out using GraphPad 4.0 to determine which enzyme batch or plate contributed most to the variation (see Table 6.2).

Table 6.2 Bonferroni post-tests for Sage dilution 1 data, comparing replicate plates.

Plate 1 vs. Plate 2

<table>
<thead>
<tr>
<th>Enzyme Batch</th>
<th>Plate 1</th>
<th>Plate 2</th>
<th>Difference</th>
<th>95% CI of diff.</th>
</tr>
</thead>
<tbody>
<tr>
<td>15 day old</td>
<td>0.00007875</td>
<td>0.0000315</td>
<td>-4.7E-05</td>
<td>-0.00009433 to -0.000001676</td>
</tr>
<tr>
<td>5 day old</td>
<td>0.00002025</td>
<td>0.0000315</td>
<td>1.13E-05</td>
<td>-0.00003583 to 0.000005833</td>
</tr>
<tr>
<td>fresh</td>
<td>0.0000125</td>
<td>4.75E-06</td>
<td>-7.8E-06</td>
<td>-0.00005483 to 0.00003933</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Enzyme Batch</th>
<th>Difference</th>
<th>t</th>
<th>P value</th>
<th>Summary</th>
</tr>
</thead>
<tbody>
<tr>
<td>15 day old</td>
<td>-0.00004725</td>
<td>3.16</td>
<td>&lt; 0.05 *</td>
<td></td>
</tr>
<tr>
<td>5 day old</td>
<td>0.00001125</td>
<td>0.7523</td>
<td>&gt; 0.05 NS</td>
<td></td>
</tr>
<tr>
<td>fresh</td>
<td>-0.00000775</td>
<td>0.5183</td>
<td>&gt; 0.05 NS</td>
<td></td>
</tr>
</tbody>
</table>

Plate 1 vs. Plate 3

<table>
<thead>
<tr>
<th>Enzyme Batch</th>
<th>Plate 1</th>
<th>Plate 3</th>
<th>Difference</th>
<th>95% CI of diff.</th>
</tr>
</thead>
<tbody>
<tr>
<td>15 day old</td>
<td>0.00007875</td>
<td>0.000032</td>
<td>-4.7E-05</td>
<td>-0.00009383 to 0.000003324</td>
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<tr>
<td>5 day old</td>
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<td>1.48E-05</td>
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<td>-0.00004433 to 0.00004983</td>
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<th>Difference</th>
<th>t</th>
<th>P value</th>
<th>Summary</th>
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</tr>
<tr>
<td>5 day old</td>
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<td>0.9864</td>
<td>&gt; 0.05 NS</td>
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</tr>
<tr>
<td>fresh</td>
<td>0.00000275</td>
<td>0.1839</td>
<td>&gt; 0.05 NS</td>
<td></td>
</tr>
</tbody>
</table>

Plate 2 vs. Plate 3

<table>
<thead>
<tr>
<th>Enzyme Batch</th>
<th>Plate 2</th>
<th>Plate 3</th>
<th>Difference</th>
<th>95% CI of diff.</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Enzyme Batch</th>
<th>Difference</th>
<th>t</th>
<th>P value</th>
<th>Summary</th>
</tr>
</thead>
<tbody>
<tr>
<td>15 day old</td>
<td>-0.00004675</td>
<td>3.126</td>
<td>&lt; 0.05 *</td>
<td></td>
</tr>
<tr>
<td>5 day old</td>
<td>0.00001475</td>
<td>0.9864</td>
<td>&gt; 0.05 NS</td>
<td></td>
</tr>
<tr>
<td>fresh</td>
<td>0.00000275</td>
<td>0.1839</td>
<td>&gt; 0.05 NS</td>
<td></td>
</tr>
</tbody>
</table>
These tests revealed that 15 day-old enzyme in Plate 1 was significantly different from both Plates 2 and 3, $p<0.05$ on both occasions, so it appeared that 15 day-old enzyme in Plate 1 was the main source of variation in the experiment. However, as none of the enzyme batches on Plates 2 and 3 were significantly different from each other, the aberrant results for 15 day-old enzyme in Plate 1 were more likely due to methodological error rather than to enzyme age or freezing status.

6.2.2.2 Conclusions

From these results, it was concluded that aliquots of bovine acetylcholinesterase enzyme can be frozen for up to 15 days without affecting assay reproducibility. This allowed for the preparation of larger amounts of enzyme at a time, therefore reducing variability due to error of measurement.
6.2.3 Essential oil assay

6.2.3.1 Aims

The primary aim of the assay was to discover whether any of the essential oils used in aged care facilities exhibited dose-dependent inhibition of bovine acetylcholinesterase *in vitro*. Where applicable, the results were to be compared with reported literature values for the same oils.

The second aim was to determine from the data those oils which would be most likely, and those least likely to improve cognitive function by inhibition of cholinesterase. Based on this information, it was intended to develop two blends that could be used in a clinical trial, a potentially ‘active’ and a potentially ‘inactive’ blend.

6.2.3.2 Methods

From results of the preliminary methodological assessments, the method of Savelev et al. (2003) was modified as follows. Firstly, 5% Triton-X100, a non-ionic surfactant was used in the place of ethanol. Secondly, the enzyme was prepared in bulk and frozen in 1mL aliquots at -18°C. As the laboratory only had a 37°C incubator, the incubation time was reduced from 30 minutes to 15 minutes and absorbance measured for 15 minutes rather than 6 minutes. The available spectrophotometer only had a 405nm filter, rather than a 412nm filter but it was found that there was sufficient absorption at 405nm to carry out the test.

6.2.3.3 Materials

*Buffer:* Phosphate buffer (0.1M, pH 8.0 at 20°C) was used to prepare all materials, including the 5% Triton-X100 solvent (5g Triton/95mL buffer). Phosphate buffer (0.1M, pH 7.0 at 20°C) was used to solubilise DTNB and ATCI.

*Enzyme Stock:* Ten 1.0mL aliquots of lyophilised bovine erythrocyte enzyme, 0.31 units of enzyme activity/mg solid (Sigma C5021, Sydney, Australia) were sonicated for 20 seconds in phosphate buffer pH8 as per the instructions on the container. Each aliquot contained approximately 3.52U/mL. These were frozen at -18°C until required. Working enzyme was
prepared by thawing one aliquot and diluting with 9mL of 0.1M phosphate buffer pH 8.0. When 5µL of this diluted enzyme was added to test plates, it gave a final well concentration (final volume = 220µL) of enzyme of 0.008U/mL as used by Savelev et al. (2003).

Acetylthiocholine iodide (ATCI) (Sigma A5751, Sydney, Australia). A 0.022M solution of ATCI was prepared in phosphate buffer pH 7.0 to give final well concentration of 0.47mM. As ATCI undergoes non-enzymatic hydrolysis in light and heat, 5mL amounts were prepared and stored in a brown glass bottle at 4° C.

5,5-dithiobis-(2-nitrobenzoic acid) (DTNB) (Sigma D8130, Sydney, Australia). A 0.0132M solution of DTNB was prepared in phosphate buffer pH 7.0 to give a final well concentration of 0.3mM. DTNB also is unstable in light and heat, so 5mL amounts were prepared at a time and stored in a brown glass bottle at 4° C.

Galantamine hydrobromide from Lycoris sp. (Sigma G1660, Sydney, Australia). A stock solution (1 mg/mL) of galantamine, a known enzyme inhibitor and Alzheimer’s disease drug, was prepared in 5% Triton-X100, and serially diluted in 5% Triton-X100 as required. Galantamine was used to compare the modified method with literature values. It was too expensive for use as a positive standard, and also water-soluble, so the monoterpenoid compound 1,8-cineole was used instead as a positive standard.

Stock essential oils: 10µL of each essential oil was weighed (9.8µg +/-0.2µg) and added to 1990µL of 5% Triton-X100 (0.1 phosphate buffer, pH 8.0) in a screw-cap clear glass vial to yield a 0.01mg ±0.002 mg/mL stock solution. Mixtures were sonicated for 20 seconds and vortexed for 10 seconds to ensure complete solubilisation of the oils (non-turbid solutions).

The following oils were tested: Basil (Ocimum basilicum), Bergamot (Citrus bergamia), Black Pepper (Piper nigrum), Clary Sage (Salvia sclarea), Cypress (Cupressus sempervirens), Eucalyptus (Eucalyptus globulus), Frankincense (Boswellia carterii), Geranium (Pelargonium graveolens), German Chamomile (Matricaria recutita), Ginger (Zingiber officinalis), Grapefruit (Citrus paradisi), Juniper berry (Juniperus communis), French and Tasmanian Lavender (both Lavandula angustifolia), Lavender Sage (Salvia lavandulaefolia), Lemon (Citrus limonum), Lemongrass (Cymbopogon citratus), Lime (Citrus latifolia), Mandarin (Citrus reticulata), Peppermint (Mentha x piperita), Roman Chamomile
(Anthemis nobilis), Rosemary (Rosmarinus officinalis), Sage (Salvia officinalis), Sweet Marjoram (Origanum marjorana), Tea Tree (Melaleuca alternifolia).

The three major components of each essential oil were determined by GC-MS and appear in Table 6.5 in the Discussion section of this chapter. GC-MS specifications appear in Appendix 2.

Positive standard: 1,8-cineole (Sigma C80601) was used as a positive standard on each plate, and was prepared in the same way as the essential oils.

Equipment: A Wallac Victor 2 96-well plate spectrophotometer was used, controlled by WorkOut software (version 2). Absorbance at 405nm and 37° C was measured over 15 minutes, with each well measured for 0.1 seconds. Automated shaking for 10 seconds was carried out prior to commencement with 1 second automated shaking between each run. Clear plastic Spectra U-bottomed 96 well plates (Perkin Elmer, Roweville, Victoria) were used, and absorbance was measured without lids as the condensation on the lids interfered with the absorbance readings.

6.2.3.3.1 Preparation of dilution series

140 µL of essential oil samples and 1,8-cineole positive control were added to 140 µL of 5% Triton solution and then serially diluted 5 times with 5% Triton solution to achieve a six-fold 1 in 2 dilution series. The dilution series therefore contained final concentrations of approximately 5% Triton-X100. Based on the results of the preliminary investigations with Sage oil, it was assumed that the small increases of Triton-X100 in the dilutions would not significantly contribute to variation in the assay results. When 20 µL of sample was transferred from the dilution plate to the test plate, the final well concentrations for the samples started at 0.454 mg/mL.

6.2.3.3.2 Addition of solutions to test plate

Solutions were added to sample and 1,8-cineole standard wells in the test plate as follows: 5 µL enzyme, 20 µL sample/standard from dilution plate, 5 µL DTNB, 185 µL phosphate buffer. The plates were shaken with lids on for 1 minute on an orbital shaker and then incubated for 15 minutes at 37° C. After incubation, 5 µL of ATCI was added to each well and
plates were immediately transferred without their lids to the spectrophotometer maintained at 37°C throughout the assay. The final volume in each well was 220µL. Figure 6.2 shows the different quantities of reagents used for the different wells. As the solvent was shown in the preliminary work to interfere with the assay, but with a large amount of variability from plate to plate, it was decided to include two controls and two blanks, with and without solvent. The presence of control and blank without solvent would allow for calculation of the amount that the solvent inhibited the enzyme and the non-enzymic hydrolysis of the substrate in the blank.

Figure 6.2 Volume (µL) of reagents added to the test plate.

<table>
<thead>
<tr>
<th>Order of addition</th>
<th>Control</th>
<th>Blank</th>
<th>Blank + Solvent</th>
<th>+ Control Solvent</th>
<th>+ 1,8-cineole standard</th>
<th>Sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enzyme</td>
<td>5</td>
<td>-</td>
<td>-</td>
<td>5</td>
<td>5</td>
<td>5</td>
</tr>
<tr>
<td>Std/sample</td>
<td>-</td>
<td>-</td>
<td>20 solvent</td>
<td>20 solvent</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>DTNB</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td>5</td>
<td>5</td>
</tr>
<tr>
<td>Buffer</td>
<td>205</td>
<td>210</td>
<td>190</td>
<td>185</td>
<td>185</td>
<td>185</td>
</tr>
</tbody>
</table>

Shake on orbital plate shaker for 1 minute with lid on
Transfer to 37°C incubator in closed box for 15 minute incubation
ATCI 5 5 5 5 5 5

Place in spectrophotometer and take absorbance readings without lid

Each test plate had 6 replicate wells for control, blank, control + 20µL solvent, blank + 20µL solvent, 2 or 3 replicate wells for 6 serial dilutions of 1,8-cineole as a positive standard, and 2 or 3 replicate wells for 6 serial dilutions of essential oil sample. Three or four essential oils were tested on each plate.

6.2.3.3.3 Measurements

The Absorbance/time curves for each sample were calculated by WorkOut, the software program supplied with the spectrophotometer. The WorkOut software also automatically calculated maximum rate of absorbance change for different sample concentrations, averaging the slopes of the curve at 5 points. This was suggested by the software help menu to provide a good estimate of maximum rate of change of absorbance.

The maximum rate values were then transferred to Excel 5.0 (Microsoft), averaged, blank-subtracted and compared to controls to create dose-response curves and determine the inhibitory activities of the oils. The blank-subtraction step was necessary due to non-enzymic hydrolysis of the substrate at pH 8.0, and because the solvent inhibited the non-enzymic hydrolysis and the enzyme to a significant extent.
6.2.3.3.4 Replication

Each sample was replicated three times on each plate, and the controls and blanks were replicated six times. Each plate was replicated a minimum of two times, using the same aliquot of enzyme for replicate plates.

6.2.3.4 Results

6.2.3.4.1 Rate of non-enzymic hydrolysis

To ensure that the assay was working correctly, the rate of non-enzymic hydrolysis in the blank wells without solvent was calculated and compared with the published results by Ellman et al. (1961). Using mean values from twelve experiments, the mean rate (maximum slope) of non-enzymic hydrolysis of acetylthiocholine at 37°C (i.e. the blank) was equivalent to 0.0015 (sd 0.0003) Abs/min. Ellman et al. (1961), reported a rate of 0.0016 absorbance units per minute at 25°C, also indicating that the assay temperature did not appear to significantly affect the rate of non-enzymic hydrolysis.

The mean rate of non-enzymic hydrolysis of the substrate in blanks containing 5μL of 5% Triton was equivalent to 0.00032 (sd 0.00003) Abs/minute, 5 times less than the blank without solvent. The fact that 5% Triton significantly reduced the non-enzymatic hydrolysis reinforced the need to have blank and control wells with 5% Triton on every plate.

6.2.3.4.2 Solvent inhibition of enzyme

As 5% Triton inhibited the enzyme to some degree, the blank and control wells contained the same concentration of solvent as the sample wells. The extent of solvent inhibition was calculated using the following equation:

\[
\text{Inhibition due to Triton} = \frac{((C-BL)-(CT-BLT))}{(C-B)}
\]

where \( C \) = Control (containing enzyme but no sample), \( BL \) = blank (containing no enzyme and no sample), \( CT \) = Control with 5% Triton and \( BLT \) = Blank with 5% Triton. This method of accounting for solvent inhibition was used for all the following results.
Absorbance/time curves were produced for all samples, controls and blanks, and the maximum slopes of these lines was taken to be the measure of inhibition of the enzyme. When percentage inhibition of controls was plotted against inhibitor concentration (IC), several essential oil samples exhibited dose-dependent inhibition. Mean percentage inhibitions at each dosage were calculated for each sample. The standard deviation between replicates and between plates was less than 15%. As natural log equations generated using Microsoft Excel 2000 appeared to best approximate the curves, IC$_{50}$ values for the oils displaying dose-dependent inhibition of the enzyme were calculated from the natural log equations.

Oils showing dose-dependent inhibition of the enzyme were (in order from most to least active): Lime (*Citrus latifolia*), Sage (*Salvia officinalis*), Rosemary (*Rosmarinus officinalis*), Eucalyptus (*Eucalyptus globulus*), Lavender Sage (*Salvia lavandulaefolia*), Lemon (*Citrus limonum*), Sweet Marjoram (*Origanum marjorana*), Tea tree (*Melaleuca alternifolia*) and Black Pepper (*Piper nigrum*). Cypress (*Cupressus sempervirens*) and Frankincense (*Boswellia carterii*) showed linear dose-dependent inhibition. The rest of the oils did not display dose-dependent inhibition.

A summary of the percentage inhibitions at 0.454mg/mL (approximately 2.9mM if calculated on an average molecular weight for monoterpenoid compounds of 154) is shown in Table 6.3. Results of individual assays are shown in Appendix 2, Table 2.

<table>
<thead>
<tr>
<th>Oil name</th>
<th>N plates</th>
<th>Av. % inhibition at 0.454 mg/mL</th>
<th>Range of % inhibition at 0.454 mg/mL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lime</td>
<td>5</td>
<td>90.9</td>
<td>62.2 to 105.1</td>
</tr>
<tr>
<td>Blend 1 (Cypress, Euc, Lime)</td>
<td>3</td>
<td>82.9</td>
<td>69.9 to 91.3</td>
</tr>
<tr>
<td>Galantamine*</td>
<td>3</td>
<td>79.5</td>
<td>71.1 to 86.5</td>
</tr>
<tr>
<td>Sage</td>
<td>8</td>
<td>63.9</td>
<td>51.8 to 86.7</td>
</tr>
<tr>
<td>1,8-cineole</td>
<td>41</td>
<td>54.1</td>
<td>33.7 to 75.1</td>
</tr>
<tr>
<td>Cypress</td>
<td>5</td>
<td>52.9</td>
<td>35.7 to 62.9</td>
</tr>
<tr>
<td>Rosemary</td>
<td>3</td>
<td>51.9</td>
<td>46.9 to 55.6</td>
</tr>
<tr>
<td>Eucalyptus globulus</td>
<td>3</td>
<td>46.8</td>
<td>45.4 to 49.6</td>
</tr>
<tr>
<td>Lavender Sage (ABP)</td>
<td>2</td>
<td>45.8</td>
<td>44.6 to 46.9</td>
</tr>
<tr>
<td>Lavender Sage (Baldwins)</td>
<td>8</td>
<td>43.1</td>
<td>29.6 to 55.7</td>
</tr>
<tr>
<td>Sweet Marjoram</td>
<td>3</td>
<td>33.5</td>
<td>31.3 to 35.8</td>
</tr>
</tbody>
</table>
Oil name | N plates | Av. % inhibition at 0.454 mg/mL | Range of % inhibition at 0.454 mg/mL
--- | --- | --- | ---
Tea Tree | 5 | 30.2 | 27.8 to 33.4
Lemon | 6 | 29.2 | 19.2 to 44.5
Black Pepper | 3 | 25.0 linear | 24.6 to 25.9
Frankincense | 5 | 25.8 linear | 8.3 to 35.7
Lavender Tasmanian | 6 | 25.4 NDD | 14.9 to 36.0
Lavender French | 6 | 25.3 NDD | 17.8 to 40.0
Roman Chamomile | 5 | 20.0 NDD | 15.1 to 24.7
Juniper berry | 3 | 19.2 NDD | 17.8 to 20.6
Blend 2 (Mandarin, Ginger, Lemongrass) | 2 | 18.7 NDD | 17.3 to 20.7
German Chamomile | 6 | 16.4 NDD | 2.2 to 36.0
Bergamot | 3 | 12.3 NDD | 5.7 to 19.4
Grapefruit | 6 | 8.4 NDD | -0.5 to 24.2
Clary Sage | 3 | 3.1 NDD | 1.7 to 3.8
Basil | 6 | 1.9 NDD | -15.5 to 21.8
Geranium | 3 | -2.8 NDD | -3.1 to -2.7
Lemongrass | 6 | -2.9 NDD | -7.6 to 0
Mandarin | 6 | -4.5 NDD | -11.9 to 8.0
Ginger | 5 | -7.4 NDD | -16.8 to 9.7

*Galantamine final well concentration was 0.98mM, and for essential oils and 1,8-cineole it was 2.9mM. NDD = no dose-dependent change from serial dilutions.

The variability between assays done on different days was determined by comparing the range of results for 1,8-cineole (included as a positive standard on all plates). The average percentage inhibition for 2.9mM 1,8-cineole ranged from 33.7% to 75.1%. Figure 6.3 displays the means and standard errors for cineole, with the dotted line indicating when a new batch of 5% Triton was used. On each occasion three plates were used with the same enzyme so as to provide adequate replication of the samples.

Figure 6.3 Cineole replicates.
There was a significant difference (two way t-test, \( t = -2.880, p=0.005 \)) between the mean maximum slopes for 2.9mM 1,8-cineole prepared with old and new batches of 5% Triton (means were 0.000148 and 0.000164 respectively). This finding explained the variability between replicates of essential oils done with old or new Triton. However, the order of potency of the oils showing dose-dependent inhibition was the same in both batches of Triton.

### 6.2.3.4.4 Selection of essential oils for clinical trial blends

The primary aim of the laboratory work was to select essential oils for two aromatherapy blends for a clinical trial. Blend 1 was formulated to contain equal proportions by weight of Lime, Cypress and Eucalyptus oils (the ‘active’ blend, labelled Treatment A in the clinical trial). These oils were chosen because they showed the greatest AChE inhibitory activity in the laboratory tests, and also formed a pleasant odour when blended. The Sage oils were not used because aromatherapists in Australia do not recommend the use of Sage oil, and the Ethics Committee could have refused to approve the clinical trial if Sage oil had been used. Blend 2 contained equal proportions by weight of Mandarin, Lemongrass and Ginger (the ‘inactive’ blend, Treatment B in the clinical trial). These oils did not show any dose-dependent inhibition of the enzyme, and also formed a pleasant odour when combined. Figure 6.4 displays the results for the two blends, with 1,8-cineole as a standard.

**Figure 6.4 Inhibitory effect of two essential oil blends and 1,8-cineole standard on AChE activity compared to control (repeats n=3, error bars = standard error).**
The shape of the dose-response curves of Blend 1 and 1,8-cineole suggest dose-dependent activity of the inhibitors. Using the best-fitting natural logarithm equations generated by Microsoft Excel 2000, the IC₅₀ values for Blend 1 and 1,8-cineole were 0.0938mg/mL, and 1.814mg/mL respectively. Blend 2 did not show dose-dependent inhibition though the maximum inhibitory value at 0.454 mg/mL was 21% that of controls. It was decided that these two blends would be sufficiently different in likely AChE inhibitory activity for the clinical trial, and that added benefit from testing other mixtures of essential oils would be slight.

6.2.3.4.5 Comparison with literature values

The comparison of the results found during this project and those reported in the literature are shown in Table 6.4 and summarised below.

Table 6.4 Comparison of research (bovine erythrocyte enzyme) with literature values (human brain and bovine erythrocyte enzyme).

<table>
<thead>
<tr>
<th>Sample</th>
<th>Literature results</th>
<th>This research (bovine erythrocyte)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>IC₅₀ mg/mL</td>
<td>% inhibition (sd) at 0.09 mg/mL</td>
</tr>
<tr>
<td>Lime (expressed)</td>
<td>0.029</td>
<td>66% (3.6)</td>
</tr>
<tr>
<td>Lavender Sage (S. lavandulaefolia)</td>
<td>0.07</td>
<td>63% (3.7)</td>
</tr>
<tr>
<td>Sage (S. officinalis)</td>
<td>0.03</td>
<td>-</td>
</tr>
<tr>
<td>Clary Sage</td>
<td>0.05</td>
<td>52% (0.8)</td>
</tr>
<tr>
<td>Rosemary</td>
<td>0.07</td>
<td>21% (5.7)</td>
</tr>
<tr>
<td>Lavender</td>
<td>0.07</td>
<td>17% (11.2)</td>
</tr>
<tr>
<td>Lemon (expressed)</td>
<td>0.04</td>
<td>0.0%</td>
</tr>
<tr>
<td>Lemongrass</td>
<td>0.06</td>
<td>0.0%</td>
</tr>
<tr>
<td>Cineole</td>
<td>0.06</td>
<td>0%</td>
</tr>
<tr>
<td>Galantamine</td>
<td>5.0x10⁻⁷ M</td>
<td></td>
</tr>
</tbody>
</table>

* Perry (2000) used Lavender Sage oil with 30% camphor, 15% 1,8-cineole, and 15% bornyl acetate.
** Savelev used Lavender Sage oil with 26.8% 1,8-cineole, 24.7% camphor, 6.6% alpha-pinene.
@Lavender Sage oil used in this research had 28% 1,8-cineole, 27% camphor and 6% camphene.

Savelev et al. (2003) reported an IC₅₀ of 0.05 mg/mL for Salvia lavandulaefolia. The two different brands of Salvia lavandulaefolia oil tested in this study had IC₅₀ values of 0.908 and 0.608 mg/mL. Savelev et al. (2003) also reported an IC₅₀ value of 0.06 mg/mL for 1,8-cineole
and Miyazawa et al. (1998) reported 0.04mg/mL, whereas IC$_{50}$ values for 1,8-cineole obtained in this study ranged from 0.04-1.34 mg/mL.

The difference in values for the *Salvia lavandulaefolia* oils suggests that the 5% Triton solvent may have interfered with the active compound or compounds in the oils. However, the wide range of results for 1,8-cineole in the current study as compared to the literature values suggests that the modified experimental method (using 5% Triton as a solvent) did not deliver precise results. This is partly accounted for by the differences between old and new batches of solvent, but the range of values is still wide between different plates and between different experiments.

Although it is questionable whether results from bovine erythrocyte enzyme and human brain enzyme can be compared, Perry et al. (1996) reported the IC$_{50}$ of Lime oil to be 0.029 mg/mL and it was found to be 0.038 mg/mL in this study. Sage and Rosemary oils were also shown to have inhibitory activity at 0.09 mg/mL against human brain enzyme by Perry et al. (1996) at 52% and 17% respectively. This research showed Sage and Rosemary oils had 43% and 30% inhibition at 0.11 mg/mL respectively. Surprisingly, Perry et al. (1996) did not find 1,8-cineole to show any inhibition, even at 0.723mg/mL (4.7mM). However, because a different solvent system was used in the current study, comparisons are probably of little use other than to note that the Lime, Sage and Rosemary oils still rank among the more potent inhibitors of the AChE enzyme, as found in other studies.

Perry et al. (1996) report a galantamine IC$_{50}$ value of 5.0 x 10$^{-7}$ M, whereas this study found an IC$_{50}$ 5.9 x 10$^{-5}$M, which is 100-fold less potent. Unfortunately, galantamine was not tested by Savelev et al. (2003), or Miyazawa et al. (1998) who used the bovine erythrocyte enzyme. Galantamine was not chosen as a standard for the current study as it does not have a terpenoid structure, and therefore could have completely different interactions with the Triton solvent.

6.2.4 Discussion

The primary aim of the current study was to investigate whether there was dose-dependent inhibition of bovine acetylcholinesterase by any of the essential oils used in aged care facilities. While several of the essential oils studied appeared to show dose-dependent inhibition of the AChE enzyme, there were discrepancies between the results found here, and those reported by other researchers. Further work could have been carried out to investigate
this, but as the main aim of the research was to find out which oils exhibited AChE inhibitory activity for the clinical trial, not to replicate previous work, the replication studies were not carried out. Repetition of the work of Savelev et al., (2003) could have been done to see whether their results could have been replicated with the different spectrometer and incubation conditions.

Future research in this area should include comparisons of human and bovine enzyme, with a positive essential oil control and galantamine control on each plate. Other solvents could be tested, including a head-to-head comparison between ethanol and non-volatile surfactants such as Triton X-100 and Tween 20 or Tween 80. To further identify possible sources of experimental error, it would be useful to have two researchers run the experiments in tandem in the same laboratory.

The chemical compositions of the essential oils revealed that some essential oils would have been expected to inhibit the enzyme due to their high content of the known inhibitors. Table 6.5 shows the GC-MS results for the three major components of essential oils used in this research. Grey shaded cells are compounds that exhibited AChE-inhibition at less than 1.0mM in previous reports (see references in Table 1, Appendix 2). The order of potency found in the literature from the most to least potent of the shaded compounds, was: Delta-3-carene > 1,8-cineole > alpha-pinene > alpha-terpinene (see Table 1, Appendix 2). Compounds that exhibited inhibition at greater than 1.0mM were not considered to be sufficiently potent to contribute significantly to an essential oil’s overall activity (synergy aside). For example, beta-pinene reached 50% inhibition at 1.47 mM, but limonene and camphor did not reach 50% inhibition even at 2.0mM.
Table 6.5 Essential oils used in aged care, showing top three major constituents as analysed by GC-MS.

<table>
<thead>
<tr>
<th>Oil/compound</th>
<th>Botanic name</th>
<th>Compound 1</th>
<th>Compound 2</th>
<th>Compound 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>1,8-cineole</td>
<td>(Sigma C80601)</td>
<td>1,8-cineole 100%</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Basil</td>
<td>Ocimum basilicum</td>
<td>Estragole 83%</td>
<td>Linalool 15%</td>
<td>Alpha-bisabolene 1.5%</td>
</tr>
<tr>
<td>Bergamot</td>
<td>Citrus bergamia</td>
<td>Linalyl acetate 46%</td>
<td>Limonene 25%</td>
<td>Linalool 23%</td>
</tr>
<tr>
<td>Black Pepper</td>
<td>Piper nigrum</td>
<td>Beta-caryophyllene 31%</td>
<td>Limonene 13%</td>
<td>Sabinene 10%</td>
</tr>
<tr>
<td>Clary Sage</td>
<td>Salvia scabra</td>
<td>Linalyl acetate 64%</td>
<td>Linalool 22%</td>
<td>Alpha-terpineol 6%</td>
</tr>
<tr>
<td>Cypress</td>
<td>Cupressus sempervirens</td>
<td>Alpha-pinene 60%</td>
<td>Delta-3-carene 19%</td>
<td>Limonene 5%</td>
</tr>
<tr>
<td>Eucalyptus</td>
<td>Eucalyptus globulus</td>
<td>1,8-cineole 76%</td>
<td>Alpha-pinene 14%</td>
<td>Aromadendrene 3%</td>
</tr>
<tr>
<td>Frankincense</td>
<td>Boswellia carterii</td>
<td>Alpha-pinene 87%</td>
<td>Limonene 5%</td>
<td>Delta-3-carene 2%</td>
</tr>
<tr>
<td>Geranium</td>
<td>Pelargonium graveolens</td>
<td>Citronellol 32%</td>
<td>Geraniol 16%</td>
<td>Citronellyl formate 15%</td>
</tr>
<tr>
<td>German Chamomile</td>
<td>Matricaria recutita</td>
<td>Trans-beta farnesene 44%</td>
<td>Alpha-bisabolol 28%</td>
<td>Chamazulene 8%</td>
</tr>
<tr>
<td>Ginger</td>
<td>Zingiber officinalis</td>
<td>Alpha-zingiberene 46%</td>
<td>Beta-sesquiphellandrene 11%</td>
<td>Ar-curcumene 8%</td>
</tr>
<tr>
<td>Grapefruit</td>
<td>Citrus paradisi</td>
<td>Limonene 97%</td>
<td>Beta-pinene 2%</td>
<td>Para-cymene 1%</td>
</tr>
<tr>
<td>Juniper Berry</td>
<td>Juniperus communis</td>
<td>Alpha-pinene 54%</td>
<td>Beta-pinene 34%</td>
<td>Limonene 6%</td>
</tr>
<tr>
<td>Lavender French</td>
<td>Lavandula angustifolia</td>
<td>Linalyl acetate 43%</td>
<td>Linalool 37%</td>
<td>Beta-caryophyllene 6%</td>
</tr>
<tr>
<td>Lavender Sage</td>
<td>Salvia lavandulaefolia</td>
<td>Camphor 29%*</td>
<td>1,8-cineole 29%</td>
<td>Alpha-terpinene 7%</td>
</tr>
<tr>
<td>Lavender (Baldwins)</td>
<td>Salvia lavandulaefolia</td>
<td>1,8-cineole 28%</td>
<td>Camphor 27%*</td>
<td>Camphene 6%</td>
</tr>
<tr>
<td>Lavender Tasmanian</td>
<td>Lavandula angustifolia</td>
<td>Linalyl acetate 42%</td>
<td>Linalool 40%</td>
<td>Trans-beta-ocimene 4%</td>
</tr>
<tr>
<td>Lemon</td>
<td>Citrus limonum</td>
<td>Limonene 76%</td>
<td>Beta-pinene 11%</td>
<td>Para-cymene 8%</td>
</tr>
<tr>
<td>Lemongrass</td>
<td>Cymbopogon citratus</td>
<td>Geraniol 51%</td>
<td>Neral 37%</td>
<td>Geranyl acetate 6%</td>
</tr>
<tr>
<td>Lime</td>
<td>Citrus latifolia</td>
<td>Limonene 54%</td>
<td>Gamma-terpinene 14%</td>
<td>Terpinolene 13%</td>
</tr>
<tr>
<td>Mandarin</td>
<td>Citrus reticulata</td>
<td>Limonene 76%</td>
<td>Gamma-terpinene 17%</td>
<td>Para-cymene 2%</td>
</tr>
<tr>
<td>Peppermint Australian</td>
<td>Mentha x piperita</td>
<td>l-menthol 47%</td>
<td>l-menthone 23%</td>
<td>Isomenthone 6%</td>
</tr>
<tr>
<td>Roman Chamomile</td>
<td>Anthemis nobilis</td>
<td>Isobutyl 18%</td>
<td>Angelate ester 17%</td>
<td>Angelate ester 13%</td>
</tr>
<tr>
<td>Rosemary</td>
<td>Rosmarinus offcinalis</td>
<td>1,8-cineole 57%</td>
<td>Alpha-pinene 13%</td>
<td>Camphor 9%*</td>
</tr>
<tr>
<td>Sage</td>
<td>Salvia officinalis</td>
<td>Camphor 35%*</td>
<td>1,8-cineole 29%</td>
<td>Alpha-pinene 7%</td>
</tr>
<tr>
<td>Sweet Marjoram</td>
<td>Origanum marjorana</td>
<td>Terpinen-4-ol 23%</td>
<td>Cis-sabinenene 19%</td>
<td>Gamma-terpinene 14%</td>
</tr>
<tr>
<td>Tea Tree</td>
<td>Melaleuca alternifolia</td>
<td>Terpinen-4-ol 40%</td>
<td>Gamma-terpinene 20%</td>
<td>Alpha-terpinene 9%</td>
</tr>
</tbody>
</table>

* Camphor has been demonstrated to reduce the efficacy of 1,8-cineole (Savelev et al, 2003). Grey cells are compounds with known IC50 values of less than 1.0mM.

Several oils contained high percentages of alpha-pinene, 1,8-cineole, delta-3-carene and alpha-terpinene, which had been identified as having IC50 values less than or equal to 1.0mM (Table 1, Appendix 2). Based on the synergy work by Savelev et al. (2003), it could be further expected that oils such as Eucalyptus with both alpha-pinene and 1,8-cineole would be more...
potent inhibitors than those with only one of these compounds. Similarly, oils that contained both 1,8-cineole and camphor such as Rosemary, Sage and Lavender Sage were expected to be less effective inhibitors due to the observed antagonism (Savelev et al. 2003). Although delta-3-carene was not tested for synergy or antagonism by Savelev and colleagues it was the one of the most potent terpenoid inhibitors reported in the literature, with an IC$_{50}$ of 0.20mM (Miyazawa and Yamafuji, 2005), and therefore Cypress oil with alpha-pinene and delta-3-carene was expected to be about as potent as Eucalyptus. The expected order of potency for the oils assayed based on these assumptions was therefore:

- Eucalyptus, Cypress >
- Frankincense, Juniper (due to alpha-pinene content) >
- Rosemary, Sage and Lavender Sage (due to camphor inhibition of 1,8-cineole) >
- Tea Tree (due to alpha-terpinene content) >
- remainder of the oils.

The actual order of potency of the oils assayed was:

- Lime >
- Sage, Cypress, Rosemary, Eucalyptus, Lavender Sage, Sweet Marjoram >
- Tea Tree, Lemon, Black Pepper, Frankincense.

The rest of the oils, including Juniper, showed low inhibition that was not dose-dependent.

Given their major constituents, the positions of Lime and Sage were higher than expected (in the case of Lime, much higher), whereas Frankincense and Juniper were lower than expected. This could have been due to inhibition of the alpha-pinene by minor constituents of each oil. Sweet Marjoram, Lemon and Black Pepper displayed unexpected inhibition as they did not contain any of the top four constituents, although the inhibition was not as powerful as the 1,8-cineole containing oils.

These results suggest that there may be hitherto undiscovered inhibitor compounds or synergism in both Lime and Sage oil. (+)-Limonene, the major component of Lime oil was
shown to have only very weak inhibition (Miyazawa et al. 1997). The fact that both Mandarin and Lemon oils contain high percentages of limonene, but were not strong inhibitors, also suggests that limonene is not responsible for the strong activity of Lime oil.

It is possible that coumarins present in Lime oil contribute to the inhibition, as auraptene, a coumarin from Grapefruit oil showed 17-24% inhibition of controls at 1.62µg/mL (Miyazawa et al., 2001). Coumarins from Korean Angelica (Angelica gigas, an oil not used in aromatherapy) also showed inhibition ranging between 0.028-0.067mM, although the enzyme assay method may not be comparable as it was not described in the report (Kang et al., 2001).

Perry et al. (1996) suggested that perhaps the Lime oil activity they observed could be due to organophosphate pesticides, but if this was the case, then other citrus peel oils like Lemon, Mandarin and Grapefruit would have also been likely to show the same inhibition. Activity-guided fractionation of Lime and Sage oils could lead to the discovery of potent, novel acetylcholinesterase inhibitors.

The second aim was to determine which oils would be most and least likely to show activity in a clinical aromatherapy trial and to test two blends of oils that would also be acceptable in terms of odour and cost. Two blends were created and tested alongside 1,8-cineole and galantamine. Blend 1 contained Cypress, Lime and Eucalyptus oils, and exhibited inhibitory activity at 0.454 mg/mL to the same degree as the individual oils. Blend 2 contained Mandarin, Lemongrass and Ginger oils, and exhibited very little inhibitory activity at 0.454 mg/mL similar to that of the individual oils. Both blends also smelled acceptable to laboratory colleagues and co-workers, and were reasonably priced.

6.2.5 Conclusions

These results show that some essential oils used in aged care aromatherapy inhibit bovine acetylcholinesterase but at relatively high concentrations, compared to galantamine. There is some suggestion that essential oils affect human brain enzyme and bovine erythrocyte enzyme
similarly, but further assessment of essential oils with human brain enzyme is required to establish this conclusively. Two selected essential oil blends were considered likely to show a difference in therapeutic effects in a clinical trial of aromatherapy for cognitive function in dementia, based on the differences in their \textit{in vitro} inhibitory activity against the acetylcholinesterase enzyme.
7 Investigating cognitive effects of aromatherapy on people with dementia living in residential care facilities

7.1 Clinical Study Methods

7.1.1 Introduction

The aim of this clinical study was to investigate cognitive effects of an essential oil blend with known \textit{in vitro} acetylcholinesterase activity on people with dementia living in residential aged care facilities.

It was hypothesised that twelve weeks of treatment with a lotion containing essential oils with \textit{in vitro} acetylcholinesterase-inhibiting properties would show a statistically significant improvement in participants’ cognitive ability and behavioural characteristics compared to baseline, and controls at each data measurement point. This hypothesis and its derivation from the literature and preparatory laboratory work are outlined in Chapters 5.

7.1.2 Study Population

The study population was selected from residents of aged care facilities that participated in the survey of aromatherapy use in aged care facilities in the Northern Rivers area (Chapter 4). Ten facilities within a 150 km radius were involved in the study.

7.1.2.1 Inclusion criteria

The primary inclusion criteria were a diagnosis of dementia or short-term memory problems, and a Mini-Mental State Examination (MMSE) score between 10 and 26 in the last 12 months. A baseline MMSE was carried out by researchers once the consent forms had been obtained, and participants scoring outside the 10-26 range were excluded from the study.
The diagnosis of dementia or short-term memory problems was confirmed from medical records, although type and severity of dementia was not quantified using accepted standards like DSM-IV or ICD-10.

Furthermore, participants also had to:
1) be more than 65 years old;
2) already be on an aromatherapy care plan or be deemed by staff to be unlikely to be disturbed by the study treatment;
3) have English as their first language;
4) have been living in the nursing home for more than 3 months, to reduce likelihood of confusion due to change of environment.
Residents with non-acute concomitant diseases were allowed to participate if their disease(s) were medically controlled.

7.1.2.2 Exclusion criteria

Participants were excluded from the study at baseline if they had:
1) had a myocardial infarction or stroke in previous 3 months;
2) epilepsy;
3) eczema, psoriasis or dermatitis around the neck and shoulders area;
4) known allergy to Eucalyptus, Cypress, Ginger, Lemongrass, Lime or Mandarin essential oils or aqueous cream;
5) an adverse reaction to treatment patch-tests given during screening process;
6) current treatment with anti-cholinesterase or anti-cholinergic drugs;
7) vision or hearing impairments that would prevent them from undertaking the MMSE;
8) extreme behaviour difficulties that would pose problems for the researchers during the MMSE.

Criteria 1) - 5) were included to protect the participants with a high medical risk or who would be more likely to experience adverse reactions to the treatments. Criterion 6) was included to prevent confounding of the results by the use of drugs with similar activity to the treatment preparations.
7.1.2.3 Information collected from medical records

To control for the possible differences in illness levels between groups that may have affected their ability to respond favourably to the treatments, participants’ medical records were investigated to ascertain the presence or absence of certain diseases and also to ascertain the levels and types of concurrent medications people were receiving.

Hypertension, cancer and diabetes were recorded as these are high risk diseases for persons with dementia, and indicators of possible serious illness. Asthma and hay fever were recorded because the aromas may have exacerbated people’s asthma (a possible reason for withdrawal). Arthritis was recorded because it is a common source of pain and discomfort that can contribute to a person’s depressed mood and thus affect outcomes of the study. Furthermore, people with arthritis usually are medicated with anti-inflammatory drugs that may be protective in Alzheimer’s disease (Etminan et al., 2003).

7.1.2.4 Recruitment

An information session about the study was held at each facility for residents and their carers prior to any invitation to participate in the study. Consent forms were available at the information session, and residents and their carers were informed that they may withdraw from the study at any time that they wish to. If participants or their carers could not attend the session, an information sheet was given to them by the facility staff who then also supervised the signing of the consent forms.

7.1.3 Study design

A randomised, double-blind, controlled, multi-centre clinical study was conducted using three treatments (Treatment A, Treatment B and a control with no essential oils) in a parallel design over a four and a half month period (May-August 2005). The study comprised a four week initial wash-out period, twelve weeks of treatment and a two week return to baseline period, with measurements taken according to the schedule shown in Figure 7.1.
The study was carried out over 18 weeks, with a four week baseline washout phase, a 12 week active treatment phase and a 2 week post-treatment washout. Key: R = Randomisation point, N = NOSGER observations, M = MMSE data collected, V = visits to facilities.

7.1.3.1 Primary outcome measure

The primary outcome measure was change in mean scores between baseline and end-point on the standardised Mini-Mental State Examination (MMSE) (Molloy et al., 1991), originally developed in 1975 (Folstein et al., 1975) (see Appendix 3). Although the MMSE is normally used as a tool for staging dementia, several dementia drug trials have used it as a primary or secondary outcome measure (Burns et al., 2004, Iwasaki et al., 2004, Orgogozo et al., 2002, Orrell et al., 2005).

It was hypothesised that participants treated with Treatment A would show an increase in their MMSE score between baseline and end-point, and participants treated with the Treatment B and the plain control lotion would either show no change or a slight decrease over the three months of the study between baseline and end-point.

7.1.3.1.1 Sample size estimation

The mean annual rate of change on the MMSE for people with mild to moderate dementia is approximately -3.6 points (sd. 3.5) per 12 month period (Swanwick et al., 1998, Holmes & Lovestone, 2003) although annual rate of decline is not necessarily constant for each individual (Holmes & Lovestone, 2003). This would suggest a decline rate of -1.2 points per 3 month period. An acetylcholinesterase inhibitor study resulted in a mean MMSE increase after 3 months treatment of +1.4 points (sd. 3.6) (Hager et al., 2003). Therefore, if the
essential oil treatments were to be considered effective AChE inhibitors *in vivo*, a mean MMSE increase of at least +1.4 points should be expected.

A previous aromatherapy study in people with moderate dementia (Bowles et al., 2002) reported a mean MMSE score increase of 3.12 points (sd. 2.75) after one month’s aromatherapy lotion treatment. However, the sample size was small (n=8).

After discussion with a statistician, an estimate of the mean MMSE score difference of +/- 2.0 points (sd. 3.0) was chosen to calculate the sample size required for a three month aromatherapy study. A power calculation for a three-arm study indicated that the total number of people required was between 75-98 people allowing for 25% drop-outs. Table 7.1 below shows the sample size for a power of 0.8 and 0.9.

<table>
<thead>
<tr>
<th>Power</th>
<th>N per arm</th>
<th>Sample size</th>
<th>Sample size + 25%</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.9</td>
<td>26</td>
<td>78</td>
<td>98</td>
</tr>
<tr>
<td>0.8</td>
<td>20</td>
<td>60</td>
<td>75</td>
</tr>
</tbody>
</table>

Two-tailed estimate of study sample size for a three arm study, allowing an additional 25% for withdrawals. Estimated likely mean difference (sd) = 2.0 (3.0), alpha = 0.05.

### 7.1.3.2 Secondary outcome measures

A secondary outcome measure was mean change between baseline and endpoint scores on the Nurses’ Observation Scale for Geriatric Patients (NOSGER). It was hypothesised that NOSGER scores for people on Treatment A would decrease (i.e. an improvement), and that mean scores for people on the other two lotions would either stay the same or increase over the treatment period.

The Nurses’ Observation Scale for Geriatric Residents (NOSGER) (Spiegel et al., 1991) is a 30 question care-staff rating scale, covering mood, memory, activities of daily living, social behaviour, disturbed behaviour and independent activities of daily living. The scale was designed to be rated by care staff. The NOSGER has also been used as a secondary outcome measure in dementia drug trials (Hager et al., 2003, Orgogozo et al., 2002, Monsch et al., 2004) (see Appendix 3).
7.1.3.3 Treatment formulae

The essential oils and aqueous base lotion were supplied by TP Health, Ballina, NSW and were assessed by gas chromatography and mass spectrometry (GC-MS) to ascertain the proportions of their major constituents. All the oils met with expected industry standards for the oils of that type, and also conformed to expected organoleptic quality (based on expert evaluation by laboratory staff).

An aqueous base lotion containing stearic acid, glycerine, isopropyl myristate, Germaben II (propylene glycol, diazolidinyl urea, methylparaben, propylparaben), dimethicone, cetearyl alcohol, Carbomer 940 and purified water was used as the base for both treatment lotions and as the placebo control as shown in Figure 7.1. The formulation of the two treatment lotions is shown in Table 7.2.

Each essential oil in Treatment A showed the strongest acetylcholinesterase inhibition \textit{in vitro} out of all the aromatherapy oils tested. Treatment A also showed similar AChE inhibition to its component oils. None of the oils in Treatment B showed acetylcholinesterase inhibition \textit{in vitro} (see Figure 6.4, Chapter 6), nor did Treatment B. To match the perceived odour intensity of Treatment A, Treatment B had a slightly lower concentration of essential oils.

Table 7.2 Formulae of treatment lotions.

<table>
<thead>
<tr>
<th>Component</th>
<th>Treatment A</th>
<th>Treatment B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aqueous base lotion</td>
<td>673 mg/g</td>
<td>660 mg/g</td>
</tr>
<tr>
<td>Purified water</td>
<td>300 mg/g</td>
<td>320 mg/g</td>
</tr>
<tr>
<td>Eucalyptus oil</td>
<td>0.009 mg/g</td>
<td>-</td>
</tr>
<tr>
<td>Cypress oil</td>
<td>0.009 mg/g</td>
<td>-</td>
</tr>
<tr>
<td>Lime oil</td>
<td>0.009 mg/g</td>
<td>-</td>
</tr>
<tr>
<td>Mandarin oil</td>
<td>-</td>
<td>0.010 mg/g</td>
</tr>
<tr>
<td>Lemongrass oil</td>
<td>-</td>
<td>0.005 mg/g</td>
</tr>
<tr>
<td>Ginger oil</td>
<td>-</td>
<td>0.005 mg/g</td>
</tr>
<tr>
<td>% of essential oil in blend</td>
<td>27 mg/g</td>
<td>2 mg/g</td>
</tr>
</tbody>
</table>

The treatments were formulated by Spectrum Herbals Pty Ltd, Alstonville, NSW and mechanically packaged into 500mL opaque HDPE plastic bottles with manual pump attachments. Each full pump operation delivered 1.6 mL (+/- 0.2) of lotion. Packaging was carried out under The Australian Code of Good Manufacturing Practice for Medicinal Products (2000). Each lotion bottle was weighed prior to dispensing, and the initial weight written on the label. At the end of the study all lotion bottles were collected from the facilities and weighed to ascertain the amount of lotion that had been used.
7.1.3.4 Application methods

During each phase of the study, 1.6 mL of lotion (1 pump) was applied to the neck and shoulders of each participant before midday. In most cases, participants were in single rooms with their own private ensuite shower. Participants who shared bathrooms were given the treatment lotions in their own rooms to avoid odour cross-contamination on application. A laminated instruction sheet was hung in each shower area or room, with diagrams illustrating the treatment application to remind staff to apply the lotions. Participants were requested to use non-fragranced soap, and staff members were asked to refrain from wearing strong perfume and essential oils if they were applying the lotions.

Care staff wore gloves to apply the lotions and initialled a study sign-sheet to indicate whether they had applied the lotion each morning. On occasions when participants did not receive the lotion (sick or away), staff were requested to contact the researcher as soon as possible.

7.1.3.5 Treatment regime and dosage

During the 4 week baseline washout period, all participants received control lotion. During the 12 week treatment period, participants were randomised to receive either treatment or control lotion. Treatment A delivered a dose of 48 mg of essential oil blend, whereas Treatment B delivered a dose of 36 mg of essential oil blend. After the treatment period, all participants returned to 1 daily pump of the control lotion for 2 weeks.

7.1.3.6 Randomisation and blinding

A list of eligible participants was given to a member of the research team for randomisation. Participants were randomly allocated into sets of three in each facility until all participants had been allocated to a treatment or control. The allocation to treatment was done by repeated selection of one of three marbles with a 0, 1 or 2 on them out of a cloth bag.

Participating facilities were given a numerical code, and participants from each facility were given a three digit code, the first of which was the facility number, the next two being the number of the person in the study. The staff member undertaking the randomisation labelled the control and treatment bottles appropriately, and the researcher remained blinded to allocation until the code was broken at the end of the study.
Facility care staff and participants were blinded to the differences in anti-cholinesterase activity of the essential oils in each treatment lotion. Pilot testing revealed that the odour of both lotions was gone from the skin within half an hour of application, and that the odours were not readily discernible at normal inter-personal distances. As the MMSE testing was always carried out at least an hour after treatment application, the researchers were not unblinded by smelling the lotions on the participants.

7.1.3.7  Procedural protocols

Ethics approval was obtained from the Human Research Ethics Committee at Southern Cross University (approval number ECN-04-201). Approval for the study was obtained from the head office or facility director of each facility prior to commencement.

Training sessions were given in each facility to ensure staff compliance with treatment protocols and use of the NOSGER. Seven facilities elected to have just one person complete the NOSGER forms, whereas the other three had various numbers of staff complete them.

Information sessions for residents and relatives were offered in each facility. Signed consent forms were obtained after the sessions from relatives and, where possible, from participants as well.

7.1.3.7.1  Screening techniques

Once residents were accepted as potential participants in the study, a screening meeting was arranged with each resident. During this initial meeting, the researchers carried out an olfactory test, a patch test and the initial MMSE test.

7.1.3.7.2  Olfactory test procedure

Participants were asked to smell each lotion to assess their olfactory ability. They were asked a) if they could smell anything, b) if so, whether they liked the smells and c) whether they could identify either smell. It was considered necessary to evaluate whether the odours were acceptable to people in the trial prior to commencement, to avoid withdrawals and poor treatment compliance due to dislike of the odours.
7.1.3.7.3 Patch-test procedure

Participants were asked to apply a small amount of Treatment A and Treatment B lotions one to either side of their neck prior to the MMSE. The plain lotion was not patch-tested. They were asked how the lotions felt before the MMSE and after the MMSE (a 10-15 minute time interval). As an adverse reaction would exclude the individual from the study, any adverse changes, such as reddening of the skin, or itching, were noted.

7.1.3.8 Collection of demographic data

Age, gender, weight, length of stay in the facility, medical, smoking and educational histories and concomitant medications were collected by the researcher before the end of the study. This was done by consultation of the client records and medication charts, and if necessary by asking staff and residents.

7.1.3.9 Statistical methods

Data were entered into a spreadsheet, and approximately 10% of entries were randomly double-checked for accuracy. Descriptive statistics were generated for the variables using SPSS v11.0 for Windows. A one-way ANOVA was used to assess the similarity of participant groups at baseline.

The study had a hierarchical structure, so student’s t-tests were not appropriate statistical tests for the data. A multi-level statistical model was created to analyse the primary and secondary outcome measures. Variance components due to ‘facility’, ‘participant within facility’ and ‘observations within participant’ were calculated using the restricted maximum likelihood (RML) estimation model (Snijders & Bosker, 1999) in MLwiN software (Goldstein, 2003). The fixed effects were the three different treatments. Response variables were generated from the MMSE and NOSGER measurements.
7.2 Results

7.2.1 Participants

Eleven facilities were initially approached, with 644 potential participants. After screening, 98 people were randomised from 10 facilities to treatment groups. Of these, complete data sets were collected for 72 people. It was decided that it was appropriate to analyse only complete data sets. The mean age for 58 women whose data was analysed was 85.8 years (sd 6.2, range 68-97), and for the 14 men was 83.1 (sd 6.5, range 72-94). All participants had a diagnosis of cognitive impairment, short-term memory loss or dementia on their admission notes, but no formal assessment of dementia type was carried out. Participant flow within the facilities and within the treatment groups is illustrated in Figure 7.2.

7.2.1.1 Withdrawals from the study prior to data analyses

Nine people from facilities 1 and 2 were omitted from the final analyses as there was a mix-up between the treatment lotions for the participants and it was not possible to determine who had received which lotion at the end of the treatment period. Two people were withdrawn by the researcher after discovery that their baseline MMSE scores were outside the inclusion criteria.

One person with Huntington’s disease was entered into the study. A decision was made to omit their data from the final analyses because they also turned out to be less than 65 years old and therefore outside the inclusion criteria.

Five people withdrew from the study on their own initiative, either because they felt it wasn’t working (2) or because they developed itchy skin (2). However, the itchy skin in both cases occurred on the legs and arms, which did not have lotion applied to them. One person moved to another facility and was withdrawn from the study. The eight people who died during the study had previous histories of serious chronic illness. The treatments were not considered to have contributed to the causes of death. The total number of people withdrawn from the study or lost to follow-up was 26 out of 98, or 26.5%.
Figure 7.2 Participant flow diagram

Assessed
N=644

Excluded
N=546

Randomised
N=98

Control
N=31

Commenced treatment
N=28

Lost to follow up
N=5
(Died n=3
Withdrawn n=2)

Analysed
N=23

Treatment A
N=36

Commenced treatment
N=30

Lost to follow up
N=3
(Died n=3)

Analysed
N=27

Treatment B
N=31

Commenced treatment
N=27

Lost to follow up
N=5
(Died n=2
Relocated n=1
Withdrawn n=2)

Analysed
N=22
7.2.1.2 Treatment of missing data

Missing data points were excluded from the analyses by variable. For example, MMSE scores of subjects with one missing NOSGER score were still analysed.

7.2.2 Comparison of dependent variables between treatment groups

Tables for the following descriptive data can be found in Appendix 3 for further details.

7.2.2.1 Age

Group A, Group B and Control had mean ages of 86.3 years (sd 6.7), 86.6 (sd 5.5) and 82.8 (sd 6.2) respectively. One-way ANOVA revealed that the differences between groups were not significant, $F = 2.696$, $p=0.075$.

7.2.2.2 Weight

Group A, Group B and Control had mean weights of 61.5 kg (sd 15.2), 64.8 (sd 12.0), 63.6, (sd 14.9) respectively. One-way ANOVA revealed that the differences between groups were not significant, $F = 0.313$, $p = 0.733$.

7.2.2.3 Length of residence in the facility

Group A, Group B and Control had mean lengths of stay in the facility of 28.3 months (sd 26.3), 38.3 (sd 31.1), 35.6 (sd 42.0). One-way ANOVA revealed that the differences between groups were not significant, $F = 0.604$, $p = 0.550$.

7.2.2.4 Gender

Group A had 78% women, 22% men (n=27 analysed), Group B had 86% women, 14% men (n=22 analysed), and the Control group had 78% women, 22% men (n= 3 analysed).

7.2.2.5 Recorded illnesses

Hypertension was recorded for 33% of participants, cancer for 10%, diabetes for 14%, asthma or hay fever for 17% and arthritis for 35%. None of these conditions were significantly more
prevalent in any of the treatment groups (all p>0.065). Arthritis was more prevalent in Group B, approaching statistical significance, \( F = 2.849, p = 0.065 \).

### 7.2.2.6 Concurrent medications

The mean number of medications taken by the whole sample was 8.6 (sd 3.8, range 0-18). The means for the treatment groups were not significantly different, \( F = 0.873, p = 0.422 \). Seventy-one percent of participants were on analgesics, 61% on anti-hypertensives, 51% for prevention of stroke, 44% for prevention of constipation, 36% on heart-stabilising medication, and 35% on anti-depressants. Medications for anxiety, insomnia, psychotic behaviour, epilepsy, thyroid deficiency, B12 and folate deficiency and lowering cholesterol were taken by less than 20% of the participants, but more than 10%. There were no significant differences by one-way ANOVA between the groups at \( p = 0.05 \) for any of the types of medications.

### 7.2.2.7 Educational levels

All except one participant attended school until at least age 12. Seventy-five percent attended an additional couple of years of secondary school. Three participants had some form of tertiary or further trade education (one was a doctor, one a qualified music teacher and the other, a nurse).

### 7.2.2.8 Smoking history

Sixty-eight percent of the participants had never smoked, 28% used to smoke but gave up for various reasons, and only 4% still smoked daily.

### 7.2.2.9 Olfactory ability

Participants were asked to smell the two essential oil lotions and state whether they could smell them, whether they liked the smells and whether they could identify them (see Table 7.3).
Table 7.3 Results of lotion smelling exercise.

<table>
<thead>
<tr>
<th>Response type</th>
<th>Can smell A</th>
<th>Can smell B</th>
<th>Likes A</th>
<th>Likes B</th>
<th>Identifies A</th>
<th>Identifies B</th>
</tr>
</thead>
<tbody>
<tr>
<td>No</td>
<td>16</td>
<td>14</td>
<td>5</td>
<td>4</td>
<td>26</td>
<td>34</td>
</tr>
<tr>
<td>Yes</td>
<td>51</td>
<td>53</td>
<td>45</td>
<td>43</td>
<td>8</td>
<td>4</td>
</tr>
<tr>
<td>Wrong word</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>14</td>
<td>12</td>
</tr>
<tr>
<td>Can’t smell</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>16</td>
<td>14</td>
</tr>
<tr>
<td>No answer</td>
<td>-</td>
<td>-</td>
<td>1</td>
<td>6</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>Total</td>
<td>67</td>
<td>67</td>
<td>67</td>
<td>67</td>
<td>67</td>
<td>67</td>
</tr>
</tbody>
</table>

A = treatment lotion A; B = treatment lotion B.

Olfactory ability was fairly well distributed among the treatment groups, with people in Group A slightly less able than the other two groups (see Table 7.4). A one-way ANOVA showed no significance between groups at p = 0.05.

Table 7.4 Ability to smell lotions by treatment group, n=72.

<table>
<thead>
<tr>
<th>Treatment Group</th>
<th>Can’t smell</th>
<th>Can smell odour</th>
<th>1 Can smell odours</th>
<th>2</th>
<th>Missing</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>4</td>
<td>0</td>
<td>17</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>A</td>
<td>7</td>
<td>5</td>
<td>13</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>0</td>
<td>3</td>
<td>19</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

A = treatment lotion A; B = treatment lotion B.

7.2.2.10 Patch test results

There were no adverse reactions noted within 10 minutes of application of Treatment A or Treatment B during the screening meeting.

7.2.3 Compliance of lotion application

Compliance of lotion application was estimated by weighing the used bottles after each phase of the study (see Table 7.5). During the baseline washout, an average daily amount of 1.283 g (sd 0.577) of plain lotion was used on all participants. The difference between treatment groups was not significant by one-way ANOVA (F=2.995, p=0.055). During the treatment phase, Group A received 119 g (sd 38), Group B 121 (sd 38), and the control group 124 g (sd 37). The difference between treatment groups was not significant by one-way ANOVA.
(F=0.099, p=0.905). Using the total group mean, a daily average application was 1.1g of lotion, which was 0.5g less than the intended dose. Density of the lotion was assumed to be 1.0g/mL.

Table 7.5 Lotion compliance descriptive statistics.

<table>
<thead>
<tr>
<th>Treatment Group</th>
<th>Plain lotion weight</th>
<th>Average daily amount of plain lotion (g)</th>
<th>Tx lotion used</th>
<th>Average daily amount of Tx lotion (g)</th>
<th>Number of signatures during Tx</th>
<th>Number of days away or in hospital during Tx</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>Mean 35.504</td>
<td>1.145</td>
<td>124.11</td>
<td>1.1266</td>
<td>83.59</td>
<td>.79</td>
</tr>
<tr>
<td></td>
<td>N 28</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>SD 16.8933</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A</td>
<td>Mean 35.442</td>
<td>1.229</td>
<td>118.27</td>
<td>1.0764</td>
<td>90.96</td>
<td>.77</td>
</tr>
<tr>
<td></td>
<td>N 33</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>SD 11.7715</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>Mean 41.487</td>
<td>1.482</td>
<td>116.54</td>
<td>1.0589</td>
<td>91.75</td>
<td>.46</td>
</tr>
<tr>
<td></td>
<td>N 31</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>SD 14.7231</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>Mean 37.498</td>
<td>1.283</td>
<td>119.49</td>
<td>1.0860</td>
<td>89.00</td>
<td>.68</td>
</tr>
<tr>
<td></td>
<td>N 92</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>SD 14.5959</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Rx = treatment; SD = standard deviation

7.2.4 Description of multi-level statistical model

Due to the hierarchical nature of the study design, it is not correct to presume that MMSE scores of participants from different facilities can be amalgamated into composite treatment groups for statistical analysis. It is necessary to account for possible contributions to variance from the different levels of the study design such as having participants grouped in facilities (Snijders & Bosker, 1999).

A multi-level (repeated measures) model was used for both the primary outcome MMSE and secondary outcome NOSGER test data. For both sets of data, the levels of random variation were the variance due to facility, participants within each facility, and observations within each participant. The fixed effects variables were added in the following order: treatment type; measurement occasion; and the interaction between treatment type and measurement occasion. The significance of the contribution of the treatment type by measurement occasion interaction determined whether changes in scores over time differed significantly by treatment type.
7.2.4.1 MMSE multi-level model results

7.2.4.1.1 MMSE variance components

The software program MLwiN (Rasbash et al., 2004) produced a variance components model to estimate the variance of the three sampling levels: facility, participant, observations within participant (see Figure 7.3). The -2*log likelihood (Iterative Generative Least Squares (IGLS) Deviance or Maximum Likelihood (ML) Deviance) statistic was also calculated, and was subsequently used to determine the significance of the fixed effects.

The variance components in the initial model were used to determine whether variance from a particular component was statistically significant. The estimated variance was divided by the standard error to give a one-tailed z-score. The cut-off for one-tailed z-scores ($\alpha = 0.05$) is 1.64.

As the facility variance $v_{0k}$ z score = $0.444/1.045 = 0.425$, therefore the variance from the ‘facility’ level of structure added no significant contribution to the MMSE score variance. Variance due to individuals within facilities $u_{0jk}$ was $z = 12.365/2.444 = 5.059$, which was significant. The variance of observations within individuals $e_{0ijk}$ also contributed significant variance to the overall test scores, $z = 5.442/0.530 = 10.268$.

The following fixed effects variables were added to the model:
• Treatment type;
• Occasion (of measurement);
• Interaction between Treatment type and Occasion.

Models with different fixed effects were compared using the -2 Log Likelihood (-2LL) statistics. The difference between the -2LL statistics for pairs of models with different fixed effects is distributed as chi-square with degrees of freedom equal to the difference in the number of fixed effect variables.

### 7.2.4.1.2 MMSE: Treatment effects

Treatment type was the first fixed effect variable added to the model. Figure 7.4 shows the model with dummy variables for Treatment type.

**Figure 7.4 MMSE: Treatment effects.**

\[
\begin{align*}
\text{MMSE}_{ijk} &\sim N(\beta_0 + \beta_1 \text{TX\_TYPE\_1} + \beta_2 \text{TX\_TYPE\_2}, \Omega) \\
\beta_0 &= 22.053(0.802) + \mu_{0k} + \varepsilon_{0jk} \\
\mu_{0k} &\sim N(0, \Omega_\mu) : \Omega_\mu = \begin{bmatrix} 0.407(1.020) \end{bmatrix} \\
\varepsilon_{0jk} &\sim N(0, \Omega_\varepsilon) : \Omega_\varepsilon = \begin{bmatrix} 5.441(0.530) \end{bmatrix}
\end{align*}
\]

Comparison of the likelihood deviance results for the two models was done by subtracting the -2log likelihood (IGLS Deviance) of the treatment effects model from the initial model. The difference in likelihood deviances was 1448.749 – 1445.818 = 2.931. A chi-square test using this statistic determined there were no significant differences among treatments overall (\(\alpha = 0.05\), df=2, p=0.231).
7.2.4.1.3  MMSE: Treatment and Occasion effects

The next version of the model contained dummy variables for both Treatment type and Occasion (of measurement) fixed effects (see Figure 7.5).

Comparison of the likelihood deviances from the Treatment effects model and this one gave a difference of 1445.818 – 1427.124 = 18.694. A chi-squared test revealed that Occasion contributed significantly to the variance, when the Treatment variable was included in the model (α = 0.05, df = 3, p = 0.0003).

Figure 7.5 MMSE: Treatment + Occasion (of measurement)

<table>
<thead>
<tr>
<th>MMSE_{ijk}</th>
<th>~ N(X\beta, \Omega)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MMSE_{ijk} = \beta_{0ijk}\text{CONS} + -1.751(1.047)TX_TYPE_1_{ijk} + -1.231(1.101)TX_TYPE_2_{ijk} + 0.771(0.378)OCCASION_2_{ijk} + -0.333(0.380)OCCASION_3_{ijk} + 1.144(0.382)OCCASION_4_{ijk}</td>
<td></td>
</tr>
<tr>
<td>\beta_{0ijk} = 21.652(0.835) + v_{ijk} + u_{ijk} + e_{ijk}</td>
<td></td>
</tr>
<tr>
<td>\left[ v_{ijk} \right] \sim N(0, \Omega_v) : \Omega_v = \begin{bmatrix} 0.415(1.018) \end{bmatrix}</td>
<td></td>
</tr>
<tr>
<td>\left[ u_{ijk} \right] \sim N(0, \Omega_u) : \Omega_u = \begin{bmatrix} 12.265(2.408) \end{bmatrix}</td>
<td></td>
</tr>
<tr>
<td>\left[ e_{ijk} \right] \sim N(0, \Omega_e) : \Omega_e = 5.057(0.493)</td>
<td></td>
</tr>
</tbody>
</table>

-2*loglikelihood(JGLS Deviance) = 1427.124(283 of 288 cases in use)

Variables: [v_{ijk}] = Facility; [u_{ijk}] = Participant within facility; [e_{ijk}] = observations within participant; \Omega = [estimated variance per item (standard error)]; TX\_TYPE\_1 = Treatment A; TX\_TYPE\_2 = Treatment B; Occasion = repeated test occasions 1-4.

7.2.4.1.4  MMSE: Treatment, Occasion and “Treatment*Occasion” interaction effects

To determine whether the difference between Occasions was significantly different between Treatments, the variance due to the Treatment by Occasion interaction dummy variables were included in the model (see Figure 7.6).
Figure 7.6 MMSE: Treatment + Occasion + ‘Treatment by Occasion’ interaction effects.

\[
\begin{align*}
\text{MMSE}_{ijk} & \sim N(\mu, \Omega) \\
\text{MMSE}_{ijk} = & \beta_{0ijk} \text{CONS} + -1.708(1.195)TX\_TYPE\_1_{ijk} + -1.531(1.252)TX\_TYPE\_2_{ijk} + \\
& 0.713(0.677)\text{OCCASION}\_2_{ijk} + -0.765(0.677)\text{OCCASION}\_3_{ijk} + \\
& 1.322(0.677)\text{OCCASION}\_4_{ijk} + 0.169(0.920)T1^0O2_{ijk} + 0.314(0.920)T1^0O3_{ijk} + \\
& -0.673(0.925)T1^0O4_{ijk} + -0.031(0.961)T2^0O2_{ijk} + 1.039(0.968)T2^0O3_{ijk} + \\
& 0.221(0.968)T2^0O4_{ijk} \\
\beta_{0ijk} & = 21.731(0.907) + v_{0ijk} + u_{0jk} + e_{ijk}
\end{align*}
\]

Again, the difference in -2LL deviances was compared: 1427.124 – 1423.885 = 3.239. A chi-squared test revealed that the interaction between Treatment Type and Occasion does not account for a significant amount of variance in MMSE scores (\(\alpha = 0.05, \text{df} = 6, p = 0.778\)).

This result indicates that Treatment Type did not account for the observed differences in MMSE scores, and therefore that results for the sample should be considered as a whole.

7.2.4.1.5 MMSE: Occasion effects only

Given that Treatment Type did not make a significant difference to the changes in scores over Occasion, it is appropriate to model only these changes in the whole sample. The model was repeated with dummy variables for Occasion only (see Figure 7.7).
Figure 7.7 MMSE: Occasion effects only.

\[ \text{MMSE}_{ijk} \sim N(\gamma_0, \Omega) \]
\[ \text{MMSE}_{ijk} = \beta_0 + \text{CON} + 0.771(0.378)\text{OCCASION}_2 + -0.332(0.380)\text{OCCASION}_3 + 1.146(0.382)\text{OCCASION}_4 \]
\[ \beta_0 = 20.612(0.549) + \nu_{ik} + \epsilon_{ijk} \]
\[ \nu_{ik} \sim N(0, \Omega_v) : \Omega_v = \begin{bmatrix} 0.467(1.056) \end{bmatrix} \]
\[ \epsilon_{ijk} \sim N(0, \Omega_e) : \Omega_e = \begin{bmatrix} 12.398(2.432) \end{bmatrix} \]
\[ \epsilon_{ijk} \sim N(0, \Omega_e) : \Omega_e = \begin{bmatrix} 5.057(0.492) \end{bmatrix} \]

Again, the \(-2\text{LL deviance}\) was compared with the initial model, 1448.749 \(- 1430.042 = 18.707\). A chi-squared test indicated that Occasion contributed significantly to the variance in MMSE scores (\(\alpha = 0.05, \text{df} = 3, p = 0.0003\)).

The estimated whole sample mean scores for each occasion (± 95% confidence interval) were: Occasion 1, 20.2 (± 1.08); Occasion 2, 21.4 (± 1.07); Occasion 3, 20.3 (± 1.07); Occasion 4, 21.8 (± 1.08). Figure 7.8 shows these results graphically.

Figure 7.8 Mean MMSE scores for whole sample.

Error bars = standard error.
The frequencies of mean score differences between Week 16 and Week 4 (i.e. the difference between the end and beginning of the treatment lotions) are shown in Figure 7.9.

A positive number on the Score difference scale (x-axis) indicates an improvement during the treatment, whereas a negative number indicates a decline in scores over the treatment period. Ten out of seventy-two people on the trial (13.9%) experienced an improvement of greater than 3 points during the trial. The mean MMSE score of these ten people at Week 4 was 15.8 (sd 4.09) and the mean scores improved to 19.5 (sd 4.03) by the end of the treatment period, Week 16.

Figure 7.9 Frequency of mean score differences on the MMSE (Week 16 minus Week 4).

7.2.4.2 Summary of results from multi-level model analysis

The purpose of using a multi-level repeated measures model to analyse the data was to appropriately model the data given their non-independence arising from repeatedly measuring the same facilities, and within facilities, the same participants over occasion. The results can be summarised as follows:

- There was no significant variance at facility level.
- There was no significant difference between treatment groups on changes over time.
- The changes over the time on the whole sample were significant.
7.2.4.3 **NOSGER multi-level model results**

The same multi-level model approach was used to examine the NOSGER scores (details in Appendix 3). The main results were:

- There was no significant variance due to the facility level random effects variable.
- The variance in NOSGER scores came from within individuals and observations within individuals.
- There were no significant differences between Treatment type, Occasion or Treatment by Occasion fixed effects variables.

The descriptive statistics for the three different NOSGER occasions, illustrate the non-significant trend towards a decrease in scores over time, suggesting a mild improvement in function and behaviours (see Table 7.6).

| Table 7.6 Descriptive statistics for NOSGER scores by whole group |
|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|
|                   | N  | Mean  | Std. Deviation | Minimum | Maximum |
| NOSGER 1          | 70 | 73.04 | 16.836         | 31      | 108     |
| NOSGER 2          | 71 | 72.13 | 18.762         | 33      | 115     |
| NOSGER 3          | 66 | 72.77 | 15.988         | 39      | 108     |

7.3 **Discussion**

7.3.1 **Summary of main findings**

The study was designed as a randomised, controlled, parallel group, multi-centre clinical study to compare the effect of two aromatherapy lotions and an unscented control on cognitive function of people with mild to moderate dementia in residential care.

The research hypothesis stated that people receiving Treatment A would be expected to show an improvement between mean baseline and endpoint MMSE and NOSGER scores, whereas scores for people receiving Treatment B or control lotions would be expected to remain the same or deteriorate.

Analysis of the data revealed no significant differences between the effects of treatments on score changes on either the MMSE or NOSGER. However, on the MMSE, the differences between the measurement occasions were significant when the group was considered as a whole.
7.3.2 Comments about the outcome measures

The multi-level model results for the MMSE and NOSGER indicate that among the random effects, there was significant contribution to the variance from the individuals and from the differences between test-scores, but not from the ‘facility’ level. This meant that modelling differences between the facilities had little effect on the model.

The fact that there were no differences between either of the treatments or the placebo was disappointing, but the differences between MMSE measurement occasions are worthy of further discussion. In this study, the MMSE tests were not carried out at equal time intervals. The differences between the mean scores on the four testing occasions (see Figure 7.8) could be explained by a ‘learning’ effect between the test occasions that were separated by only 4 or 2 weeks. A ‘learning’ effect is where participants score better in a repeat of the same test, presumed to be due to participants remembering and learning the expected answers to questions. These findings are similar to a much larger trial of the MMSE on 1,648 outpatients with dementia, where the test was repeated within 2 weeks (Doraiswamy & Kaiser, 2000).

In two trial phases, all participants received just placebo lotion. These two phases were the initial baseline (4 weeks long) and the post-treatment washout phases (2 weeks long). The greatest improvement in mean scores was seen during the post-treatment washout phase, which was also the shortest time between two repeated tests. This suggests that the time between tests was short enough so that participants could have remembered their responses from the previous test.

The initial increase in mean scores between Week 0 and Week 4 could also be attributed to the so-called ‘Hawthorne effect’ which is “the tendency for research participants to change their behaviour because they are part of a study” (James & Talbot, 2005). The extra daily attention from the nursing staff, the novelty of massage with lotions and the sense of importance at being in a trial could have increased participants’ alertness and therefore their ability to perform well on the MMSE (Perry & Hodges, 1999).

The longest time between MMSE tests, 3 months showed a decrease in MMSE scores of -1.103 which is similar to the expected rate of decline for people with mild to moderate
dementia (Han et al. 2000). However, the mean score at Week 16 after 3 months of treatment had not decreased below the initial baseline measurement.

As it seems necessary to account for a possible learning effect and or ‘Hawthorne’ effect (James & Talbot, 2005), the more accurate baseline measurement is most likely to be the one after the 4 weeks of placebo lotion. This means that the overall result of the clinical trial shows that daily massage with a lotion, whether it contains low dosages (<3%) of essential oils or not, does not significantly protect against cognitive decline in dementia, but nor does such aromatherapy treatment exacerbate it.

The conclusion that the MMSE is vulnerable to ‘learning’ effects, even with a 4 week time separation between repeat testing is one that should be noted by future dementia researchers. Using the concept of “responders” and “non-responders” found in other dementia drug research (Lucchi et al., 2004), it is possible that there are “responders” and “non-responders” to aromatherapy. The finding that the ten people who did improve by 3 or more points during the treatment period had a low initial mean score (15.8, sd 4.09) may indicate that the lotion treatment itself was helpful for improving cognitive function in people with more severe dementia, regardless of essential oil content.

However, it is not generally appropriate to do post-hoc analyses such as these, as the sample size is too small to draw meaningful conclusions. A repeat of this trial with sufficient numbers of participants to allow for a meaningful analysis of the “responders” could provide insight into this hypothesis.

7.3.3 Protocol issues

7.3.3.1 Randomisation

Randomisation was carried out by facility, using the list prepared by the facility. These were arranged in room number order, which although apparently random, may have added bias if residents with more severe dementia were allocated adjacent rooms. It would have been better to sort participants by their baseline MMSE scores within each facility, and then randomise in sets of three. This would have meant that the baseline scores for each treatment group would have been more even, although this was not necessary as baseline scores were not significantly different anyway.
7.3.3.2 Measurement scale choice rationale

The MMSE was chosen as the primary outcome measure of cognitive function because:

- It takes 10-15 minutes to administer, which was important given the logistics of testing 98 people in the given time-frame.
- It is able to be administered by a non-clinician (i.e. the researcher and assistant)
- It was already used in all the facilities, so participants were familiar with both the scale and the administration methods. This should have led to less variability in the baseline scores, and possibly diminished possible learning effects on repeated measures.

Factor analysis was not carried out due to the small number of participants in each arm.

The NOSGER was chosen because it covered functional, behavioural and mood characteristics not covered by the MMSE, and could be recorded by nursing staff. Spiegel et al. (1991) intended the NOSGER to encompass 6 dimensions: Memory, Independent Activities of Daily Living, Self-care (or Activities of Daily Living), Mood, Social Behaviour, and Disturbing Behaviour. Initially it was intended to investigate these sub-factors and correlations with the MMSE scale, but seeing as there were no significant effects due to treatment, and no significant differences between any of the measures, it was unnecessary to do sub-factor analyses.

Sensitivity of the NOSGER dimensions to change over time was tested over a 3 month period, and Memory and IADL dimensions were found to be most sensitive to change (Tremmel & Spiegel, 1993). Absolute differences were given arbitrary clinical significance: 0-1 point = no change, 2-3 points = small change, 4 or more points = large change. In this study, the change in mean scores between baseline and end-point were similar to the mean absolute score changes (± 1.92) reported by Tremmel and Spiegel (1993). It can therefore be suggested that the application of lotions, whether with essential oils or not, did not clinically affect the functional, behavioural or mood characteristics of the study population.
7.3.3.3 *Treatment regime and dosage*

A treatment regime of single daily aromatherapy treatments was chosen as this would be minimally disruptive to nursing care routines. There was no suggestion of improved efficacy for greater frequency of treatments from the survey of aromatherapy practices (see Chapter 2).

One approach to determining an appropriate daily dose for clinical trials would be to extrapolate from the laboratory results for the blend that inhibited the enzyme. The *in vitro* IC$_{50}$ of the Treatment A blend was 0.076 mg/mL, but there is no known translation coefficient to compare *in vitro* bovine erythrocyte enzyme inhibition with *in vivo* human brain enzyme inhibition, and the use of a solvent in the assay further complicates the issue, as aromatherapy is not normally carried out with 5% Triton-X100 added to the blend.

However, even if the required plasma concentration for effective treatment was equal to the *in vitro* assay IC$_{50}$, i.e. 0.076 mg/mL, the daily dose would have had to have been about 148 mg per day, which is closer to a 10% dilution of essential oils (presuming 25% dermal absorption and 5 litres of plasma (Tisserand & Balacs, 1995)).

The reason for not using a 10% dilution was an ethical one, in spite of the use of 10% Melissa oil by Ballard et al. (2002) (see review of paper in Chapter 1). Essential oil dosages between 1-5% are widely accepted as safe and appropriate for use in aromatherapy by nurses and aromatherapy professionals (Maddocks-Jennings & Wilkinson, 2004; Meyer, 2001). Lower doses are recommended for the frail aged and for young children and babies. The dose used in the clinical trial was therefore kept within accepted aromatherapy guidelines for frail elderly patients (1-3% essential oils in a blend) (Battaglia, 1995; Price & Price, 1995).

Furthermore, while Melissa oil may be safe at 10% dilution, Lemongrass oil (used in Treatment B) contains high percentages of citral, a known dermal sensitiser (NTP, 2001). Some would argue that because the sensitising effects of citral were shown to be quenched with limonene in guinea pig dermis (Hanau et al., 1983), and limonene is present in high percentages in Mandarin oil (also in Treatment B), then Treatment B should not cause dermal sensitisation. However, Lemongrass oil is usually used at much lower percentages in an aromatherapy treatment than 3.3% dilution (i.e. one third of a 10% dilution). It was considered preferable to err on the side of caution due to the vulnerability of the study population, and keep within accepted aromatherapy dosage guidelines.
The results of the clinical trial showed that dermal applications of approximately 30 mg a day for three months of cholinesterase-inhibiting essential oils did not significantly affect cognitive function. Presuming 25% absorption of the dermal dosage of the 30 mg of essential oil used in the current clinical trial study (Tisserand & Balacs, 1995), plasma levels could be expected to be around 0.0015 mg/mL.

The ‘no observed adverse effect level’ (NOAEL) is considered to be the highest dosage level at which a drug substance causes none of the specified adverse health reactions. For example there could be cytotoxic NOAELs or developmental NOAELs. Unfortunately, there was no readily available NOAEL data for dermal applications of the whole essential oils used in this study. To check that the dosages used in the trial were at least expected to be safe, comparisons were made with oral NOAELs for the major components of oils in the blends.

The NOAEL_{oral} for 1,8-cineole, the major component in Eucalyptus oil (Treatment A) was 300 mg/kg of body weight per day for male Fischer344 rats in a 28-day ingestional study (NTP, 1987). The mouse NOAEL_{oral} for 1,8-cineole was 32 mg/kg of body weight per day over an 80 week study period (Roe et al., 1979).

The dosage levels used in the clinical trial were therefore 20,000-fold lower than the oral mouse NOAEL for 1,8-cineole, so were extremely unlikely to cause adverse effects. However, it appears as though the dosage levels were too low to cause any beneficial effect either.

A dose regimen trial should now be conducted to ascertain the dermal NOAEL in the study population, and indeed whether higher doses of AChE-inhibiting essential oils can affect cognitive function without serious adverse effects.

7.3.3.4 Absence of a ‘no-treatment’ arm

A ‘no-treatment’ arm was considered in the study design phase, but was not included. The decision to exclude the ‘no-treatment’ arm in the final protocol was made because the research hypothesis concerned the effect of essential oils, compared to no essential oils. Another study could be done with the ‘no-treatment’ arm, which could explore whether essential oils, or massage, are the most effective components of an aromatherapy treatment.
7.3.3.5 Sample size

The sample size for each arm was initially calculated expecting the mean change on the MMSE between end-point and baseline to be ± 2.0 (sd 3.0). The actual mean change was -1.103 (sd 3.081).

The lack of statistical significance for the results may be due to insufficient power, and a repeat of the study should consider increasing the sample size using these figures to estimate numbers of subjects per arm.

7.3.3.6 Withdrawals

Although a 25% withdrawal rate was allowed for in the calculation of sample size, 12 of the 25 withdrawals were due to technical errors in the carrying out of the study or from poor communication. Better training of staff as to the importance of each person’s unique identifying code and use of an allocation check-list could have reduced the errors in this regard.

The number of withdrawals due to death could possibly have been reduced by conducting the study during summer months, as nursing staff had the opinion that more aged care facility residents die during winter.

7.3.3.7 Olfactory testing

This clinical trial was designed to test the cognitive effects of essential oils. The hypothesis was the anti-cholinesterase compounds would enter the bloodstream and affect cognition by modulating brain acetylcholinesterase function. Although the clinical trial did not yield statistically significant results, several papers suggest that inhaled odours have an impact on cognitive function during inhalation (Ilmberger et al., 2001, Ludvigson & Rottman, 1989, Millot et al., 2002).

If the intention had been to assess the effects of odour on cognitive ability, the olfactory ability of participants would have been assessed more rigorously. The method used in the trial of asking people with dementia whether they could smell an odour or not, is purely subjective. A triangular forced-choice test between two non-scented and one scented lotion
would have been more rigorous (Green & Swets, 1966), or the use of electrocardiogram (ECG) recording equipment adapted to measure alertness (Torii et al., 1988). However, the possible arousing effect due to olfactory effects of the odours would most likely have diminished in this trial, as the MMSE was administered at least one hour after lotion application.

7.3.3.8 MMSE administration

On reflection, the MMSE was probably not sensitive enough to detect small cognitive changes in dementia over three months. However, the accuracy of the MMSE can be increased if certain considerations are taken into account (Tombaugh & McIntyre, 1992). The following considerations of Tombaugh & McIntyre were taken into the account by the researcher:

- If people felt unwell on the day of the test, they were re-tested on another day. This could have introduced bias into the study as they would have had more days of treatment, but the results did not show this.
- Keeping the site of the test the same was considered important for repeated measures. In most cases, people were tested in their rooms, but sometimes it was in a common area with other people present, which may have compromised their responses.
- The pentagon drawing and the ‘Spell WORLD backwards’ questions were the least reliably scored between raters. All MMSE tests were re-scored by the investigator prior to data-entry as a double check.

7.3.4 Possible sources of bias

7.3.4.1 Multiple study centres

Although it was necessary to recruit sufficient numbers of participants for the study, and therefore a greater number of facilities had to be involved, it would have made it easier to carry out the study in with more people from fewer facilities. Fortunately the contribution of ‘facility’ to the variance in the outcome measures was not statistically significant.
7.3.4.2 **Vulnerability of study population**

Aged people with dementia living in aged care facilities are vulnerable on account of their physical and mental frailty. This study protocol was designed to minimise disturbance and discomfort to participants but some of the compromises may have affected the outcomes of the study.

For example, only residents already receiving aromatherapy as part of their normal care regime, or who were judged by staff to be undisturbed by participation in the study were invited to participate. People with severe behaviours or high confusion levels were therefore excluded from the study. This prevented the observation of the effects of aromatherapy on people with more severe dementia, on whom it may have had a greater effect than the less troubled people included in the trial.

7.3.4.3 **Dementia diagnosis**

Ideally it would have been best to employ an inclusion criterion based on a formal diagnosis of dementia using an accepted guide like the DSM-IV or ICD-10. The inclusion of many different types of dementia in the population of the study may have prevented observation of treatment effects in certain types of dementia. However, the trial was intended to evaluate the usefulness of aromatherapy for cognitive function in dementia, and it was not known at the start whether certain types of dementia would be more responsive to treatment than others.

The results for some individuals suggest that aromatherapy was indeed helpful for some people, and it may be instructive to consider the concept of “responders” versus “non-responders” as other dementia trials have done (Lucchi et al., 2004, Lanctôt et al., 2003). However, as the research design did not include this, post-hoc analysis of this nature is inappropriate and may give biased results due to inadequate power.

7.3.4.4 **Impact of concomitant disease and medications**

Another possible source of bias was made by including people with concomitant diseases, such as diabetes or cardiovascular disease. It would have been impossible to recruit sufficient numbers of participants within the study time-frame if stricter inclusion criteria had been used. However, examination of the descriptive statistics shows that the incidence of disease
types and medication types were comparable between the treatment groups, so it is unlikely that the results were biased in favour of either treatment group.

On the other hand, as most people were taking several medications (mean n=8.51, sd 3.675) this may well explain why the low dosages of essential oils used in the study had an insignificant impact on cognitive function of the participants. It is unlikely that the low dosages of the oils would have interacted or been able to overcome the effects of the medications.

7.3.4.5 Education levels

Education levels have been shown to be the most important external variable for the MMSE (Tombaugh & McIntyre, 1992). Fortunately, the educational levels in the study population were fairly homogenous, with 75% of people finishing school by age 15. Tombaugh & McIntyre (1992) recommend that the MMSE should not be used if people do not have at least a grade eight education and fluent English.

7.3.4.6 Smoking status

Nicotine is an acetylcholine agonist that may improve cognitive function in people with Alzheimer’s disease (Mihailescu & Drucker-Colin, 2000). However, only three people in the trial still smoked, so it was not possible to do a sub-factor analysis to determine if there was a combined benefit of nicotine use and aromatherapy treatment. The MMSE scores from the three people are unlikely to have biased the study outcomes.

7.3.4.7 Inter-rater reliability

It was not possible to coordinate inter-rater reliability tests of the NOSGER raters either between facilities or within facilities due to the distances and time constraints involved. NOSGER scores could have been affected by the fact that three facilities had multiple raters for the observations. However, the multi-level model did not show any significant effect due to the ‘facility’ variable, so it suggests that lack of inter-rater reliability tests did not compromise the study outcomes. In future studies, however, it would be preferable to build inter-rater reliability tests into the staff training program.
7.3.4.8 Study liaison

As it was a multi-centre study, it was necessary to keep in contact with the facilities on a regular basis. One staff member from each facility emerged as the best contact person, and the researcher kept in contact with them by telephone and email. This person was also requested to advise the researcher of any changes, such as participant adverse reactions, illnesses or demise. In several facilities, this person was also the staff member responsible for making the NOSGER observations, but it was not practical in all facilities for this to be the case. This introduced some variability in the NOSGER scores.

Agreements were made at the start of the study with each facility as to what times and days of the week suited each facility best for visits from the researchers. This was confirmed by telephone contact prior to each visit.

Study liaison was on the whole successful, but on reflection, refreshment of training of staff and checking of compliance of lotion application would have been better if done more frequently.

7.3.4.9 Compliance

The average daily dosage given was 1.1g, rather than the intended 1.6g. This failure to apply the intended dosage means that the results had to be interpreted for the new real dosage levels, rather than the intended one. Future trials should have weekly checks of compliance and ongoing staff reminders or training. Some participants had to be hospitalised, but fortunately hospital stays were not extended for more than a few days in most instances.
8 General discussion of the research

8.1 Overview

This thesis reports an investigation into the effects of aromatherapy on cognitive function in people with dementia. A four-phase cascading research framework was designed to provide a rigorous approach to the studies, comprising: investigation of the research situation by literature review and survey; hypothesis and research plan generation; quantitative laboratory assessment of the hypothesis and; a clinical trial to test the results of the laboratory assessment in an appropriate human population. It appears that the framework developed was indeed useful in the execution of the project.

The framework provided a guideline for the three research projects, which flowed from one to another based on the results of the previous step(s). New demographic information about aromatherapy practices in aged care facilities in the Northern Rivers area of NSW was obtained through administration of a survey and essential oils and application methods identified for the next stages of laboratory work and clinical trial; the *in vitro* acetylcholinesterase-inhibiting properties of 25 essential oils were investigated, and two blends created for the clinical trial based on their *in vitro* AChE inhibitory activity; and a three month clinical trial of some of these essential oils in people with dementia showed no significant difference between aromatherapy treatments and a non-scented control treatment on improvement of cognitive function as measured by the Mini-Mental State Examination.

This chapter discusses the main findings of the research project and suggests possible future avenues of research.

8.2 Main findings

8.2.1 Phase One: Investigation – literature review and survey

The literature review of aromatherapy use in dementia revealed that no previous trials had tested dermal applications of aromatherapy for the improvement of cognitive function in dementia. The only paper reporting on the effects of essential oils on cognitive function
concerned a six week trial of 50-150µL of ingested Lavender Sage (Salvia lavandulaefolia) essential oil. The report showed that vigilance scores in people with dementia significantly improved during the treatment phase compared to the control (Perry et al., 2003).

A survey of aromatherapy practices in 28 Northern Rivers aged care facilities identified that aromatherapy is used for 59% of residents, and that half of these facilities give aromatherapy to all of their residents with dementia. Staff perceived aromatherapy to be most efficacious for reduction of agitation, anxiety, muscle tension, pain, restlessness, confusion and insomnia, and increase of relaxation.

Use of aromatherapy was perceived by directors of care to reduce the use of sedative medications, implying that aromatherapy is perceived more as a relaxing, calming therapy than as an agent to improve cognitive function. Only 2.8% of the stated uses for different essential oils pertained to improvement of cognitive function in dementia, which was surprising, given the accepted aromatherapy uses of essential oils like Rosemary, Basil and Peppermint for alertness and improvement of memory in healthy younger people (Battaglia, 1995).

Only 12 adverse events were reported, 9 of which were associated with Lavender oil, and only two of which were associated with dermal irritation. This suggests that dermal applications of aromatherapy can be considered to be very low risk, given the number of treatments being administered daily in the study sample (n=483).

8.2.2 Phase Two: Hypothesis and research plan

As there had been little research on the cognitive effects of essential oils on people with dementia, and the literature review showed some therapeutic potential for essential oils with cholinesterase-inhibiting properties, an hypothesis was generated to investigate this area. The hypothesis was that:

“A blend of essential oils with demonstrated in vitro anti-cholinesterase activity would improve the cognitive function of people with mild to moderate dementia more than a blend of oils with no in vitro activity and more than a non-scented control lotion.”
The research plan comprised screening of essential oils identified in the survey for *in vitro* acetyl cholinesterase activity, development of two blends of essential oils for use in the clinical trial, and design and execution of a clinical trial to compare the two blends to be developed in the laboratory phase.

### 8.2.3 Phase Three: Laboratory work

Twenty-five essential oils identified in the aromatherapy survey were screened for inhibitory activity in an *in vitro* assay of bovine erythrocyte acetylcholinesterase. *In vitro* inhibition of the membrane-bound bovine enzyme is taken to be an indicator of potential pharmacological activity in human Alzheimer’s disease (Savelev et al., 2003), where acetylcholinesterase inhibition is still thought to be a useful pharmacotherapy for the improvement of cognitive function and also more recently, of behavioural and psychological symptoms of dementia (Ibach & Haen, 2004).

Lime (*Citrus latifolia*), Sage (*Salvia officinalis*), Cypress (*Cupressus sempervirens*) and Rosemary (*Rosmarinus officinalis*) essential oils, and the terpenoid 1,8-cineole all inhibited the enzyme to ≥ 50% of controls at 0.454 mg/mL. Lime, Sage and Rosemary oils had previously inhibited human brain enzyme, although Rosemary oil was more active in this study than in the research of Perry et al. (1996). This could be due to the difference in chemical composition of the two Rosemary essential oils or a difference between bovine erythrocyte and human brain enzyme.

The Cypress oil assayed in this study contained 60% alpha-pinene and 19% delta-3-carene, both previously identified as enzyme inhibitors (Miyazawa & Yamafuji, 2005), so it is not surprising that Cypress oil inhibits the enzyme as a whole. However, although both Juniper and Frankincense were expected to show good inhibition due to their high contents of alpha-pinene (54% and 87% respectively), neither of them did. This raises the question of competitive interactions between different essential oil components as investigated by Savelev et al. (2003).

Other bovine erythrocyte enzyme assays of 1,8-cineole showed the following IC\(_{50}\) values: 0.041mg/mL (Miyazawa et al., 1998) and 0.06mg/mL (Savelev et al., 2003). This study showed a mean IC\(_{50}\) value for 1,8-cineole of 0.59mg/mL (range 0.044-1.528 mg/mL). The
variability could have been caused by the use of different batches of 5% Triton-X100 as a solvent for the essential oils in this study, but is not the only source of possible error.

However, in spite of the wide variability in the results, the essential oils were grouped into 4 groups, the first group being those that exhibited dose-dependent inhibition and achieved ≥ 50% inhibition at 0.454 mg/mL, the last group being those with very little inhibition at 0.454 mg/mL. Three essential oils were selected from the first and last group to create pleasant-smelling, affordable aromatherapy blends for the subsequent clinical trial of aromatherapy for cognitive function in dementia. The results of the laboratory testing yielded two blends with distinctly different acetylcholinesterase inhibiting properties that were used in a clinical trial in Phase Four.

### 8.2.4 Phase Four: Clinical trial

Although the clinical trial was designed using a sample size estimate based on literature values for changes in MMSE scores, the actual changes in MMSE scores in the control group were smaller than expected (-1.103 over 3 months, literature estimate -1.2 over 3 months (Holmes & Lovestone, 2003)). This suggests that either the literature values for MMSE change scores are not applicable from one population to another, or perhaps that the placebo treatment had a beneficial effect along with the aromatherapy treatments. Unfortunately, it also means that the sample size was not sufficiently large enough for the study to be adequately powered.

The major flaw in the experimental design was the absence of a no-treatment control arm. However, the hypothesis for the trial was to compare the effects of aromatherapy treatments with a non-scented treatment, which was achieved. The results showed a non-significant decline in mean MMSE scores (from 21.38 to 20.28) from the start of treatment to the end of treatment, and it was found that the major variance between results was not due to Treatment Group, but rather to Testing Occasion. Interestingly, Treatment B, (comprised of ‘inactive’ essential oils) showed less of a decline than Treatment A, although the differences were not statistically significant.

The use of repeated MMSEs showed a possible learning effect although the differences did not reach statistical significance in this study. The mean scores between the initial screening
test and the base-line test increased after one month on non-scented lotion. However, this improvement did not continue after three months of continued treatment with non-scented lotion in the Control group, suggesting a learning effect in MMSEs repeated within four weeks. This was further demonstrated when comparing the end test and post-treatment washout test, which also showed an increase of mean scores.

The MMSE was found to be an imprecise tool for the measurement of cognitive change in a short-term trial, as the standard deviations for the mean scores was in the range of 3.9-4.4 points. The standard deviations could perhaps have been smaller if the initial scores of the sample population had been within a tighter range (18-23, rather than 10-26), as Han et al., (2000) commented in their review on the use of the MMSE as a tracking tool for cognitive change in dementia. The reason the initial range was so large in the current project was to obtain a sufficient sample size within the same geographical area, but future projects should keep the range of initial MMSE scores tight if the MMSE is to be used as a measurement tool.

There were no statistically significant improvements or declines in functional ability, mood, behaviours or sociability during the treatment period, as assessed by the NOSGER. This suggests that the dosage of essential oils used was not sufficiently large enough, or that the effects of aromatherapy do not persist beyond the time when the treatment can be smelled, if indeed there are any objectively measurable effects of aromatherapy on cognitive function.

As this was the first clinical trial of dermal aromatherapy for improvement of cognitive function in dementia, several methodological issues were identified that could increase the rigour of future clinical investigations on this topic.

8.3 Comment on methodological issues

8.3.1 Investigative phase

The research questions driving the creation of the survey were not defined clearly enough. Certain crucial questions were omitted, such as: ‘How effective is aromatherapy for cognitive improvement in dementia?’ and ‘What dosages are used?’. Other issues such as the amount of staff aromatherapy training and perceptions of environmental influences on dementia
behaviours, while interesting, were not crucial to the research question, and should have been omitted.

The survey did not distinguish between respondents’ beliefs about aromatherapy in general and their experiences with aromatherapy in the facility. It would have been better if this could have been more clearly delineated, because beliefs are possibly more vulnerable to change over time, whereas experiences are more likely to provide objective data. Furthermore, the survey was not repeated with any of the respondents, so the test-retest validity of the survey is in question.

8.3.1.1 Methodological improvements to investigative techniques

The survey could be redesigned to better answer how effective aromatherapy is perceived (by staff) to be for improvement of cognitive function in dementia in each facility. All the questions could be edited with this in mind, though it is still useful to include questions about mood and behavioural symptoms, as these are likely to correlate with cognitive changes (Thompson et al., 2004). Professional survey design advice could be sought to improve the validity and rigour of the survey design, which was not possible in the tight budget and time-frame of the current study.

The survey should be given to three nursing staff members from each facility, including the person responsible for the aromatherapy care planning. This would allow for inter-rater reliability tests to be carried out, and would control for respondent variation. Aromatherapy care planners are possibly more likely to be ‘believers’ in aromatherapy, although most of the people appeared keen to present their true observations of aromatherapy, as evidenced by the bi-modal distribution of opinions. A repeat of the survey could be undertaken within one month in all facilities tested. This would allow for test-retest validity of the survey to be assessed.

Once modified and validity-tested, such a survey could be carried out in different areas of Australia, to investigate whether the Northern Rivers survey results are typical of aromatherapy practices in aged care in the wider population. This would increase the relevance of the data to a wider population, and provide more fuel for future research on the validation of aromatherapy use for dementia.
As some oils are used quite widely for relief of arthritis (see Table 4.8), it would be interesting to assess whether the anti-inflammatory effects of the oils also have protective effects on people’s brain function (Townsend and Pratico, 2005).

8.3.2 Laboratory phase

The acetylcholinesterase assay results were widely variable between replicates and between occasions, and the source of variability was not clearly identified, although the choice of Triton-X100 as a solvent and operator errors (e.g. reagent preparation and pipetting) appear to be the major contributors.

8.3.2.1 Methodological improvements to laboratory phase

The experiment could be repeated in tandem (from reagent preparation through to data analysis) with another person equally skilled in the technique, to determine the extent of operator error. Once established whether the major source of variability was from operator error or not, comparison of the effects of Triton-X100 and other solvents could be carried out systematically. Different solvents may affect the inhibitory properties of the different essential oil compounds, due to interactions with polarity and molecular size.

Galantamine and 1,8-cineole should be used on every plate as positive controls unless the variability between plates can be reduced to acceptable levels.

Exact reproduction of a previous method should be attempted and compared with the modified method used in this study.

Once the methodological issues of the cholinesterase assay are resolved, greater numbers of essential oils could be screened, including essential oils of the same type from different origins, and of different ages. The most active oils could be further assessed for more potent inhibitors, in particular Lime essential oil. De-terpenised Lime oil is available and may well contain the active components, as the monoterpenes limonene and gamma-terpinene have only mild in vitro inhibitory activity.
8.3.3 **Clinical trial phase**

The MMSE was chosen as the primary outcome measure mainly due to its availability, the speed of administration and calculation, its popularity in aged care, and the fact that it can be administered by non-clinicians. However, its wide variability does make it insensitive to small changes, and future research would either have to go for a longer time, or use a different, more sensitive scale.

The dosage regime used in the study was based on safe aromatherapy practices for the frail elderly, as no evidence was found in the literature to guide estimations of efficacy.

Compliance of treatment application by care staff was also a possible source of variation in the results, as the average daily application of lotions was 0.5g less than the intended dose, and in some cases was much less due to missed days (see Section 7.2.3). Tighter monitoring of staff by the researcher, and retraining where appropriate could have improved the compliance.

### 8.3.3.1 Methodological improvements to clinical trial

It could have been preferable to carry out an initial open-label dose regimen pilot trial of Treatment A to determine whether a dosage regime with significant effects on cognitive function does exist. Dermal application and ingestional routes could also have been compared to elucidate the most effective application method.

In a future repeat of this trial, the treatment phase should be for twelve months, with repeated cognitive tests at two-monthly intervals to control for the ‘learning’ effect and variability of cognitive function within the disease. A ‘no-treatment’ arm should be included. The ADAS-cog scale should be used as well as the MMSE in future research, as it would provide comparability with other dementia drug trials. More objective behavioural and functional ability scales than the NOSGER could also be used.

The population tested should have more similar dementia stages, preferably between 18-23 on the MMSE, and of Alzheimer’s type without other types of dementia. Formal assessment of dementia should be carried out for each participant using one of the standard lists of diagnostic criteria, for example the DMS-IV.
Once an effective aromatherapy dosage regime is established, a head-to-head clinical trial comparing aromatherapy with one of the cholinesterase drugs could further investigate the usefulness of aromatherapy as a cost-effective dementia therapy.

### 8.4 Suggestions for aromatherapy research in aged care and dementia

As aromatherapy appears to be widely used in aged care, and it is perceived as an effective therapy for agitation and anxiety and arthritic pain, all common ailments in ageing residents, it appears worthy of further investigation. Future aromatherapy research projects could benefit from the use of the methodological framework employed in this thesis, and could include:

- evaluation of essential oils as multi-potent agents in dementia, with potential anti-inflammatory, anti-oxidant, anti-cholinesterase and neurotransmitter modulating capabilities;
- objective evaluation of aromatherapy for both psychological and physical sources of discomfort in aged care residents;
- determination of ‘best practice’ for use of aromatherapy in aged care, especially for residents with dementia, including most effective application methods and dosage regimes;
- prospective evaluation of the use of sedatives in the presence and absence of aromatherapy;
- quantitative evaluation of pharmaceutical cost-savings in facilities that use aromatherapy;
- evaluation of the effects of aromatherapy on staff members and visitors to the facility, not just residents;
- determination of pharmacological effects of essential oils, whether physical or psychological.

### 8.5 Conclusions

This research project has contributed to the knowledge of aromatherapy use in dementia in three ways. Firstly, the main research hypothesis was disproved, results showing that the aromatherapy regime used in the clinical trial (oils, dosage frequencies and application
methods) does not offer significant cognitive benefit to people with mild to moderate dementia over a three month period as tested by the MMSE. Further dosage exploration work should be done to establish whether dermally applied essential oils can have a dose-dependent effect on cognitive function at a safe concentration. Modifications to the trial protocol that may have produced statistical and clinical significance include: lengthening the time period to 12 months; narrowing the inclusion criteria; using more sensitive measurement instruments, and increasing the dosages of essential oils or using ingestion rather than dermal application.

Secondly, results of the survey of aromatherapy practices in aged care facilities suggest that aromatherapy is being used for a whole raft of other conditions, other than the improvement of cognitive function. These include the management of arthritis and related musculoskeletal pain, the reduction of insomnia and restlessness and the reduction in anxiety and agitation. It is easy to imagine how persistent pain or inability to get comfortable could cause insomnia and restlessness, and how pain and lack of sleep could cause increases in anxiety and agitation, regardless of whether a person has dementia or not. Although the ‘care-giver burden’ literature reviewed in Chapter 2 suggested that aggressive behaviours added most to care-giver burden, reduction of any of these conditions would improve residents’ well-being, and consequently reduce carers’ stress.

The results of the survey did not provide objective measures about the reduction of arthritic pain or the reduction of insomnia, although the existing use of aromatherapy for these conditions suggests that the nurses perceive aromatherapy to be effective. Also the fact that aromatherapy was perceived to reduce the use of sedative and analgesic medications supports the suggestion that further quantitative research should be carried out to ascertain how effective aromatherapy actually is for these conditions in aged care facilities, and which application methods are most effective. Quantification of these results could lead to the wider use of aromatherapy in aged care.

The third contribution of the project is the development of the four-phase cascading research framework that offers a template for research in other areas with little existing research. For example, an intensive care researcher could firstly use a survey to explore the variety and extent of different aromatherapy practices in intensive care, followed by a laboratory exploration of possible pharmacological properties of essential oils, and concluded by a randomised controlled trial to finally test the selected practices.
Appendix 1

Published paper

Authors’ contributions
A survey of aromatherapy practices in aged care facilities in northern NSW, Australia

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Summary

Directors of care and aromatherapy care planners from 28 residential aged care facilities were surveyed about their practices and perceptions of aromatherapy in their facility. A total of 1032 (59\%) residents in these facilities received aromatherapy, with 47\% receiving daily treatments. The treatments were applied by assistants in nursing in most facilities, with activities officers and registered nurses also applying the treatments. The one essential oil used by all facilities was Lavender (Lavandula angustifolia) followed by most facilities using Tea Tree (Melaleuca alternifolia), Geranium (Pelargonium graveolens), Eucalyptus (Eucalyptus globulus) and Bergamot (Citrus bergamia). Commercial blends were used by 15/28 facilities. The choice of individual essential oils and blends suggests that aromatherapy is considered effective for both behavioural or psychological symptoms and physical ailments like arthritic pain. Nearly all facilities used foot baths and hand, foot, limb and neck-and-shoulders massage. The average monthly cost of materials per person was AUD 4.50. Directors of care also perceived that aromatherapy moderately reduces the amount of pharmaceuticals used. The main types of pharmaceuticals perceived to be reduced by aromatherapy were sedatives and analgesics.

Considering all these findings, it appears as though aromatherapy is being used extensively in aged care facilities to manage symptoms of dementia and age-related physical discomfort. It is clear from this survey however, that further research is required to determine a ‘best practice’ for aromatherapy in dementia and aged care.

Keywords: Aromatherapy; Aged care; Dementia; Essential oil; Drug reduction; Survey
**Introduction**

Aromatherapy is growing in popularity in aged care facilities in Australia and elsewhere in the world. It is thought to be a useful complementary therapy for people with dementia, in particular for managing the difficult or challenging behaviours that arise as the disease progresses. Several reports of aged care aromatherapy trials suggest that aromatherapy can reduce agitation and anxiety in residents with dementia and also help them sleep (Mitchell, 1993, Smallwood et al. 2001, Ballard et al. 2002, Bowles et al. 2002 and Holmes et al. 2002). A review of these trials shows that they all used different treatment protocols for managing dementia subjects. This most likely reflects that there is currently no consensus among aromatherapists as to what constitutes an effective aromatherapy treatment regime for aged care residents with dementia.

To develop a more informed understanding about aromatherapy in aged care facilities we set out to ascertain current practice in the regional area surrounding our University. This commenced with a focus group of aged care staff using aromatherapy. It became clear from this focus group that the practice of aromatherapy in aged care facilities is highly variable. It also emerged that aromatherapy was being used for other conditions, not just management of dementia symptoms, and that treatments were designed around management of symptoms, rather than whether a resident had dementia or not.

Against this background, we undertook a survey aimed to describe existing variations in practices and perceptions of aged care staff using aromatherapy and, if possible, to identify the extent to which aromatherapy is being used for the management of dementia in aged care.

**Methods**

**Aims**

The aims of the survey were to:

- Describe the variations in aromatherapy practices in aged care facilities in northern NSW.
- Identify how effective aromatherapy is perceived to be for dementia.
- Determine if any aromatherapy practices correlate significantly with perceptions of efficacy for dementia.
• Describe aromatherapy practices related to management of dementia.

**Participants**

Forty-eight residential aged care facilities were identified and contacted in north-eastern NSW. Twenty-eight of these met the inclusion criteria of having aromatherapy care plans and at least one resident with dementia. Facilities were excluded if they only used essential oils to improve ambient odour ($n = 3$). The Director of Care and the aromatherapist or person responsible for designing the aromatherapy care plans filled in the surveys.

**Survey design**

The survey was in two parts, one part filled out by directors of care and the other part by aromatherapy care planners. Part one of the survey completed by the directors of care covered numbers of residents with and without dementia, categories of staff applying aromatherapy, average monthly cost of aromatherapy materials and perceptions about the extent to which aromatherapy reduces the use of pharmaceuticals, and what types of pharmaceuticals are reduced. Part two of the survey completed by the aromatherapy care planners covered treatment frequency, application method, types of oils and blends used in treatments, and perceptions of efficacy.

Questions included closed-ended “yes/no” questions, open-ended free response questions and 10 cm visual analogue scales on which to mark a response along a continuum; for example, for the question “Which aspects of aromatherapy are helpful for dementia?” the two descriptor phrases at either end of the scale were “Not at all helpful” and “Extremely helpful”. Yes/no questions were coded 1/0, and responses on visual analogue scales were measured to ±0.1 cm. Free responses were grouped by thematic similarity for ease of analysis, for example ‘sleeplessness’ and ‘insomnia’ were grouped together. Surveys were collected without any identifying marks, thus ensuring privacy and confidentiality of information. Ethics approval from the Human Research Ethics Committee at Southern Cross University was obtained.

**Data analysis**

Descriptive statistics for most of the raw data were generated, and factor analysis (principal components extraction and Varimax rotation) was used to create new variables.
summarising perceptions of efficacy. Pearson’s correlations were used to correlate perceptions of efficacy with the descriptive statistics. The statistical package used was SPSS for Windows v11.01 (Statistical Package for Social Sciences, LEAD Technologies, Inc).

**Results**

*Numbers and percentages of residents receiving treatment*

The total number of residents in the participating facilities was 1767 comprising 827 (47%) with dementia and 940 (53%) without dementia. This is lower than the Australian population estimate of 60% of aged care residents having dementia (Access-Economics, 2003). For the survey, the numbers of people having dementia was estimated by the Directors of Care, and may be an underestimate due to the lack of formal geriatrician’s diagnosis for many residents. A total of 1032 (59%) residents received aromatherapy including 468 residents with dementia which was 57% of the total number of residents with dementia and 564 residents without dementia which was 60% of the total number of residents without dementia.

The facilities varied as to whether they used aromatherapy as a treatment for all residents, or only for a few residents. Only five facilities (18%) give all of their residents aromatherapy treatments. 14 facilities (50% of the facilities surveyed) give all of their dementia residents aromatherapy (though not necessarily daily), whereas only nine facilities (32% surveyed) give all of their non-dementia residents aromatherapy.

*Perceptions of efficacy*

Perceptions of efficacy of aromatherapy for various behaviours and symptoms were collected from aromatherapy care planners, with the aim of comparing their perceptions with the various aromatherapy practices. Perceptions were measured on 10 cm visual analogue scales. The two highest mean scores were ‘reduction in arthritic pain’ (8.3, SD = 1.4, range 4.7–10); and ‘increase in relaxation’ (8.0, SD = 1.6, range 3.3–10). However, most of the data was bi-modal with opinions clustered in two groups, either near the top or the bottom end of the range. As it is not statistically valid to compare the means of bi-modal variables, factor analysis was used to group highly correlated items into five new variables (see Methods).
One of these variables was composed of the following items: reduction of agitation, anxiety, muscle tension, pain, restlessness, confusion and insomnia, and increase of relaxation. As agitation, anxiety, restlessness, confusion and insomnia are all symptoms of dementia (Cummings, 1987) we selected this variable as an indicator of how useful aromatherapy was perceived to be for dementia.

This variable was then correlated with other survey items to determine their relationship to the perceived effectiveness of aromatherapy for dementia and is referred to in the text as the ‘perceptions of efficacy’ variable for dementia symptoms. As our focus in this article is the use of aromatherapy for dementia, we have not reported the correlations with the other factor analysis variables.

**Aromatherapy regimens**

In the survey, responses to the question “How often do residents receive aromatherapy treatments?” were either: daily; three or more times a week; 1–2 times a week; or less than once a week, to fit with the established care plan protocols. Seventy-five percent (75%) of dementia residents receiving aromatherapy were treated daily or >3 times per week, whereas 65% of non-dementia residents receiving aromatherapy were treated daily or >3 times per week as shown in Table 1.

**Table 1 Number of residents receiving specific aromatherapy regimens**

<table>
<thead>
<tr>
<th>Group</th>
<th>Aromatherapy regimen</th>
<th>Daily (43%)</th>
<th>&gt;3 per week (32%)</th>
<th>1–2 per week (15%)</th>
<th>&lt;1 per week (10%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dementia (n = 468)</td>
<td>201</td>
<td>150</td>
<td>70 (15%)</td>
<td>46 (10%)</td>
<td></td>
</tr>
<tr>
<td>Non-dementia (n = 564)</td>
<td>282</td>
<td>73 (13%)</td>
<td>113 (20%)</td>
<td>96 (17%)</td>
<td></td>
</tr>
</tbody>
</table>

The numbers of residents in each group (dementia or non-dementia) are the sum of residents in each group receiving aromatherapy in the 28 facilities.

**Daily treatments and perceptions of efficacy**

From the focus group discussions, it appeared that daily treatments for residents with dementia would be more efficacious, and therefore should be perceived as such by aromatherapy care planners. Eleven facilities gave 100% of their treated dementia residents daily treatments, and nine facilities gave 100% of their treated non-dementia residents daily treatments. However, there were no statistically significant correlations between numbers
of residents with or without dementia receiving daily treatments and the ‘perceptions of efficacy’ variable for dementia symptoms.

**Number of aromatherapy applications per day**

The greatest number of aromatherapy applications per day was nine, the fewest was one, with most facilities giving two applications a day for residents receiving daily treatments. In discussion with the focus group prior to the survey, most people said that their aromatherapy care plan protocol allowed for two or three scheduled applications per day, with ‘p.r.n’ (applications as required) also listed as an option for most people. There was no statistically significant correlation between the number of aromatherapy applications per day and ‘perceptions of efficacy’ variable for dementia symptoms.

**Cost of aromatherapy materials**

Directors of care were asked to estimate their average monthly cost of aromatherapy materials if the facility was responsible for purchasing the materials. Twenty-two facilities responded to this question, with the remaining six facilities stating that residents or family purchased the materials. The average monthly cost of materials per person receiving aromatherapy treatments was AUD4.50 (SD AUD4.80). Costs ranged from AUD0.42 to AUD21.05 per person per month. Over half of the facilities (13/22–59%) were spending AUD4.00 or less. Cost of staff wages was not included in these calculations.

*Figure 1. Estimated monthly cost per person for aromatherapy materials. Dollar ranges have been simplified for ease of reading. For example, the range 2–4 means AUD2.01–AUD4.00*
There was no statistically significant correlation between cost of aromatherapy materials and ‘perceptions of efficacy’ variable for dementia symptoms.

**Perceptions about reduction of pharmaceutical use**

Directors of care were asked to rate on a 10-cm visual analogue scale to what extent they felt aromatherapy reduced the use of pharmaceuticals in the facility, as well as what types of pharmaceutical were reduced. Aromatherapy was perceived as moderately useful in reducing pharmaceutical use. The mean response on the 0–10 scale was 5.5 (SD = 2.43) and the usefulness ranged from 0.4–9.0/10 (n = 28 facilities).

In 22 facilities, directors of care reported a reduction in the use of sedatives. This correlated significantly with perceptions of overall reduction of pharmaceutical use (Pearson’s = 0.491, two tailed sig. = 0.008, n = 28). The 11 other classes of drugs perceived by directors of care to be reduced by aromatherapy use were: analgesics, benzodiazepines, antipsychotics, aperients, anti-depressants, arthritis medication, diuretics, respiratory drugs, anti-hypertensives, skin care creams and anti-fungal preparations.

Table 2 shows the drug classes perceived to be reduced by Directors of Care and the numbers of residents receiving aromatherapy in responding facilities. The numbers and percentages suggest that the perceptions of sedative and analgesic reduction are based on extensive observations, not just on a few people. For example, reduction of sedative use was reported by 22/28 Directors of Care. About 860 residents received aromatherapy in those facilities, which represented 63% of the total number of residents in the responding facilities.
Table 2. Types of drug reduced by aromatherapy

<table>
<thead>
<tr>
<th>Type of drug</th>
<th>Facilities reporting % (n)</th>
<th>Residents receiving aromatherapy % (n)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sedatives</td>
<td>79% (22)</td>
<td>63% (860)</td>
</tr>
<tr>
<td>Analgesics</td>
<td>39% (11)</td>
<td>79% (486)</td>
</tr>
<tr>
<td>Benzodiazepines</td>
<td>21% (6)</td>
<td>84% (229)</td>
</tr>
<tr>
<td>Anti-psychotics</td>
<td>21% (6)</td>
<td>42% (168)</td>
</tr>
<tr>
<td>Aperients</td>
<td>14% (4)</td>
<td>69% (158)</td>
</tr>
</tbody>
</table>

Types of drug perceived by the Directors of Care (n = 28 facilities) that were reduced by aromatherapy. The percentage of residents receiving aromatherapy is calculated as a percentage of the total number of residents represented by the reporting facilities. Types of medication listed by only one or two facilities are not reported in the table.

**Categories of staff giving aromatherapy treatments**

Aromatherapy was applied by assistants in nursing (AINs) in 96% (27/28) facilities and Activities Officers, in 68% facilities. Registered nurses (RNs) applied aromatherapy in 60% of facilities, and Diversional Therapists did so in 40% of facilities. Physiotherapy aides applied aromatherapy in 32% of facilities. Only four facilities employed the services of a visiting aromatherapist. In two cases, this aromatherapist was not the aromatherapy care planner who responded to the survey but a person employed by the residents on an individual basis.

Occupational therapists and physiotherapists were least represented among health professionals applying aromatherapy and were only identified in one facility. Volunteers and family members were involved in aromatherapy application in 43% and 39% of facilities, respectively.

**Application methods**

The most prevalent application methods were foot baths and massage of hands, feet, neck and shoulders, and limb joints (>25 facilities, >89%). Other application methods included, in descending order: drops on tissue, clothing or pillow; abdominal massage; after-shower cream; skin integrity cream; vaporiser in bedroom; spray and vaporiser in the facility (14–22 facilities, 50–79%). The least prevalent applications methods were: steam inhalation, compresses, wound-dressings, full body massage and baths (<10 facilities, <36%).

One application method significantly correlated with ‘perception of efficacy’ variable for dementia symptoms which was the use of aromatherapy spray (Pearson’s = 0.516, two tailed sig. = 0.008, n = 25).
Essential oil usage

The list of oils in the survey was generated by a focus group of aged care nursing staff as being likely to be used in aged care. Respondents were also asked to list any other oils they used that were not on the list. The only oil that was used by every facility was lavender (*Lavandula angustifolia*). Tea tree (*Melaleuca alternifolia*) oil was next most widely used (75% of facilities). Table 3 lists the oils used and the percentage of facilities using that aromatherapy oil. Aromatherapy oils were listed in the survey only as common names.

Table 3. Percentage of facilities using different essential oils

<table>
<thead>
<tr>
<th>≥50%</th>
<th>25–50%</th>
<th>&lt;25%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lavender (100%)</td>
<td>Cypress (39%)</td>
<td>Orange 21%</td>
</tr>
<tr>
<td>Tea tree (75%)</td>
<td>Roman chamomile (39%)</td>
<td>Myrrh 18%</td>
</tr>
<tr>
<td>Bergamot (64%)</td>
<td>Basil (36%)</td>
<td>Peppermint 18%</td>
</tr>
<tr>
<td>Eucalyptus (64%)</td>
<td>Frankincense (36%)</td>
<td>Lemongrass 14%</td>
</tr>
<tr>
<td>Geranium (64%)</td>
<td>German chamomile (32%)</td>
<td>Melissa 14%</td>
</tr>
<tr>
<td>Ginger (61%)</td>
<td>Lemon (32%)</td>
<td>Ylang-ylang 14%</td>
</tr>
<tr>
<td>Rosemary (57%)</td>
<td>Lime (32%)</td>
<td>Cedarwood 11%</td>
</tr>
<tr>
<td>Clary sage (54%)</td>
<td>Sandalwood (29%)</td>
<td>Rosewood 11%</td>
</tr>
<tr>
<td>Sweet marjoram (54%)</td>
<td>Clove bud 7%</td>
<td></td>
</tr>
<tr>
<td>Black pepper (50%)</td>
<td>Grapefruit 7%</td>
<td></td>
</tr>
<tr>
<td>Juniper (50%)</td>
<td>Mandarin 7%</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Niaouli 7%</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Patchouli 7%</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Sage 7%</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Benzoin 4%</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Jasmine 4%</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Neroli 4%</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Pine 4%</td>
<td></td>
</tr>
</tbody>
</table>

Most readers familiar with the aromatherapy indications for the different oils will notice that there are several oils that would be useful for sedation, relief of anxiety and mood enhancement; for example: lavender, geranium, bergamot, clary sage and sweet marjoram. Others would be more useful for management of infections, and physical ailments, such as tea tree, eucalyptus, black pepper and juniper. The use of these oils by over 50% of the facilities suggests that aromatherapy is considered effective for behavioural and psychological symptoms and for physical ailments.
**Commercial blends used**

Fifteen of the 28 respondents 15/28 (54%) stated that their facilities used commercial aromatherapy blends. In each case, they used two or more different blends. There was no significant correlation between the number of oils used and the number of blends used. The facility with the lowest usage of individual oils (only lavender oil) used two blends, a calming and a joint relief blend. Blend types were categorised as used for: calming; sleep promoting; arthritis management; mood uplifting or nurturing; for respiratory ailments; for oedema management.

**Health conditions for which aromatherapy is used**

Respondents were also asked to indicate the primary conditions for which they used the oils. As shown in Table 4, of the 463 responses from the 28 surveys, 46% related to behavioural and psychological symptoms, of which agitation and anxiety type behaviours were the most prevalent. Management of arthritis symptoms and musculoskeletal discomfort were the next most prevalent conditions, followed by management of infections, mainly respiratory diseases and topical fungal infections. Although the listed behavioural and psychological conditions correspond well with symptoms commonly found in residents with dementia, survey respondents did not usually distinguish whether they thought the symptom was due to dementia.

<table>
<thead>
<tr>
<th>Table 4. The conditions that essential oils are used for, total number of responses = 463, from 28 surveys.</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Conditions that oils are used for</th>
<th>% of responses</th>
</tr>
</thead>
<tbody>
<tr>
<td>Behavioural and psychological symptoms (46%)</td>
<td></td>
</tr>
<tr>
<td>Agitation, anxiety, restlessness, aggression, tension, stress, lack of calmness</td>
<td>25</td>
</tr>
<tr>
<td>Depression, withdrawal, apathy, grief, moodiness</td>
<td>12</td>
</tr>
<tr>
<td>Confusion, concentration, poor memory, 'mind and spirit'</td>
<td>5</td>
</tr>
<tr>
<td>Insomnia</td>
<td>3</td>
</tr>
<tr>
<td>Physical ailments (54%)</td>
<td></td>
</tr>
<tr>
<td>Arthritis, rheumatism, joints, muscles, aches, pains, stiffness</td>
<td>21</td>
</tr>
<tr>
<td>Infections-respiratory, fungal infections, viral infections, wound healing</td>
<td>16</td>
</tr>
<tr>
<td>Skin-integrity, dryness, itch, care</td>
<td>6</td>
</tr>
<tr>
<td>Poor circulation, oedema, fluid retention, altering blood pressure</td>
<td>4</td>
</tr>
<tr>
<td>Digestive system-constipation, indigestion</td>
<td>4</td>
</tr>
<tr>
<td>Other ailments-emphysema, diabetes, hormonal imbalance, palliation</td>
<td>3</td>
</tr>
</tbody>
</table>
Most of the oils listed were recommended for several different conditions, including behavioural or psychological symptoms and physical symptoms. The seven oils recommended for only physical conditions were: black pepper, eucalyptus, ginger, juniper, lemongrass, peppermint and tea tree. All the other oils had at least one recommendation for use with a behavioural or psychological symptom. Conditions for which commercial blends are used are shown in Table 5. Some blends were used for more than one condition. As with the essential oils, calming and relaxing blends are considered to be different than blends to aid sleep.

Table 5. Conditions that blends are used for. Total number of facilities that use blends = 15, total number of responses = 69. The missing 1% is due to a blend used for ‘therapist balance’.

<table>
<thead>
<tr>
<th>Conditions that blends are used for</th>
<th>% of responses</th>
</tr>
</thead>
<tbody>
<tr>
<td>Behavioural or psychological symptoms (61%)</td>
<td></td>
</tr>
<tr>
<td>Agitation, anxiety, restlessness, aggression, tension, lack of calmness</td>
<td>23</td>
</tr>
<tr>
<td>Depression, grief, emotional imbalance</td>
<td>16</td>
</tr>
<tr>
<td>Confusion, concentration, poor memory,</td>
<td>–</td>
</tr>
<tr>
<td>Insomnia</td>
<td>22</td>
</tr>
<tr>
<td>Physical ailments (38%)</td>
<td></td>
</tr>
<tr>
<td>Arthritis, rheumatism, joints, muscles, aches, pains, stiffness</td>
<td>25</td>
</tr>
<tr>
<td>Infections - respiratory, fungal infections, viral infections, wound healing</td>
<td>6</td>
</tr>
<tr>
<td>Skin-integrity, dryness, itch, care</td>
<td>3</td>
</tr>
<tr>
<td>Poor circulation, oedema, fluid retention, altering blood pressure</td>
<td>4</td>
</tr>
<tr>
<td>Digestive system - constipation, indigestion</td>
<td>–</td>
</tr>
<tr>
<td>Other ailments - emphysema, diabetes, hormonal imbalance, palliation</td>
<td></td>
</tr>
</tbody>
</table>

Discussion

Variations in aromatherapy practices

The main outcome of this survey has been to highlight the lack of homogeneity of aromatherapy practices in aged care, which suggests that there has yet to be a ‘best practice’ protocol developed. The survey also demonstrates that aromatherapy is used quite widely in residential aged care facilities for people with and without dementia and that it is relatively inexpensive (average monthly cost of AUD4.50 per person).

The perception by the directors of care that aromatherapy use moderately reduces use of pharmaceuticals (particularly sedatives) indicates the potential economic value of
aromatherapy in aged care. Cost effectiveness of aromatherapy in aged care should be further investigated by health care economists.

Foothaths and massage of hands, feet and limbs were most commonly used, probably because they are easy and relatively non-invasive application methods. Some of the least used methods are more likely to take longer or be impractical in an institutional setting (for example steam inhalation and full body massage). However, the most effective delivery route for maximisation of the pharmacological effects of essential oils is yet to be established.

Interestingly, only half the facilities used essential oils in a vaporiser in the public areas of the facility. This could be due to a number of reasons. These could range from the possibility that aromas can be offensive to some people through to the rationale that if the essential oils are being used as therapeutic agents, it may be unethical to have them present in the air for people for whom they have not been prescribed, including staff and visitors. The latter reason underlines the need for establishment of the pharmacological effects of essential oils.

One of the issues facing aromatherapy as a profession is the issue of effective dosage. We chose not to collect data about the concentration of oils used in the various application methods, mainly because at this stage there is no established proof of effective dosage for essential oils for different conditions, and the difficulty of trying to record the information for each different application method, and for each varying condition would have proved too time-consuming for this pilot survey. However, we feel it is important to highlight the fact that 47% of residents receive aromatherapy treatments every day, usually 1–2 times a day, or as needed. This frequency of application suggests that aromatherapy may be being used in a similar way to other types of medication that aim to keep blood-levels of specific drug substances within a therapeutic range.

Given the oils used and conditions they were used for, this study confirms that aromatherapy is being used for a mixture of behavioural, psychological and physical symptoms. Aromatherapy appears to be most effective for management of agitation, anxiety, nervous tension, depression and sleeplessness. It is also used widely for
management of physical pain and infections. Resolving pain and infection may also help reduce behavioural and psychological symptoms.

Commercial blends, although not often recommended by aromatherapists in general practice, appear to be useful for arthritic pain, agitation and anxiety, sleeplessness and depression. In a busy aged care facility, using blends may be the answer to time pressures, although many aromatherapists would still prefer to prepare an individualised blend for each person.

**How effective is aromatherapy perceived to be for dementia?**

The data relating to perceptions of efficacy of aromatherapy had mostly bi-modal distribution. This implies that there are care planners who perceive aromatherapy to be very effective for various symptoms of dementia, and others who perceive it to be less effective, with not much of a middle ground. This in itself is unexpected, especially as there are no correlations with the percentage of residents being treated or percentage of residents with dementia. The propensity to perceive aromatherapy as effective may relate more to optimistic or enthusiastic character traits of the care planners.

**Correlations of perceptions of efficacy with aromatherapy practices**

Use of factor analysis to derive the new variable related to ‘perceptions of efficacy’ for dementia symptoms is a fairly standard statistical procedure. However, given the fact that the new variable only correlated significantly with one item of aromatherapy practice (use of sprays), there are two possibilities that arise: firstly that the new variable was not helpful as a summary of opinions about efficacy for dementia; secondly that the varieties of aromatherapy practice really do not correlate with perceptions of efficacy. Given the individuality of human perceptions, the second possibility is more likely. In retrospect, additional survey questions could have more directly revealed correlations between perceptions of efficacy and aromatherapy practices.

**Aromatherapy practices and dementia management in aged care facilities**

A comprehensive comment about aromatherapy practices for dementia management involves combining the findings from a number of components of the survey.
Aromatherapy is used frequently for dementia residents – 75% of residents with dementia receive >3 aromatherapy treatments per week, compared with 25% that receive it less frequently. We presume that aromatherapy is perceived to be effective enough to justify the cost of frequent treatment.

Aromatherapy is seen as a more useful treatment for dementia residents than non-dementia residents – 14/28 facilities (50%) give 100% of their total dementia residents aromatherapy, whereas only 9/28 facilities (32%) give 100% of their total non-dementia residents aromatherapy.

Reduction of sedatives by use of aromatherapy is reported by more directors of care than reduction of other classes of drugs. Sedatives are used to manage agitation, anxiety and aggression in dementia (Borson and Raskind, 1997). This result suggests a potential role for aromatherapy in dementia care.

The conditions that care planners are using essential oils for include many behavioural and psychological symptoms of dementia such as agitation, anxiety, sleeplessness and depression.

Considering these findings, it appears as though aromatherapy is being used extensively in aged care facilities to manage symptoms of dementia. It is clear from this survey however, that further research is required to determine a ‘best practice’ for aromatherapy in dementia and aged care.

**Conclusions**

The results of this survey indicate that aromatherapy warrants further investigation as a complementary therapy in residential aged care. The percentage of treated residents represented by the survey is large, and it is perceived as an effective therapy for agitation and anxiety and arthritic pain, all common ailments in ageing residents. In particular, it appears to be worth investigating the extent of pharmaceutical cost reduction in facilities using aromatherapy. Possible future research projects include:

- Objective evaluation of aromatherapy for both psychological and physical sources of discomfort in aged care residents.
• Determination of ‘best practice’ for use of aromatherapy in aged care, especially for residents with dementia, including most effective application methods and dosage regimes.
• Prospective evaluation of the use of sedatives in the presence and absence of aromatherapy.
• Quantitative evaluation of pharmaceutical cost-savings in facilities that use aromatherapy.
• Evaluation of the effects of aromatherapy on everyone in the facility, not just residents.
• Determination of pharmacological effects of essential oils, whether physical or psychological.

Acknowledgements
We thank the Northern Rivers Aged Care Aromatherapy Network who assisted in the survey design and consulted on many issues prior to the survey. Special thanks go to all the survey participants who gave their time to complete the survey in the midst of their already busy schedules. We also thank Dr. Lyndon Brooks and Margaret Rolfe from Southern Cross University for their statistical analysis advice.

References (to published paper)


Appendix 2

*GC-MS Specifications for data in Table 6.5*

**GC Agilent 6890 specifications**: Oven initial temp 50° C, rate 4 degrees·min\(^{-1}\), oven final temp 350° C; Front inlet: split column, split ratio 300:1, initial temp 280° C, pressure 29.10 psi, split flow 402.9mL/min, total flow 407.6mL/min, Helium gas.

**Capillary column** SGE BPX50, max temp 350° C, length 50.0m, diameter 220.00µm, constant flow, initial flow 1.3mL/min, nominal init pressure 29.12 psi, average velocity 33 cm/sec, outlet MSD.

**Automatic injector Agilent 7683**: Sample washes (2), sample pumps (3), injection volume 0.1µL of neat oil, syringe size 5.0µL, post-injection Solvent A washes (4), plunger speed (slow).

**MS Agilent 4973 Network Mass Selective Detector Acquisition parameters**: Scan mode, solvent delay 0 min (no solvent used), EM Absolute (false), EM offset (0), Resulting EM voltage (1611.8); Scan parameters: Low mass 35.0, high mass 450.0, threshold 25, Sample # 2, A/D samples (4), plot 2 low mass 50.0, plot 2 high mass 550.0; MS zones: MS Quad 150° C max 200° C, MS Source 230° C, max 250° C.
### Appendix 2 Table 1

Table 1. Terpenoid inhibitor compounds in bovine or human erythrocyte acetylcholinesterase (Miyazawa & Yamafuji, 2005, Miyazawa et al., 1998, Miyazawa et al., 1997, Savelev et al., 2003, NS Perry et al., 2000).

<table>
<thead>
<tr>
<th>Compound</th>
<th>IC50 or % inhibition ± s.e.</th>
<th>IC50 in mM</th>
<th>First author/date</th>
<th>Enzyme source</th>
</tr>
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<tbody>
<tr>
<td>Viridiflorol</td>
<td>25 µg/mL</td>
<td>0.11mM</td>
<td>Miyazawa 1998</td>
<td>Bovine</td>
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<tr>
<td>Elemol</td>
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<td>Bovine</td>
</tr>
<tr>
<td>(+)-3-carene</td>
<td>-</td>
<td>0.20mM</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
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<tr>
<td>1,8-cineole</td>
<td>41 µg/mL</td>
<td>0.26mM</td>
<td>Miyazawa 1998</td>
<td>Bovine</td>
</tr>
<tr>
<td></td>
<td>60 µg/mL ± 10</td>
<td>0.38mM</td>
<td>Savelev 2003</td>
<td>Bovine</td>
</tr>
<tr>
<td></td>
<td>4.7mM 100%</td>
<td>0.67mM</td>
<td>Perry 2000</td>
<td>Human</td>
</tr>
<tr>
<td>(+)-piperitenone oxide</td>
<td>64µg/mL</td>
<td>0.45mM</td>
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<tr>
<td></td>
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<td>0.63mM</td>
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<td>Bovine</td>
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<tr>
<td>(+)-pulegone</td>
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<tr>
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<td>Bovine</td>
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<td>Bovine</td>
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<td>1.0mM</td>
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<td>Bovine</td>
</tr>
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<td>1.38mM</td>
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<tr>
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<td>Bovine</td>
</tr>
<tr>
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<tr>
<td></td>
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<tr>
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<td>Miyazawa 1998</td>
<td>Bovine</td>
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<td>Bovine</td>
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<td>&gt;2.0 mM</td>
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<td>Bovine</td>
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<tr>
<td>(-)-isopulegel</td>
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<tr>
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<td>Bovine</td>
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<tr>
<td>Terpinen-4-ol</td>
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<td>10.3 mM</td>
<td>Mills 2004</td>
<td>Eel</td>
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<tr>
<td>(-)-menthyl acetate</td>
<td>41µg/mL 35%</td>
<td>-</td>
<td>Miyazawa 1998</td>
<td>Bovine</td>
</tr>
<tr>
<td>(+)-menthofuran</td>
<td>82µg/mL 33%</td>
<td>-</td>
<td>Miyazawa 1998</td>
<td>Bovine</td>
</tr>
<tr>
<td>(-)-linalyl acetate</td>
<td>82µg/mL 38%</td>
<td>-</td>
<td>Miyazawa 1998</td>
<td>Bovine</td>
</tr>
<tr>
<td>(-)-carvone</td>
<td>164µg/mL 43%</td>
<td>-</td>
<td>Miyazawa 1998</td>
<td>Bovine</td>
</tr>
<tr>
<td>(-)-borneol</td>
<td>250 µg/mL 19% 1.6mM</td>
<td>-</td>
<td>Savelev 2003</td>
<td>Bovine</td>
</tr>
<tr>
<td></td>
<td>1.0mM 22.6%</td>
<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
</tr>
<tr>
<td>(+)-borneol</td>
<td>1.0mM 22.2%</td>
<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
</tr>
<tr>
<td>Bornyl acetate</td>
<td>250 µg/mL 23%</td>
<td>-</td>
<td>Savelev 2003</td>
<td>Bovine</td>
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<tr>
<td>Caryophyllene oxide</td>
<td>250 µg/mL 35%</td>
<td>-</td>
<td>Savelev 2003</td>
<td>Bovine</td>
</tr>
<tr>
<td>Gamma-terpinene</td>
<td>4.7mM 29.6%</td>
<td>-</td>
<td>Perry 2000</td>
<td>Human</td>
</tr>
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<td>Linalool</td>
<td>500 µg/mL 18%</td>
<td>-</td>
<td>Savelev 2003</td>
<td>Bovine</td>
</tr>
<tr>
<td></td>
<td>IC90&gt;5M; 12% 4.7mM</td>
<td>-</td>
<td>Perry 2000</td>
<td>Human</td>
</tr>
<tr>
<td>(+)-camphor</td>
<td>500 µg/mL 39% 3.3mM</td>
<td>-</td>
<td>Savelev 2003</td>
<td>Bovine</td>
</tr>
<tr>
<td></td>
<td>IC90&gt;10M; 4.7mM 27%</td>
<td>-</td>
<td>Perry 2000</td>
<td>Human</td>
</tr>
<tr>
<td></td>
<td>1.0mM 26.4%</td>
<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
</tr>
<tr>
<td>(-)-camphor</td>
<td>1.0mM 21.2%</td>
<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
</tr>
<tr>
<td>(+)-cis-verbenol</td>
<td>1.0mM 17%</td>
<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
</tr>
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<td>(-)-myrenol</td>
<td>1.0M15%</td>
<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
</tr>
<tr>
<td>(+)-trans-myrtanol</td>
<td>1.0mM 37.1%</td>
<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
</tr>
<tr>
<td>(-)-trans-myrtanol</td>
<td>1.0mM 37.4%</td>
<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
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<tr>
<td>Thymol</td>
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<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
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<tr>
<td>(+)-fenchol</td>
<td>1.0mM 37.7%</td>
<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
</tr>
<tr>
<td>(+)-fenchone</td>
<td>1.0mM 23.3%</td>
<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
</tr>
<tr>
<td>(-)-fenchone</td>
<td>1.0mM 28.2%</td>
<td>-</td>
<td>Miyazawa 2005</td>
<td>Bovine</td>
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</table>
Table 2. Dose-dependent inhibition of bovine AChE by essential oils. Mean IC$_{50}$ and percentage inhibition of control at 0.454mg/ml final well concentration for all samples showing dose-dependent inhibition, from most potent to least potent.

<table>
<thead>
<tr>
<th>Expt #</th>
<th>Oil name</th>
<th>Mean IC$_{50}$ mg/mL</th>
<th>IC$_{50}$ Inter-plate sd.</th>
<th>Replicates (n x plate)</th>
<th>Mean inhibition at 0.454 mg/mL</th>
<th>% at Inter-plate St. dev (%)</th>
</tr>
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<tbody>
<tr>
<td>15</td>
<td>Galantamine</td>
<td>0.007</td>
<td>0.001</td>
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<td>78.4*</td>
<td>4.0</td>
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<td>16</td>
<td>Galantamine</td>
<td>0.010</td>
<td></td>
<td>3x1</td>
<td>70.9*</td>
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<td>10</td>
<td>Lime</td>
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<td>0.010</td>
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<td>0.012</td>
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<td>Salvia officinalis</td>
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<td>0.019</td>
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<td>3.6</td>
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<td>Blend 1 (Treatment A)</td>
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<td>0.078</td>
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<td>53.9</td>
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<td>Sage</td>
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<td>Sage</td>
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<td>0.012</td>
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<td>48.9</td>
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<tr>
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<td>Cypress (linear equation)</td>
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<td>0.033</td>
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<td>Cineole</td>
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<td>3x3</td>
<td>52.9</td>
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<td>48.2</td>
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<td>42.3</td>
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<td>46.8</td>
<td>2.4</td>
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<td>Lavender Sage 3</td>
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<td>0.406</td>
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* Galantamine % inhibition = 0.036 mg/mL
Appendix 3

Demographic results

Table 1. Age, weight and length of residence in facility, sorted by treatment type, n=72.

<table>
<thead>
<tr>
<th>Demographic</th>
<th>Treatment Group</th>
<th>N</th>
<th>Mean</th>
<th>Std. Dev.</th>
<th>Minimum</th>
<th>Maximum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient age at April 05 (years)</td>
<td>Control</td>
<td>23</td>
<td>82.78</td>
<td>6.237</td>
<td>68</td>
<td>92</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>27</td>
<td>86.30</td>
<td>6.661</td>
<td>70</td>
<td>96</td>
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<tr>
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<td>B</td>
<td>22</td>
<td>86.55</td>
<td>5.492</td>
<td>74</td>
<td>97</td>
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<td>Patient weight at last measurement (kg)</td>
<td>Control</td>
<td>22*</td>
<td>63.55</td>
<td>14.893</td>
<td>35</td>
<td>89</td>
</tr>
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<td></td>
<td>A</td>
<td>25*</td>
<td>61.48</td>
<td>15.172</td>
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<td>88</td>
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<tr>
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<td>B</td>
<td>21*</td>
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<td>12.980</td>
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<td>85</td>
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<tr>
<td>Number of months resident in facility at April 05</td>
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<td>35.59</td>
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</table>

Some data were missing from certain residents’ files.

Table 2. Patient gender by treatment group, n=72.

<table>
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<th>Treatment type</th>
<th>Female</th>
<th>Percent</th>
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<td>78.3</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>21.7</td>
</tr>
<tr>
<td>Total</td>
<td>23</td>
<td>100.0</td>
</tr>
<tr>
<td>A</td>
<td>21</td>
<td>77.8</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>22.2</td>
</tr>
<tr>
<td>Total</td>
<td>27</td>
<td>100.0</td>
</tr>
<tr>
<td>B</td>
<td>19</td>
<td>86.4</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>13.6</td>
</tr>
<tr>
<td>Total</td>
<td>22</td>
<td>100.0</td>
</tr>
</tbody>
</table>

Table 3. Presence of conditions indicative of illness status and possibly able to affect dementia status or response to study treatments.

<table>
<thead>
<tr>
<th>Condition</th>
<th>Treatment type</th>
<th>Frequency</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hypertension</td>
<td>Control</td>
<td>6</td>
<td>26.1</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>10</td>
<td>37.0</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>8</td>
<td>36.4</td>
</tr>
<tr>
<td>Cancer</td>
<td>Control</td>
<td>2</td>
<td>8.7</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>2</td>
<td>7.4</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>3</td>
<td>13.6</td>
</tr>
<tr>
<td>Diabetes</td>
<td>Control</td>
<td>2</td>
<td>8.7</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>2</td>
<td>7.4</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>6</td>
<td>27.3</td>
</tr>
<tr>
<td>Asthma/Hay fever</td>
<td>Control</td>
<td>5</td>
<td>21.7</td>
</tr>
<tr>
<td>Condition</td>
<td>Treatment type</td>
<td>Frequency</td>
<td>Percent</td>
</tr>
<tr>
<td>-----------------</td>
<td>----------------</td>
<td>-----------</td>
<td>---------</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>3</td>
<td>11.1</td>
</tr>
<tr>
<td>Arthritis</td>
<td>Control</td>
<td>6</td>
<td>26.1</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>7</td>
<td>25.9</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>12</td>
<td>54.5</td>
</tr>
</tbody>
</table>

Table 4. Mean number of concurrent medications by treatment type.

<table>
<thead>
<tr>
<th>Treatment group</th>
<th>N</th>
<th>Mean</th>
<th>Std. Dev.</th>
<th>Minimum</th>
<th>Maximum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>22</td>
<td>9.32</td>
<td>3.747</td>
<td>3</td>
<td>18</td>
</tr>
<tr>
<td>A</td>
<td>27</td>
<td>7.93</td>
<td>3.862</td>
<td>0</td>
<td>15</td>
</tr>
<tr>
<td>B</td>
<td>22</td>
<td>8.86</td>
<td>3.720</td>
<td>3</td>
<td>15</td>
</tr>
</tbody>
</table>

Table 5. Prevalence of different medications in the treatment groups, n=72.

<table>
<thead>
<tr>
<th>Medication</th>
<th>Treatment group</th>
<th>Frequency</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hypertension</td>
<td>Control</td>
<td>16</td>
<td>69.6</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>16</td>
<td>59.3</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>12</td>
<td>54.5</td>
</tr>
<tr>
<td>Heart medications (includes arrhythmia, angina)</td>
<td>Control</td>
<td>7</td>
<td>30.4</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>12</td>
<td>44.4</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>7</td>
<td>31.8</td>
</tr>
<tr>
<td>Stroke prevention (includes aspirin and anticoagulants)</td>
<td>Control</td>
<td>11</td>
<td>47.8</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>16</td>
<td>59.3</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>10</td>
<td>45.5</td>
</tr>
<tr>
<td>Cholesterol lowering meds</td>
<td>Control</td>
<td>1</td>
<td>4.3</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>5</td>
<td>18.5</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>2</td>
<td>9.1</td>
</tr>
<tr>
<td>Anti-depressants</td>
<td>Control</td>
<td>7</td>
<td>30.4</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>10</td>
<td>37.0</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>8</td>
<td>36.4</td>
</tr>
<tr>
<td>Anxiolytics</td>
<td>Control</td>
<td>5</td>
<td>21.7</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>2</td>
<td>7.4</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>2</td>
<td>9.1</td>
</tr>
<tr>
<td>Anti-psychotics</td>
<td>Control</td>
<td>4</td>
<td>17.4</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>4</td>
<td>14.8</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>2</td>
<td>9.1</td>
</tr>
<tr>
<td>Anti-epilepsy medications</td>
<td>Control</td>
<td>3</td>
<td>13.0</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>2</td>
<td>7.4</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>2</td>
<td>9.1</td>
</tr>
<tr>
<td>Insomnia medication</td>
<td>Control</td>
<td>4</td>
<td>17.4</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>4</td>
<td>14.8</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>5</td>
<td>22.7</td>
</tr>
<tr>
<td>Analgesia</td>
<td>Control</td>
<td>15</td>
<td>65.2</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>19</td>
<td>70.4</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>17</td>
<td>77.3</td>
</tr>
<tr>
<td>Laxative/haemorrhoid medications</td>
<td>Control</td>
<td>8</td>
<td>34.8</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>15</td>
<td>55.6</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>9</td>
<td>40.9</td>
</tr>
<tr>
<td>Medication</td>
<td>Treatment group</td>
<td>Frequency</td>
<td>Percent</td>
</tr>
<tr>
<td>------------------------------------</td>
<td>----------------</td>
<td>-----------</td>
<td>---------</td>
</tr>
<tr>
<td>Hypothyroid medications</td>
<td>Control</td>
<td>2</td>
<td>8.7</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>2</td>
<td>7.4</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>5</td>
<td>22.7</td>
</tr>
<tr>
<td>B12 injection &amp; folate supplement</td>
<td>Control</td>
<td>4</td>
<td>17.4</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>3</td>
<td>11.1</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>4</td>
<td>18.2</td>
</tr>
</tbody>
</table>

Table 6. Education level attained, n=72.

<table>
<thead>
<tr>
<th>Education level attained</th>
<th>Frequency</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Primary school (up to age 12)</td>
<td>14</td>
<td>19.4</td>
</tr>
<tr>
<td>Secondary school (up to age 15)</td>
<td>54</td>
<td>75.0</td>
</tr>
<tr>
<td>Tertiary or trade education</td>
<td>3</td>
<td>4.2</td>
</tr>
<tr>
<td>Don't know</td>
<td>1</td>
<td>1.4</td>
</tr>
<tr>
<td>Total</td>
<td>72</td>
<td>100.0</td>
</tr>
</tbody>
</table>

Table 7. Smoking history descriptive statistics n=72.

<table>
<thead>
<tr>
<th>Smoking history</th>
<th>Frequency</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Never smoked</td>
<td>49</td>
<td>68.1</td>
</tr>
<tr>
<td>Used to smoke</td>
<td>20</td>
<td>27.8</td>
</tr>
<tr>
<td>Smokes daily</td>
<td>3</td>
<td>4.2</td>
</tr>
<tr>
<td>Total</td>
<td>72</td>
<td>100.0</td>
</tr>
</tbody>
</table>
# MMSE form used in the trial

## Mini-Mental State Examination


<table>
<thead>
<tr>
<th>Question</th>
<th>Max Score</th>
<th>Comments</th>
<th>Time</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>1a What year is this?</td>
<td>1</td>
<td>accept exact answer only</td>
<td>10 s</td>
<td></td>
</tr>
<tr>
<td>b What season is this?</td>
<td>1</td>
<td>during first or last week of season</td>
<td>10 s</td>
<td></td>
</tr>
<tr>
<td>c What month of the year is this?</td>
<td>1</td>
<td>on first day of new month or last day of previous month accept either</td>
<td>10 s</td>
<td></td>
</tr>
<tr>
<td>d What is today’s date?</td>
<td>1</td>
<td>accept previous or next date as well as current date</td>
<td>10 s</td>
<td></td>
</tr>
<tr>
<td>e What day of the week is this?</td>
<td>1</td>
<td>accept exact answer only</td>
<td>10 s</td>
<td></td>
</tr>
<tr>
<td>2a What country are we in?</td>
<td>1</td>
<td>accept exact answer only</td>
<td>10 s</td>
<td></td>
</tr>
<tr>
<td>b What state are we in?</td>
<td>1</td>
<td>accept exact answer only</td>
<td>10 s</td>
<td></td>
</tr>
<tr>
<td>c What town are we in?</td>
<td>1</td>
<td>accept exact answer only</td>
<td>10 s</td>
<td></td>
</tr>
<tr>
<td>d What is the name of this nursing home or hostel?</td>
<td>1</td>
<td>accept exact answer only</td>
<td>10 s</td>
<td></td>
</tr>
<tr>
<td>e What floor of the building are we on?</td>
<td>1</td>
<td>accept exact answer only</td>
<td>10 s</td>
<td></td>
</tr>
<tr>
<td>3 “I am going to name 3 objects. After I have said all three objects, I want you to repeat them. Remember what they are because I am going to ask you to name them again in a few minutes. Ball (1 second) Car (1 second) Man (1 second). Please repeat the 3 items for me.”</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3a Ball</td>
<td>1</td>
<td>Score 1 point for correct answer on first attempt</td>
<td>20 s</td>
<td></td>
</tr>
<tr>
<td>b Car</td>
<td>1</td>
<td>Score 1 point for correct answer on first attempt</td>
<td>20 s</td>
<td></td>
</tr>
<tr>
<td>c Man</td>
<td>1</td>
<td>Score 1 point for correct answer on first attempt</td>
<td>20 s</td>
<td></td>
</tr>
<tr>
<td>4 “Spell the word WORLD” (you may help the subject spell WORLD correctly)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4a “Now spell it backwards please.”</td>
<td>5</td>
<td>If subject cannot spell WORLD even with assistance score 0.</td>
<td>30 s</td>
<td></td>
</tr>
<tr>
<td>Question</td>
<td>Correct Answer</td>
<td>Score</td>
<td>Score Criteria</td>
<td>Time</td>
</tr>
<tr>
<td>-------------------------------------------------------------------------</td>
<td>----------------</td>
<td>-------</td>
<td>----------------------------------------------------</td>
<td>------</td>
</tr>
<tr>
<td>5 “Now what were the 3 objects that I asked you to remember?”</td>
<td>Ball</td>
<td>1</td>
<td>Score 1 point regardless of order</td>
<td>10 s</td>
</tr>
<tr>
<td>5b Car</td>
<td>1 Score 1 point regardless of order</td>
<td>10 s</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5c Man</td>
<td>1 Score 1 point regardless of order</td>
<td>10 s</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6 Show wristwatch. Ask <strong>What is this called?”</strong></td>
<td>Wristwatch, or watch. Do not accept clock, time etc.</td>
<td>10 s</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7 Show pencil. Ask <strong>And what is this called?”</strong></td>
<td>Pencil only, score 0 for pen.</td>
<td>10 s</td>
<td></td>
<td></td>
</tr>
<tr>
<td>8 I’d like you to repeat a phrase after me: “No if’s, and’s or but’s”</td>
<td>Accept exact repetition only - score 0 for “no if’s or but’s”</td>
<td>10 s</td>
<td></td>
<td></td>
</tr>
<tr>
<td>9 “Read the words on this page and then do what it says.” (Hand subject the laminated sheet with CLOSE YOUR EYES on it)</td>
<td>If subject just reads and does not close eyes, repeat script up to 3 times. Score 1 point if subject closes eyes - does not have to read aloud.</td>
<td>10 s</td>
<td></td>
<td></td>
</tr>
<tr>
<td>10 “Are you right or left handed?” (Use opposite hand in statement. Take a piece of paper - hold it up in front of the subject and say: “Take this paper in your left/right hand, fold the paper in half once with both hands and put the paper down on the floor.”)</td>
<td>Takes paper in correct hand</td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>10a Takes paper in correct hand</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10b Folds it in half</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10c Puts it on floor</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>11 Hand subject a pencil and paper. Say <strong>Write any sentence on that piece of paper</strong>.</td>
<td>Sentence should make sense, ignore spelling errors.</td>
<td>30 s</td>
<td></td>
<td></td>
</tr>
<tr>
<td>12 Place design, pencil, eraser and paper in front of the subject. Say <strong>Copy this design please</strong>.</td>
<td>Subject must have drawn a 4-sided figure between two 5-sided figures. Allow multiple tries until patient is finished and hands it back.</td>
<td>60 s</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**TOTALS**
NOSGER scale used in trial

<table>
<thead>
<tr>
<th>Score sheet</th>
<th>Patient Code</th>
<th>Date</th>
<th>Shift (M, A, E)*</th>
<th>Staff initials</th>
</tr>
</thead>
</table>

We are interested in finding out how this patient has been doing in the LAST TWO WEEKS. For this purpose there are 30 statements which you should grade according to your own observations. Read each statement and assess the patient’s behaviour by marking the box which corresponds the best.

<table>
<thead>
<tr>
<th></th>
<th>All the time</th>
<th>Most of the time</th>
<th>Often</th>
<th>Sometimes</th>
<th>Never</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Shaves or puts on make-up, combs hair without help</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2.</td>
<td>Follows favourite radio or TV programmes</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3.</td>
<td>Reports she/he feels sad.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4.</td>
<td>Is restless during the night.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5.</td>
<td>Is interested in what is going on around her/him.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6.</td>
<td>Tries to keep her/his room tidy.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7.</td>
<td>Is able to control bowels.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8.</td>
<td>Remembers a point in conversation after interruption.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>9.</td>
<td>Goes shopping for small items (newspaper, groceries).</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10.</td>
<td>Reports feeling worthless.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>11.</td>
<td>Continues with some favorite hobby.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>12.</td>
<td>Repeats the same point in conversation over and over.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>13.</td>
<td>Appears sad or tearful.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>15.</td>
<td>Runs away, or tries to.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>16.</td>
<td>Remembers names of close friends.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17.</td>
<td>Helps others as far as physically able.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>18.</td>
<td>Goes out (of own room) inappropriately dressed.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>19.</td>
<td>Is orientated when in usual surroundings.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>20.</td>
<td>When asked questions, seems quarrelsome and irritable.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>21.</td>
<td>Makes contact with people around.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>22.</td>
<td>Remembers where clothes and other things are placed.</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>23.</td>
<td>Is aggressive (verbally or physically).</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>24.</td>
<td>Is able to control bladder function (urine).</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
**NURSES’ OBSERVATION SCALE FOR GERIATRIC PATIENTS (NOSGER)**

<table>
<thead>
<tr>
<th>Patient Code</th>
<th>Date</th>
</tr>
</thead>
<tbody>
<tr>
<td>25.</td>
<td>Appears to be cheerful.</td>
</tr>
<tr>
<td>26.</td>
<td>Maintains contact with friends or family.</td>
</tr>
<tr>
<td>27.</td>
<td>Confuses the identity of some people with others.</td>
</tr>
<tr>
<td>28.</td>
<td>Enjoys certain events (visits, parties).</td>
</tr>
<tr>
<td>29.</td>
<td>Appears friendly and positive in conversation with family members or friends.</td>
</tr>
<tr>
<td>30.</td>
<td>Behaves stubbornly, does not follow instructions or rules.</td>
</tr>
</tbody>
</table>

Comments about any item that needs explanation:

---

**NOSGER statistical calculations**

It was hypothesised that NOSGER scores for people in the Treatment A treatment group would decrease (i.e. an improvement), and that scores for people on the other two lotions would either stay the same or increase over the treatment period.

Using the same multi-level model as used for the MMSE scores, the levels of random variation were Facility, Participants within facility and Observations within participant. The variation due to Facility was not significant, but the variation due to both ‘Participant within facility’ and ‘Observation within participant’ was significant ($\alpha=0.05$). Comparison of the -2LL estimates revealed no significant difference between Treatment type or Occasion (of measurement).

**NOSGER multi-level model results**

**NOSGER: Variance components**

The same multi-level repeated measures model was generated by MLwiN, giving the equation shown in Figure 1.

Figure 1. **NOSGER: variance components. Variables:** $v_{kl}$ = Facility; $u_{ijk}$ = Participants within facility; $e_{ijk}$ = Observations within participant.
Variance due to Facility was not significant, $z = \frac{52.129}{41.188} = 1.266$ ($\alpha = 0.05$).
Variance due to Participants within facility was significant, $z = \frac{168.600}{48.460} = 3.479$, and also variance due to Observations within participant was significant, $z = \frac{288.134}{33.957} = 8.485$.

The following fixed effects variables were added to the model:

- Treatment type
- Occasion (of measurement)

**NOSGER: Treatment effects**

Treatment type was the first fixed effect variable added to the model (see Figure 2).

**Figure 2. NOSGER: Treatment effects, dummy variables = (T).**

The difference in -2LL deviances between the initial equation and this one was 1916.602-1915.296 = 1.306. This was not significant by chi-squared test ($\alpha = 0.05$, df=2, $p=0.520$).
The next level of the model included Treatment type and Occasion (see Figure 3).

Figure 3. NOSGER: Treatment type (T) + Occasion (O).

\[
\text{NOSGER}_{ijk} \sim N(\mathbf{XB}, \Omega)
\]

\[
\text{NOSGER}_{ijk} = \beta_0 + \mathbf{CONS} + 5.150(4.689)T_1 + 3.898(4.929)T_2 + 0.111(2.818)O + -4.306(2.818)O2 +
\]

\[
\beta_{ijk} = 68.043(4.478) + \nu_{ijk} + \mu_{ijk} + \psi_{ijk}
\]

\[
\begin{bmatrix} \nu_{ijk} \end{bmatrix} \sim N(0, \Omega_\nu) : \Omega_\nu = \begin{bmatrix} 48.208(39.722) \end{bmatrix}
\]

\[
\begin{bmatrix} \mu_{ijk} \end{bmatrix} \sim N(0, \Omega_\mu) : \Omega_\mu = \begin{bmatrix} 173.813(49.210) \end{bmatrix}
\]

\[
\begin{bmatrix} \epsilon_{ijk} \end{bmatrix} \sim N(0, \Omega_\epsilon) : \Omega_\epsilon = \begin{bmatrix} 285.793(33.957) \end{bmatrix}
\]

\[-2*\text{loglikelihood(IGLS Deviance)} = 1912.103(216 of 216 cases in use)\]

Again the difference in -2LL deviances was compared with the initial equation, 1915.296 – 1912.103 = 3.193. A chi-squared test revealed that Occasion also did not contribute significantly to the variance in NOSGER scores (\(\alpha = 0.05, \text{df}=2, p=0.203\)).

As neither Treatment type nor Occasion contributed significantly to the variance in NOSGER scores, no further levels of the model were pursued. The variance in NOSGER scores came from within individuals and observations within individuals, or from unmeasured variables.
References


Access-Economics (2003), *The dementia epidemic: economic impact and positive solutions for Australia*, Access Economics Pty Ltd, Canberra, ACT.


Jorm, AF (2001), Dementia: a major health problem for Australia (Position Paper 1), Alzheimer's Association of Australia, Higgins, ACT.


Survey information sheets, consent forms and questionnaire
DON PARTICIPANT INFORMATION SHEET

Survey of Aromatherapy Use in Northern Rivers Residential Aged Care Facilities
Southern Cross University and the Australian Centre for Complementary Medicine, Education and Research (ACCMER) invite you to participate in a study of aromatherapy use in residential aged care facilities in the Northern Rivers area. Of particular interest to the researchers are the effects of aromatherapy on residents with dementia. Participation is voluntary, and you may withdraw or discontinue at any time up until the survey is added to the anonymous pool of surveys.

Who is conducting the study?
Ms Joy Bowles (primary researcher)
Professor Stephen Myers (supervisor)

What am I required to do?
During the visit by the primary researcher (Joy Bowles), you will be asked to complete the attached survey in blue or black ink, and return it to Joy at the end of her visit. It will take approximately 10 minutes to complete. If you don't complete it while Joy is present, please return the completed survey in the stamped addressed envelope supplied, within 2 weeks. The return address is listed at the end of the survey.

Are there any risks?
There are no risks other than the possible inconvenience of the time required to complete the survey.

How will the privacy and confidentiality of my information be protected?
Your name and the name of the facility will at no time be connected with the completed survey. Once collected, the survey will be anonymous. No data will be reported that identifies your name or the facility's name or geographical location apart from the fact that it was from the Northern Rivers Area.

How can I make further inquiries about this study?
If you have any questions or concerns about this study you can contact any one of the following:
Ms Joy Bowles (primary researcher), Australian Centre for Complementary Medicine, Education & Research, School of Natural & Complementary Medicine, Southern Cross University, Lismore.
Phone: 02 6626 9338, Email: ebowle10@scu.edu.au

Professor Stephen Myers (supervisor), Australian Centre for Complementary Medicine, Education & Research, School of Natural & Complementary Medicine, Southern Cross University, Lismore.
Phone: 02 6620 3403, Email: smyers@scu.edu.au

If you have any questions that cannot be answered by these people, please contact:
Mr John Russell, Administrative Officer, Graduate Research College, Southern Cross University
Phone: 02 6620 3705, Email: jrussell@scu.edu.au
Southern Cross University
&
Australian Centre for Complementary Medicine, Education & Research

AROMATHERAPIST PARTICIPANT INFORMATION SHEET

Survey of Aromatherapy Use in Northern Rivers Residential Aged Care Facilities
Southern Cross University and the Australian Centre for Complementary Medicine, Education and Research (ACCMER) invite you to participate in a study of aromatherapy use in residential aged care facilities in the Northern Rivers area. Of particular interest to the researchers are the effects of aromatherapy on residents with dementia. Participation is voluntary, and you may withdraw or discontinue at any time up until the survey is added to the anonymous pool of surveys.

Who is conducting the study?
Ms Joy Bowles (primary researcher)
Professor Stephen Myers (supervisor)

What am I required to do?
During the visit by the primary researcher (Joy Bowles), you will be asked to complete the attached survey in blue or black ink, and return it to Joy at the end of her visit. It will take approximately 25-30 minutes to complete. If you don't complete it while Joy is present, please return the completed survey in the stamped addressed envelope supplied, within 2 weeks. The return address is listed at the end of the survey.

Are there any risks?
There are no risks other than the possible inconvenience of the time required to complete the survey. If you are responsible for the aromatherapy program in a number of different aged care facilities in the Northern Rivers area, you may be asked to complete the survey for each facility.

How will the privacy and confidentiality of my information be protected?
Your name and the name of the facility will at no time be connected with the completed survey. Once collected, the survey will be anonymous. No data will be reported that identifies your name or the facility's name or geographical location apart from the fact that it was from the Northern Rivers.

How can I make further inquiries about this study?
If you have any questions or concerns about this study you can contact:
Ms Joy Bowles (primary researcher), Australian Centre for Complementary Medicine, Education & Research, School of Natural & Complementary Medicine, Southern Cross University, Lismore.
Phone: 02 6626 9338, Email: ebowle10@scu.edu.au

Professor Stephen Myers (supervisor), Australian Centre for Complementary Medicine, Education & Research, School of Natural & Complementary Medicine, Southern Cross University, Lismore.
Phone: 02 6620 3403, Email: smyers@scu.edu.au

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Southern Cross University
&
Australian Centre of Complimentary Medicine, Education & Research

Pilot Study: Survey of aromatherapy use in
Northern Rivers residential aged care facilities

AROMATHERAPIST SURVEY

This study is being funded by The Aromatherapy Research Group, a not-for-profit organisation dedicated to researching the evidence base for aromatherapy. If you have any questions about the survey, please contact Ms Joy Bowles (primary researcher) by phone on 02 6626 9338 or email: ebowle10@scu.edu.au or Professor Stephen Myers (supervisor), phone: 02 6620 3403, email: smyers@scu.edu.au.

Please read the Participant Information Sheet before commencing the survey.
Please answer all questions using blue or black ink. Where required, make a vertical mark on each line to depict your response. In the example below, if you think aromatherapy is only slightly helpful in managing agitation you would make a vertical mark as shown.

<table>
<thead>
<tr>
<th>Agitation</th>
<th>Not at all helpful</th>
<th>Extremely helpful</th>
</tr>
</thead>
</table>

1 To what extent do you think the following are major causes of challenging behaviours of residents in this facility? Please make a vertical mark on each line corresponding to your opinion.

<table>
<thead>
<tr>
<th>Major cause of challenging behaviours</th>
<th>Does not cause challenging behaviours</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frustration</td>
<td></td>
</tr>
<tr>
<td>Inability to communicate pain or discomfort</td>
<td></td>
</tr>
<tr>
<td>Pain</td>
<td></td>
</tr>
<tr>
<td>Dementia</td>
<td></td>
</tr>
<tr>
<td>Personality interactions with staff</td>
<td></td>
</tr>
<tr>
<td>Loneliness</td>
<td></td>
</tr>
<tr>
<td>Need for attention</td>
<td></td>
</tr>
<tr>
<td>Survivors of sexual abuse</td>
<td></td>
</tr>
<tr>
<td>Drug abuse</td>
<td></td>
</tr>
<tr>
<td>Brain damage</td>
<td></td>
</tr>
<tr>
<td>Alcohol related brain damage</td>
<td></td>
</tr>
<tr>
<td>Neurological problem other than dementia, alcoholism or drug abuse</td>
<td></td>
</tr>
<tr>
<td>Infections like UTI, URTI</td>
<td></td>
</tr>
<tr>
<td>Incontinence and/or constipation</td>
<td></td>
</tr>
<tr>
<td>Personality interactions with other residents</td>
<td></td>
</tr>
<tr>
<td>Alzheimer's disease brain damage</td>
<td></td>
</tr>
</tbody>
</table>
2 To what extent do you think the following environmental factors affect challenging behaviours of residents in this facility? *Please make a vertical mark on each line corresponding to your opinion* 

<table>
<thead>
<tr>
<th>Factor</th>
<th>Strongly decreases challenging behaviours</th>
<th>Does not increase or decrease challenging behaviours</th>
<th>Strongly increases challenging behaviours</th>
</tr>
</thead>
<tbody>
<tr>
<td>Full moon</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overcast weather</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sunny weather</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sun-downing time (late afternoon)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Loud background noise</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Changing residents’ routine</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pleasant ambient music</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>If facility is short-staffed</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>When new staff start work</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Caring staff attitude</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uncaring staff attitude</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unpleasant ambient odour</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
3 How helpful do you think aromatherapy is in improving the following in this facility? *Please make a vertical mark on each line corresponding to your opinion*

<table>
<thead>
<tr>
<th>Not at all helpful</th>
<th>Extremely helpful</th>
</tr>
</thead>
<tbody>
<tr>
<td>Agitation</td>
<td></td>
</tr>
<tr>
<td>Verbal aggression</td>
<td></td>
</tr>
<tr>
<td>Physical aggression</td>
<td></td>
</tr>
<tr>
<td>Anxiety</td>
<td></td>
</tr>
<tr>
<td>Confusion</td>
<td></td>
</tr>
<tr>
<td>Restlessness in day</td>
<td></td>
</tr>
<tr>
<td>Absconding</td>
<td></td>
</tr>
<tr>
<td>Insomnia</td>
<td></td>
</tr>
<tr>
<td>Reducing nighttime restlessness</td>
<td></td>
</tr>
<tr>
<td>Increasing relaxation</td>
<td></td>
</tr>
<tr>
<td>Decreasing muscle tension</td>
<td></td>
</tr>
<tr>
<td>Constipation</td>
<td></td>
</tr>
<tr>
<td>Improving circulation</td>
<td></td>
</tr>
<tr>
<td>Reducing pain</td>
<td></td>
</tr>
<tr>
<td>Social withdrawal</td>
<td></td>
</tr>
<tr>
<td>Depression</td>
<td></td>
</tr>
<tr>
<td>Infections</td>
<td></td>
</tr>
<tr>
<td>Skin integrity</td>
<td></td>
</tr>
<tr>
<td>Arthritis</td>
<td></td>
</tr>
<tr>
<td>Grief and loss</td>
<td></td>
</tr>
</tbody>
</table>
4 Which aspects of aromatherapy help residents with dementia? *Please make a vertical mark on each line corresponding to your opinion*

<table>
<thead>
<tr>
<th>Not at all helpful</th>
<th>Extremely helpful</th>
</tr>
</thead>
<tbody>
<tr>
<td>Touch (if welcomed by resident)</td>
<td></td>
</tr>
<tr>
<td>Resident likes smell</td>
<td></td>
</tr>
<tr>
<td>Relaxing effects of essential oils</td>
<td></td>
</tr>
<tr>
<td>Calming effects of essential oils</td>
<td></td>
</tr>
<tr>
<td>One-to-one social interaction</td>
<td></td>
</tr>
<tr>
<td>Mental stimulation due to essential oils</td>
<td></td>
</tr>
<tr>
<td>Reminiscence due to smell</td>
<td></td>
</tr>
<tr>
<td>Nurturing contact</td>
<td></td>
</tr>
</tbody>
</table>

5 How many residents in this facility receive aromatherapy, and how often?

<table>
<thead>
<tr>
<th>Dementia Status</th>
<th>Daily</th>
<th>3 or more times a week (not daily)</th>
<th>1-2 times a week</th>
<th>Less than once a week</th>
<th>Never</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of residents with dementia</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Number of residents without dementia</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
6 Of those who receive daily aromatherapy treatments, what is the average number of treatments per day? Please circle as appropriate

<table>
<thead>
<tr>
<th></th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>&gt;4 (specify)</th>
</tr>
</thead>
</table>

7 Which aromatherapy treatments are used in this facility? Please circle Yes or No as appropriate

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Yes / No</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hand massage</td>
<td></td>
</tr>
<tr>
<td>Foot massage</td>
<td></td>
</tr>
<tr>
<td>Neck &amp; shoulders massage</td>
<td></td>
</tr>
<tr>
<td>Full body massage</td>
<td></td>
</tr>
<tr>
<td>Abdominal massage</td>
<td></td>
</tr>
<tr>
<td>Joint massage (e.g. knee, ankle, elbow, wrist)</td>
<td></td>
</tr>
<tr>
<td>Steam inhalation</td>
<td></td>
</tr>
<tr>
<td>Vaporiser (in resident's room)</td>
<td></td>
</tr>
<tr>
<td>Vaporiser (throughout the facility - not specific to individual residents)</td>
<td></td>
</tr>
<tr>
<td>Inhalation from tissue or drops on clothing or pillows</td>
<td></td>
</tr>
<tr>
<td>Spritzer/spray</td>
<td></td>
</tr>
<tr>
<td>Compresses</td>
<td></td>
</tr>
<tr>
<td>Baths</td>
<td></td>
</tr>
<tr>
<td>Foot baths</td>
<td></td>
</tr>
<tr>
<td>After shower body lotion/cream</td>
<td></td>
</tr>
<tr>
<td>Application to wounds/ulcers to aid healing</td>
<td></td>
</tr>
<tr>
<td>In cream to improve skin integrity</td>
<td></td>
</tr>
<tr>
<td>Other (specify)</td>
<td></td>
</tr>
</tbody>
</table>
8 Which conditions are these essential oils mostly used for in this facility, and how widely? *Please write one or two words to describe the condition you most use the oil for, and tick as appropriate to indicate how widely you use the oil for this purpose. If you vaporise the oil throughout the facility, please tick 'For most residents'. Please tick 'Never' if you do not use the oil at all in this facility.*

<table>
<thead>
<tr>
<th>Essential Oil</th>
<th>Condition oil is most used for</th>
<th>For most residents</th>
<th>For a few residents</th>
<th>Never</th>
</tr>
</thead>
<tbody>
<tr>
<td>Basil</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bergamot</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Black Pepper</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Clary Sage</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cypress</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Eucalyptus (globulus)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Frankincense</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Geranium</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>German Chamomile (blue)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ginger</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Juniper</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lavender French</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lavender Tasmanian</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lavender (other)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lime</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Melissa (Lemon Balm)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Myrrh</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Roman Chamomile</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rosemary</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sage</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sweet Marjoram</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tea Tree</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

If there are other oils that you use 'For most residents', please specify:

<table>
<thead>
<tr>
<th>Name of oil</th>
<th>Condition oil is most used for</th>
</tr>
</thead>
<tbody>
<tr>
<td>Other</td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td></td>
</tr>
</tbody>
</table>
9 If you regularly use commercial oil blends please list blend name and company name below and the conditions they are most used for.

<table>
<thead>
<tr>
<th>Blend name</th>
<th>Company name</th>
<th>Condition oil is most used for</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
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<td></td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*If you need more space, please attach a separate piece of paper.*

10 List any adverse effects of aromatherapy that have occurred in your facility. See example given.

<table>
<thead>
<tr>
<th>Adverse Effect</th>
<th>Essential oil(s) used</th>
<th>Application method</th>
<th>Your explanation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Increase in agitation</td>
<td>Lavender</td>
<td>3 drops on pillow</td>
<td>Resident didn't like the smell of Lavender.</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*If you need more space please attach a separate sheet.*

**Background information**

1 Please list your professional aromatherapy qualifications (e.g. Certificate IV Aromatherapy, Diploma of Aromatherapy). *If you have no professional qualifications, please write 'none'.*

<table>
<thead>
<tr>
<th>Qualification type</th>
<th>Number of staff with qualification</th>
<th>Training Institution or Teacher</th>
<th>Length of course</th>
<th>Year qualification obtained</th>
</tr>
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2 Please list any other professional qualifications (e.g. RN, Diploma of Remedial Massage) below.

________________________________________________________________________________
________________________________________________________________________________
________________________________________________________________________________

3 Are you paid for aromatherapy work outside of this facility? *Please circle* Yes / No
4 Which of the following aromatherapy organisations do you belong to? *Tick all that apply*

Australian Aromatic Medicine Association (AAMA)  
International Federation of Aromatherapists (Australia) (IFA)  
International Society of Professional Aromatherapists (ISPA)  
Northern Rivers Aged Care Aromatherapy Network (NRACAN)

If you have any feedback or comments about this survey, or your experience of aromatherapy in dementia care that have not been covered, please add them below:

________________________________________________________________________________
________________________________________________________________________________
________________________________________________________________________________

Thank you for your participation in this study.

Please return the completed survey to:

Joy Bowles  
School of Natural and Complementary Medicine,  
Southern Cross University,  
PO Box 157,  
Lismore NSW 2480
This study is being funded by The Aromatherapy Research Group, a not-for-profit organisation dedicated to researching the evidence-base for aromatherapy. If you have any questions about the survey, please contact Ms Joy Bowles (primary researcher) by phone on 02 6626 9338 or email: ebowle10@scu.edu.au or Professor Stephen Myers (supervisor), phone: 02 6620 3403, email: smyers@scu.edu.au

Please read the Participant Information Sheet before commencing the survey.

1) How many staff members in your facility apart from the aromatherapist have aromatherapy qualifications?_______

2) Please list the qualification details of staff members below:

<table>
<thead>
<tr>
<th>Qualification type</th>
<th>Number of staff with qualification</th>
<th>Training Institution or Teacher</th>
<th>Length of course</th>
<th>Year qualification obtained</th>
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3) Is the facility Government-owned □  Non-goverment owned □
   Other ________________?
4) Do you have a dementia-specific unit/ward? **Yes** □  **No** □

5) How many residents do you currently have? ______________

6) How many residents have a geriatrician's diagnosis of dementia? ____________

7) Who funds aromatherapy in this facility? *Please circle Yes or No as appropriate*
   
   Facility  Yes / No
   Residents/family members  Yes / No
   Other (specify) ________________

8) If funded by the facility, what is the average monthly cost of aromatherapy in this facility? $ ________________

9) Who administers aromatherapy to residents in this facility? *Please circle Yes or No as appropriate*
   
   Visiting Aromatherapist (not OT or DT)  Yes / No
   Nursing staff trained in aromatherapy  Yes / No
   Occupational therapist  Yes / No
   Diversional therapist  Yes / No
   Physiotherapist  Yes / No
   Physio aide  Yes / No
   Activities officer  Yes / No
   Physiotherapist  Yes / No
   Assistants in nursing (AINs)  Yes / No
   Registered nurses  Yes / No
   Family/friends of resident  Yes / No
   Volunteers  Yes / No
10) To what extent does the use of aromatherapy reduce the use of other pharmaceutical preparations for residents that receive aromatherapy? *Please mark on the line as appropriate*

<table>
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<th>Greatly reduces use of other pharmaceuticals</th>
<th>Does not reduce the use of other pharmaceuticals.</th>
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11) If aromatherapy reduces use of other pharmaceuticals in your facility, which ones does it reduce? *List type of pharmaceutical usage that is reduced e.g. aperient, sedative*

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End of Survey - Thank you.