Radiocarbon dating residues from stone tools

Andrea Bettina Yates
Magister Artium/Master of Arts (MA)

A thesis submitted in fulfilment of the requirements of the degree of

Doctor of Philosophy

Southern Cross GeoScience
Southern Cross University Lismore NSW Australia

7th of July 2015
DECLARATION

Thesis Declaration

I, Andrea Bettina Yates certify that the work presented in this thesis is, to the best of my knowledge and belief, original, except as acknowledged in the text, and that the material has not been submitted, either in whole or in part, for a degree at this or any other University.

I acknowledge that I have read and understood the University’s rules, requirements, procedures and policy relating to my higher degree research award and to my thesis. I certify that I have complied with the rules, requirements, procedures and policy of the University (as they may be from time to time).

Print Name:...........................................
Signature:...........................................
Date:..............................................
This dissertation focuses on the development of radiocarbon dating residues from stone artefacts. In the study, the method’s application is examined on artefact samples from archaeological assemblages. While radiocarbon dating of blood residues had been previously reported, dating accuracy required further substantiation. Compromising effects had been described, however the real impact of contamination during sampling steps and analysis protocols remained unclear. Certainly minimizing the quantity of extraneous carbon introduced into samples becomes increasingly significant with small-mass samples.

This study has four consecutive aims: (i) to test the practicability of radiocarbon dating plant residues by investigating the impact of contaminant introduction during sample preparation, (ii) to examine contaminants inherent on stone tools and their removal strategies, (iii) to test the feasibility of the method by dating residues from archaeological stone tools with a reference age and (iv) to develop adequate strategies for contaminant prevention to meet future residue dating requirements.

Methods used for this study comprised of microscopic residue and use wear analyses, including Scanning Electron Microscope Energy Dispersive X-ray (SEM-EDX) observations. Experimental studies evaluated contaminant cleaning from stone tools and residue extraction methods. Samples were radiocarbon dated using Accelerator Mass Spectrometry (AMS). Replicated stone tools were used initially to test techniques. Subsequently, selected artefacts from three archaeological sites were analyzed.

Results of this research show significant steps forward in direct residue radiocarbon dating.

The feasibility of radiocarbon dating ultra-small samples is suggested if contaminants are confined. For instance, the smallest sample of 10.5 µgC – extracted wooden residues from
replicate chert flakes – was dated in agreement with the reference age of ~ 6000 cal BP. Residues dated from an archaeological context showed promising results while emphasizing the crucial impact of contamination for age offsets. Two adhesive residues from one stone tool, which yielded 33.30 μgC and 44.38 μgC, were dated within the expected Late Holocene time frame. Wooden residues from a Mesolithic stone tool, containing 18.97 μgC, were dated successfully, and although deviating ~ 1000 years from the stratigraphical age measurement, the date may still represent the authentic age if taphonomic processes caused tool movement. For the first time it has been shown that the measurement of residues with such small carbon masses yielded dates in or near agreement to the reference age. This is particularly remarkable when considering the archaeological artefacts initially contained multiple contaminants.

Several limitations were overcome by the development of specific sampling and analytical protocols. This will limit common contamination and facilitate accurate artefact residue radiocarbon dating.
DEDICATION

It is with my deepest gratitude and warmest affection

that I dedicate this thesis

to my dearest friend

Dr Virginia A. Finch

Who has been a constant source of encouragement, inspiration, support, knowledge, wisdom and kindness
I would like to acknowledge

The Bundjalung people who are the traditional custodians of this land. I would also like to pay respect to the elders past and present of the Bundjalung nation and extend that respect to other Aboriginal people present.

Associate Professor Anja Scheffers (SCU GeoScience), for supervising in genuine kindness, believing in me when I did not, patience, compassion, availability, showing me the soul of science, rewards of research, the meaning of succinct, moral and financial support, endless editing and proof reading.

Dr. Renaud Joannes-Boyau (SCU GeoScience) for supervising with encouragement, the gift to focus on the essential, overall support, showing how to write more concisely, friendliness, accessibility, continuous proof reading and editing.

Dr Jeffrey Parr (SCU GeoScience) for introduction into microscopic residue and use-wear analyses, lab work, friendliness, support, proof reading and editing.

Dr Andrew Smith (ANSTO) for inspirational collaboration, direction, guidance, clarity, sharing enthusiasm in lithic residue radiocarbon dating research, ready availability for answering endless questions, invigorating discussions, fresh awareness, new insights, friendliness and much support in developing this project.

Fiona Bertuch (ANSTO) for teamwork, guidance in the AMS pre-treatment lab, assistance with sample treatment, patience, friendliness and availability for questions.

Dr Chris Clarkson (UQ) for introduction in Australian lithic stone tool technology, ready availability for questions, literature and inspiring discussions.
Tweed Byron Local Aboriginal Land Council for permitting and endorsing residue radiocarbon dating from stone tools and loaning artefacts for analyses.

Rob Appo, Community Development Officer - Aboriginal Community and Natural Resources, for time spent, availability, meetings and kind support.

Ian Fox for introduction in Australian Cultural matters, availability and immense support.

Journal of Archaeological Science reviewers for constructive comments and critique on manuscript drafts.

The late Dr Thomas Loy for instigating inspirational residue radiocarbon dating research.

Ursula Baumer for discussing GC/MS analyses related matters, providing methodological details, reports and related literature.

Dr Patrick Dietemann for generating and providing Gas Chromatograms and reports.

Southern Cross University (SCU) for use of tremendous facilities and support of staff.

Maxine Dawes of the Environmental Analysis Laboratory for introduction into various microscopes techniques, analysing stone tools in situ, examining uncoated samples with Scanning Electron Microscope - Energy Dispersive X-ray (SEM-EDX).

Southern Cross Geoscience staff for wonderful and kind support during the last four years.

Dr Mark Rosicki for obtaining special equipment and availability for lab related questions.

Dr Andrew Rose for introducing and providing access to Raman Spectroscopy equipment.

Laurel and Yen for always assisting with student matters. Chrisy for the interest in my work. Greg Luker, SCU GIS laboratory for providing maps. Nadia and Michelle for explaining and helping with XRD work and many, many more for their support.
SCU Division of Research for the granting the Australian Postgraduate Award which supported my study tremendously.

AINSE for an award (ALNGRA11032) granted in 2011, allowing initially AMS measurements at ANSTO.

ANSTO for supporting laboratory work and AMS measurement in 2013 through the ICCAS project in the Institute for Environmental Science.

Archaeological division of The Australian Museum Sydney, Allison Dejanovic, Dr Nina Kononenko, Dr Val Attenbrow, Dr Robin Torrence for interest in my work, support, accessibility to collections, kindness. Angela Rosenstein for her support, encouragement, friendship, hospitality, feedback and patience in listening to presentations.

Dr Birgit Gehlen for support, interest, communication, open-mindedness and involvement.

Dr Martin Heinen, for encouragement, support, humour and involvement.

Dr Bernhard Gramsch for sharing archaeological research and material from Friesack.

Dr Alfred Pawlik for friendly collaboration, sharing data and support on conferences.

Dr Jürgen Junkmanns, for instant readiness to offer expertise as experimental archaeologist, producing stone flakes and birch bark tar for this study.

Archaeologist friends from overseas, Marion, Ina, Birgit, Jutta, Bettina, Alfred, for being forgiving for the minute amount of communication during thesis writing.

Dr Virginia Finch, ‘Ginger’, dearest friend and kindred spirit, for support in so many ways, believing in me, giving confidence when the chips were down, seeing the magic in science,
listening to presentations, reading drafts, editing, kindness, wisdom, warmth, safety, compassion, embracing being human, being a beautiful soul and the best role model.

Meeta, for help in maintaining a sense of humour, lunch’s, massages, appreciation, consideration, patience, kindness, support and enduring friendship.

Satyaa, for continuous friendship, magic, insights, understanding, balance, patience, taking care of my health, proof reading, encouragement, believing in me, support, warmth, wisdom, delicious dinners.

Paul, for humour, fun, lightness, laughter and yummy dinners.

Louisa, for patience, constant friendship, sisterhood, loyalty and Yoga-lates.

Liat for poetry, heart, Christmas and Easter dinners.

Graeme, for friendship, generosity, support, presence, humour and making things possible.

Horst (Shuny) for proof reading, advice, kindness and wise words.

Byron Mediation Dance community, Liat, Geash, Marnie, Faith, Nathalie, Efrath, Dwari, Ash, Jay, Dan, Andrew, Heidi, Suryam, Laura, Bacchi, Sufyio and many more for space, no-mind, joyfulness, craziness, friendship and support in so many ways. Thank you all.

My family, Mum Ruth, sister Beate, brother Thorsten, nephew Marek for patience, love and support.

Leroy, K9 friend, for helping to keep me sane, not to take myself too seriously, daily exercise, joy, loyalty and appreciated company.

Trust into the Unknown.
This thesis was prepared in fulfilment of the requirements for the degree of Doctor of Philosophy at Southern Cross University (SCU), GeoScience, Lismore, New South Wales, Australia. Principal and co-supervisors are Associate Professor Anja Scheffers and Dr. Renaud Joannes Boyau (SCU, Australia) with Dr Jeffrey Parr as former supervisor until 2013. During this study extensive laboratory and field investigations were carried out, involving collaboration with researchers from the Australian Nuclear Science And Technology Organization (ANSTO), the University of Cologne, Collaborative Research Centre (CRC) 806, Project D4, the University of the Philippines, Archaeological Studies Program. Results from this research have been presented at four annual conferences of the Australian Archaeological Association (AAA) (giving two oral and three poster presentations) and at the Association of Archaeological Wear and Residue Analysts (AWRANA), in Leiden, The Netherlands in May 2015 (poster presentation).

Thesis organisation

This PhD thesis, by incorporating publications (a format supported by Southern Cross University), includes three peer-reviewed full-length international journal publications (Chapter 3, 4 and Chapter 5). All papers originate from the prime PhD study project. Publications are included in Chapters 3-5 and are formatted according to the guidelines set for the specific PhD thesis requirements (i.e. PhD thesis by incorporating publications) at Southern Cross University.

Appendices contain the published journal articles in their published format, conference contributions (paper abstracts and posters) as well as two publications related to this thesis.
but not part of the actual research project. Written statements of each co-author are also included in the appendices.

The format and references of the three published papers were standardized to be consistent across the thesis. The research aims were defined from the knowledge gaps revealed by the literature review. All papers and chapters are logically linked and research steps outlined in the papers consecutively build on previous results. The following is a brief outline of each chapter.

**Chapter 1: Introduction**

This chapter provides information about the research scope and background information on residue and use wear analysis as well as AMS radiocarbon dating. Key knowledge gaps, overall objectives and specific aims are presented. The research plan and steps are discussed to express how to achieve the aims and objectives of this study.

**Chapter 2: A review on residue analyses aspects relevant to radiocarbon dating**

Literature is reviewed in relation to global lithic residue research and methods used for morphological and biochemical identification, as well as elaborating on specific issues related to AMS radiocarbon dating ultra-small samples including contamination questions and preservation of residues. Several Knowledge gaps are identified, specifically around contamination in various sampling steps, analytical protocols and the handling of very small sized samples for radiocarbon dating.

**Chapter 3: AMS dating of ancient plant residues from experimental stone tools: a pilot study (published)**
In this chapter the issue of laboratory introduced contamination is examined, while preparing residues from stone tools for AMS dating. This experimental study demonstrates the feasibility of radiocarbon dating three wooden residue samples (containing as little as 10.5 μg carbon mass) from chert flakes, consistent with the reference age of ~6000 cal BP. The results suggest that AMS laboratory contamination addition is small enough to allow dating of ultra-small Holocene age residue samples:


Chapter 4: Radiocarbon-dating adhesive and wooden residues from stone tools by Accelerator Mass Spectrometry (AMS): challenges and insights encountered in a case study (published)

Based on the new knowledge gained in the experiments presented in Chapter 3, the logical next step was proceeding to test the applicability of the method on residues from archaeologically retrieved stone tools. Various cleaning methods are examined first experimentally, then on archaeological artefacts. Residues dated from an archaeological context showed promising results but also demonstrated the significance of contaminant influence on microgram-sized samples. This study also revealed previously unpredictable factors potentially responsible to cause age offsets:

Chapter 5: Residue radiocarbon AMS dating review and preliminary sampling protocol suggestions

Challenges and problems arising from the preceding residue dating studies (in Chapters 3 and 4) led to the development of a specific sampling protocol in this study section. Furthermore, a review of residue dating research and related issues points out pitfalls and recurrent problems leading to contaminated samples and age offsets. The discussed and demonstrated steps and results are hoped to promote residue research as a new archaeo-chronological tool:


Chapter 6: Synthesis and conclusion

This section summarises results and outcomes of preceding chapters, draws the overall conclusions and points out implications of this study project. Recommendations for future work are presented.

References

References of both, published journal articles and unpublished chapters are included in this section.

Appendices

The appendices contain signed author contribution statements, a list of conference abstracts associated with the work of this PhD thesis, published papers related to Chapter 3, 4 and 5 and two publications related but not part of this work.
AUTHOR CONTRIBUTION STATEMENT

Conception and design: AY (85%), AMS (7.5%), AS (2.5%), JP (2.5%), RJB (2.5%)

Samples analysis: AY (100 %)

Interpretation of results: AY (90 %), AMS (10 %)

Data collection: AY (100 %)

Thesis writing: AY (100 %)

Publication writing: AY (85 %), AMS (6 %), FB 2%; AS (1%) RJB(1%), JP (1%), BGe (1.%), BGr (1%), MH (1%), AP (1%)

Data analysis and interpretation: AY (90 %) AMS (8 %), (FB 2%)

Obtaining funding: AY (80 %), AS (20%)

Overall responsibility: AY (100 %)

Author declaration

I, Andrea Bettina Yates, the author of this thesis, certify that the contributors and conflicts of interest statements included in this thesis are correct and have been approved by all contributors.

Approved by all contributors. Signature: Date: 22/06/2015

I agree with the contents of the thesis; to being listed as a contributor; and to the conflicts of interest statement as summarised. I have had access to all the data in the study and accept responsibility for its validity.

A/Prof Anja Scheffers (AS) Signature: Date: 22/06/15

Dr Renaud Joannes-Boyau (RJB) Signature: Date: 22/06/15
Signed statements from each of the following contributors are included in Appendix 1:

Dr Andrew M. Smith (AMS)

Ms Fiona Bertuch (FB)

Dr Jeffrey Parr (JP)

Dr Birgit Gehlen (BGe)

Dr Bernhard Gramsch (BGr)

Dr Martin Heinen (MH)

Dr Alfred Pawlik (AP)
TABLE OF CONTENTS

THESIS DECLARATION ........................................................................................................... II

ABSTRACT ............................................................................................................................ III

DEDICATION .......................................................................................................................... V

ACKNOWLEDGEMENTS ....................................................................................................... VI

PREFACE ............................................................................................................................... X

AUTHOR CONTRIBUTION STATEMENT ......................................................................... XIV

TABLE OF CONTENTS ......................................................................................................... XVI

LIST OF TABLES ................................................................................................................... XXII

LIST OF FIGURES ................................................................................................................ XXIII

LIST OF ABBREVIATIONS .................................................................................................. XXVI

CHAPTER 1. INTRODUCTION

INTRODUCTION ...................................................................................................................... 1

1.1. Background .................................................................................................................... 1

1.1.1. Residue and use wear analysis on stone artefacts .................................................... 1

1.1.2. Preservation .............................................................................................................. 3

1.1.3. Previous radiocarbon dating of stone tool residues ............................................... 4

1.1.4. Radiocarbon dating by Accelerator Mass Spectrometry (AMS) ............................... 5

1.2. Definition of the problem ............................................................................................. 8

1.3. Knowledge gaps – the role of contaminants and suitable residue types ....................... 9

1.4. Research limitations ..................................................................................................... 10

1.4.1. Financial and instrument access limitations .......................................................... 10

1.4.2. Time limitations of a PhD research project .............................................................. 11

1.5. Aims and objectives ..................................................................................................... 11

1.6. Research design ......................................................................................................... 13

1.6.1. Phase 1: Pilot experiment ....................................................................................... 14

1.6.2. Phase 2: Case study .............................................................................................. 15

1.6.3. Phase 3: Sampling strategies .................................................................................. 16
1.7. Study materials and study sites .....................................................16
  1.7.1. Phase 1 .................................................................................16
  1.7.2. Phase 2 .................................................................................18
  1.7.3. Phase 3 .................................................................................21
1.8. Archaeological study sites .................................................................21
  1.8.1. Friesack 4 .............................................................................21
  1.8.2. Wesseling .............................................................................23
  1.8.3. Yelgun .................................................................................23

CHAPTER 2. A REVIEW OF RESIDUE ANALYSES ASPECTS RELEVANT TO
AMS RADIOCARBON DATING

2.1. Introduction ..................................................................................27
2.2. Definitions, methodology and case studies ......................................27
  2.2.1. Definitions ..............................................................................27
    2.2.1.1. Residue analysis .................................................................27
    2.2.1.2. Use wear analysis ...............................................................29
    2.2.1.3. Use-related residues ...........................................................29
    2.2.1.4. Non-use-related residues ..................................................33
  2.2.2. Methodology ..........................................................................33
    2.2.2.1. Optical interpretation .........................................................33
    2.2.2.2. Chemical identification .......................................................34
    2.2.2.3. Experimental studies to understand residue behaviour on
             lithic artefacts .................................................................35
  2.2.3. Case studies ............................................................................39
    2.2.3.1. Multiple tool use ...............................................................39
    2.2.3.2. Subsistence .......................................................................39
    2.2.3.3. Open sites surface stone tools versus excavated artefacts ...40
2.3. Contaminant sources and transfer to archaeological artefacts ..........40
  2.3.1. Post depositional .....................................................................40
  2.3.2. Starch ....................................................................................42
  2.3.3. Fungus ..................................................................................42
  2.3.4. Excavation, retrieval and storage of artefacts ..............................44
  2.3.5. Post excavation ......................................................................46
  2.3.6. Case study .............................................................................47
2.4. Residue Preservation

<table>
<thead>
<tr>
<th>Subsection</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.4.1. Conditions</td>
<td>48</td>
</tr>
<tr>
<td>2.4.2. Proportions of stone tools with residue preservation in lithic assemblages</td>
<td>50</td>
</tr>
</tbody>
</table>

2.5. Previous radiocarbon dating of residues from stone tools

2.6. Previous contaminant mitigation strategies

2.7. Identified aspects relevant for research directions for this study

2.8. Identified knowledge gaps relevant for research directions for this study

CHAPTER 3. AMS DATING OF ANCIENT PLANT RESIDUES FROM EXPERIMENTAL STONE TOOLS: A PILOT STUDY

ABSTRACT

3.1. Introduction

<table>
<thead>
<tr>
<th>Subsection</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>3.1.1. Identification and preservation of residue on stone artefacts</td>
<td>60</td>
</tr>
</tbody>
</table>

3.2. Materials and Methods

<table>
<thead>
<tr>
<th>Subsection</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>3.2.1. Materials and Procedure</td>
<td>63</td>
</tr>
<tr>
<td>3.2.2. Preparation for AMS radiocarbon dating</td>
<td>66</td>
</tr>
</tbody>
</table>

3.3. Results

<table>
<thead>
<tr>
<th>Subsection</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>3.3.1. Wood samples</td>
<td>69</td>
</tr>
<tr>
<td>3.3.2. Peat samples</td>
<td>69</td>
</tr>
</tbody>
</table>

3.4. Discussion

3.5. Conclusion

3.6. Acknowledgements

CHAPTER 4. RADIOCARBON-DATING ADHESIVE AND WOODEN RESIDUES ON STONE TOOLS BY ACCELERATOR MASS SPECTROMETRY: A CASE STUDY

ABSTRACT

4.1. Introduction

<table>
<thead>
<tr>
<th>Subsection</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>4.1.1. Archaeological study sites</td>
<td>84</td>
</tr>
<tr>
<td>4.1.1.1. Friesack</td>
<td>84</td>
</tr>
<tr>
<td>4.1.1.2. Wesseling</td>
<td>84</td>
</tr>
</tbody>
</table>
4.2. Materials and Methods ........................................................................................................... 85

4.2.1. Remarks on conditions of GC/MS analyses previously carried out on Friesack samples ................................................................. 85
4.2.2. Optical residue and use-wear analyses ................................................................................. 87
4.2.3. Experimental design to establish decontamination protocols ............................................. 88
4.2.4. Methodological approach on artefacts - preparation for AMS dating ............................... 89
  4.2.4.1. Cleaning pre-treatment with Decon 90, 2% diluted ......................................................... 89
  4.2.4.2. Residue Removal ............................................................................................................ 90
  4.2.4.3. AMS dating ................................................................................................................... 90
4.2.5. Establishing elemental characteristics of residues .......................................................... 90

4.3. Results and Discussion ........................................................................................................ 91

4.3.1. Optical residue and use-wear analyses .............................................................................. 91
4.3.2. Experimental design to establish decontamination protocols ........................................ 95
4.3.3. Methodological approach on artefacts - preparation for AMS dating ............................. 95
  4.3.3.1. Cleaning pre-treatment with Decon 90, 2% diluted ......................................................... 95
  4.3.3.2. Residue Removal ............................................................................................................ 97
  4.3.3.3. AMS dating ................................................................................................................... 100
    4.3.3.3.1. Adhesive residues ................................................................................................. 100
    4.3.3.3.2. Wooden residues ...................................................................................................... 102
4.3.4. Establishing elemental characteristics of residues .......................................................... 104
  4.3.4.1. OZQ689 ......................................................................................................................... 104
  4.3.4.2. OZQ695 ......................................................................................................................... 106
  4.3.4.3. OZQ696 ......................................................................................................................... 107

4.4. Discussion ............................................................................................................................ 110

4.5. Conclusion ............................................................................................................................ 113

4.6. Acknowledgements ............................................................................................................. 114

CHAPTER 5. RESIDUE RADIOCARBON AMS DATING REVIEW AND
PRELIMINARY SAMPLING PROTOCOL SUGGESTIONS

ABSTRACT ..................................................................................................................................... 116

5.1. Introduction .......................................................................................................................... 117

5.2. Aspects related to AMS radiocarbon dating ....................................................................... 119
  5.2.1. Residue identification ........................................................................................................ 119
5.2.1.1. Microscopic residue interpretation ...................................................... 119
5.2.1.2. Biochemical identification and interpretation ........................................ 119
5.2.2 Residue Preservation .................................................................................... 121
  5.2.2.1. Residue preservation in general ............................................................ 121
  5.2.2.2. Specific residue preservation ................................................................. 122
  5.2.2.3. Conditions that protect preserved residues ............................................ 122
5.2.3 Contamination .............................................................................................. 124
  5.2.3.1. Radiocarbon dating small (<100 μg) samples ...................................... 124
  5.2.3.2. Contaminant management in the AMS pre-treatment laboratory .......... 125
  5.2.3.3. Case study ............................................................................................ 125
  5.2.3.4. Contamination added in the field/storage environments ...................... 127
  5.2.3.5. Contamination added in artefact handling after retrieval ..................... 127
  5.2.3.6. Contamination added during sample processing prior to arrival in the
                     AMS laboratory .......................................................................................... 129
  5.2.3.7. In-built ages affecting radiocarbon determinations ............................... 131
5.2.4 Residue type, sample size and yielded carbon mass ...................................... 133
  5.2.4.1. Sample size and yielded carbon mass .................................................... 133
  5.2.4.2. Residue type ......................................................................................... 135
  5.2.4.3. Case study 2 ......................................................................................... 138
5.3. Discussion ...................................................................................................... 140
  5.3.1. Implications for retrieving, handling and storing archaeological objects .... 140
    5.3.1.1. Fieldwork and storage ............................................................... 142
    5.3.1.2. Cleaning and preparing ............................................................... 142
    5.3.1.3. Microscopic examination .............................................................. 143
    5.3.1.4. Chemical identification ................................................................. 143
    5.3.1.5. Extraction/removal ....................................................................... 145
5.4. Conclusion ..................................................................................................... 146
5.5. Acknowledgments ......................................................................................... 148

CHAPTER 6. SYNTHESIS AND CONCLUSION
6.1. Synthesis ...................................................................................................... 149
6.2. General conclusions ..................................................................................... 152
6.3. Limitations ................................................................................................... 153
6.4. Future work .................................................................................................. 154
  6.4.1. Further recommendations ...................................................................... 155
REFERENCES

APPENDIX 1 CO-AUTHOR CONTRIBUTION STATEMENT

APPENDIX 2 CONFERENCE PRESENTATIONS

APPENDIX 3 CHAPTER 3 - PUBLISHED MANUSCRIPT

APPENDIX 4 CHAPTER 4 - PUBLISHED MANUSCRIPT

APPENDIX 5 CHAPTER 5 - PUBLISHED MANUSCRIPT

APPENDIX 6 SIDE PUBLICATION:

APPENDIX 7 SIDE PUBLICATION:
Table 2.1. Residue and use-wear criteria for hypothesised task associations and functions (adapted from Robertson, 2011:87) ................................................................. 31

Table 2.2. Plant and inorganic materials: Residue and use-wear criteria for hypothesised task associations and functions (adapted from Robertson, 2011:88) ................................. 32

Table 3.1. Radiocarbon ages from original wood and peat samples and associated residue samples extracted by sonication and scraping, amount of carbon mass used for AMS dating, time range of calibrated radiocarbon ages (using CALIB 6.0. after Stuiver et al., 2012). ......68

Table 4.1. Summary of archaeological samples and preparation protocol .......................................................... 86

Table 4.2. Summary of residue sample dating by AMS. Radiocarbon ages are indicated in BP and previously obtained dates are from Görsdorf and Gramsch 2004 and Gramsch, 2000 and 2012. (Abbreviations: D = Decon90, 2% diluted, DCM = dichloromethane treated, AAA = Acid-Alkali-Acid, pMC = Percent Modern carbon) ........................................... 99

Table 4.3 Extracted residue from cavity of OZQ689: Weight Percentages determined from EDX data for elements >1% average atomic weight ................................................. 106

Table 4.4. Residue and rock matrix of OZQ695: Weight Percentages determined from EDX data for elements >1% average atomic weight .................................................. 106

Table 4.5 Residue and rock samples of OZQ696, in comparison with contemporary Xanthorrhoea samples: Weight Percentages determined from EDX data for elements >1% average atomic weight ............................................................ 109

Table 5.1.: Fullagar’s sampling suggestions for residue preservation during and following fieldwork (Fullagar 2006b: 189,191,195) and required sampling adjustments for AMS residue dating .............................................................................. 128

Table 5.2. Causes for residue radiocarbon dates that are too young and potential removal strategies ..................................................................................................................... 131

Table 5.3. Causes for residue radiocarbon dates that are too old and potential removal strategies .............................................................. ........................................... 132

Table 5.4. Overview of AMS radiocarbon dated archaeological residues (*from lithics, rock art, and food residues from ceramics) and correlation of residue dates with residue quantity microgram of Carbon (µgC) (if available) and associated age control ............................................. 134

Table 5.5. Sampling protocol suggestions to avoid contaminant introduction prior to AMS radiocarbon dating ........................................................................................................... 144
LIST OF FIGURES

Figure 1.1. Succession of research steps consecutively building on previous results.............. 14

Figure 1.2. Initial trial: scraping and cutting movements with replicated stone flakes on contemporary materials. ................................................................................................................................. 17

Figure 1.3. Production of chert flakes using a copper headed percussion stick and cutting movements with replicated stone flakes into wood of a known age. .......................................................... 18

Figure 1.4. Map 1: showing the sites Friesack and Wesseling in Germany, Map 2: indicating the location of Yelgun, northeast NSW in Australia (map designed by Greg Luker, SCU GeoScience). ........................................................................................................... 19

Figure 1.5. Experimental set up to evaluate cleaning techniques as preparation for sample pre-treatment for AMS dating. 1: proximal end of ventral surface of fabricated stone flakes before treatment, 2: fabricated flakes with attached contemporary birch tar and pencil graphite lines on proximal end of ventral surface, 3: removal technique, 4: proximal end of ventral surface of fabricated stone flakes after treatment. G = Graphite, BT = Birch tar; 1, 2 and 4 = microscopic images of 7 x to 10 x magnification ranges................................................. 20

Figure 1.6. Mesolithic flint stone tools from Friesack 4: 1-3 core axes, 4 scaper, 5 flake fragment with macroscopically visible dark residue................................................................. 22

Figure 1.7. Wesseling , chert flake with macroscopically visible dark residue..................... 23

Figure 1.8. Lithic assemblage from Yelgun, NSW, Australia. From above left to bottom right: Grinding dish fragment, bungwall pounders, cobbles with use wear or pitting marks, ground edge hatchets, flat chalcedony piece with cortex, flakes, cores and tools sorted by raw material. ...................................................................................................................... 24

Figure 1.9. Lithic assemblage of Yelgun, NSW, Australia: relationship of raw material to manufactured stone tool type.......................................................... 25

Figure 1.10. Yelgun, chalcedony pebble with dark residue patches.................................. 25

Figure 3.1. Production of stone flakes using antler as a percussion stick and leather leg/lap protection, scraping wood with flake, scraping fern residue with scalpel monitored under the microscope, extracting wood residue in timed sonic bath.................................................. 63

Figure 3.2. Production of stone flakes using metal headed percussion stick, plastic-coated production surface and wearing starch-free gloves, cutting into wood, sample pre-treatment with the acidic-alkali-acid method. .................................................................................64

Figure 3.3. Sequence of experiment part two, showing number of extracted samples proceeding to AMS.................................................................................................................. 66
Figure 3.4. Radiocarbon age of NA5 original wood and corresponding dates of extracted residues.

Figure 3.5. Radiocarbon age of NA2 original peat and corresponding date of extracted residue.

Figure 3.6. Radiocarbon age of NA3 original peat and corresponding date of extracted residue.

Figure 3.7. Radiocarbon age of NA4 original peat and corresponding date of extracted residue.

Figure 4.1. Macroscopic and microscopic images of stone tools containing putative adhesive residues: 1 late Paleolithic flake (OZQ695), 2 Mesolithic flake fragment (OZQ694), 3 undated cobble fragment (OZQ696), 4-9 OZQ695: Microscopic images of 4 adhesive concentration 32x mag., 5 partially droplet appearance of adhesive, 6 white mass overlays adhesive, possibly bone collagen, 115x mag., 7 pencil graphite overlaying adhesive residue, 8 modern fabric fiber, 9 possible fungus contamination. 10-12 OZQ694: Microscopic images of 10 dark droplet appearance of adhesive with brown (plant) tissue, 11 edge area with fibrous (plant) material and dark droplet like adhesive spots 100x mag., 12 right margin edges damaged and fibrous (plant) material 20x mag., 13-15 OZQ696: Microscopic images of 13 presumably adhesive chunk mixed with sand 25x mag, 14 adhesive patch cross section at 20x mag., 15 adhesive chunks and mud cracked appearance of adhesive parts at 7x mag.

Figure 4.2. Macroscopic and microscopic images of Mesolithic stone tools with identified wooden residues associated with use-wear traces. 1st row: OZQ689 (Friesack, D5/7), Core axe, (Scale 1 =1000 μm, 2 - 500 μm, 3 = 200 μm). 2nd row: OZQ690 (Friesack, B2/7), Core axe (Scale, 1 and 2 = 500 μm, 3 = 2000 μm). 3rd row: OZQ691 (Friesack, F25/10b), Core axe, (Scale 1 and 3 = 1000 μm, 2 = 200 μm). 4th row: OZQ692 (Friesack, CO15/8a), Scraper, (Scale 1 and 3 = 500 μm, 2 = 1000 μm).

Figure 4.3. Detailed picture of OZQ689 wooden residues intermixed with dark deposit from an unknown substance (possibly adhesive residue) (200x magnifications).

Figure 4.4. Residue radiocarbon ages (rounded) plotted against anticipated ages. From left to right: Both samples of OZQ696 date within expected Late Holocene age range, OZQ695 significantly too young, OZQ694 and OZQ689-U1 significantly too old, 6 OZQ689-U2 dates close to anticipated age, OZQ691 and OZQ692 too young.

Figure 4.5. SEM images from OZQ689 residue: 1 Unknown substance possibly adhesive material with silica chunk attached right side, (Scale = 50 μm); 2 and 3 Unknown organic substance (Scale = 20 μm); 4 from left to right: silica chunk, partially unknown substance and wood piece (Scale = 200 μm); 5, 6 shell with unknown residue (Scale =50 μm).
Figure 4.6. SEM images of OZQ695 residue: 1 adhesive streaks (white); 2 adhesive streaks (white) and grey traces from scraping off adhesive; 3 residue on tip area of tool (Scale 1 + 2 = 1.0 μm, Scale 3 = 10 μm). ................................................................. 107

Figure 4.7. SEM images from residue recorded on artefact OZQ696 (images 1-5) and a contemporary Xanthorrhoea sample (6): 1-3 transition from chipped part to more sealed part of sample A imaged in increasing magnification (Scale, 1 =100 μm, 2 = 10 μm, 3 = 20 μm), 4-5 structure of sample B (Scale, 4 = 20 μm, 5 =100 μm), in contrast: 6 dense and compact consistency of contemporary Xanthorrhoea sample, cracks here occurred through beam (Scale = 300 μm) .............................................................................................................. 108

Figure 4.8. Gas-Chromatogram from birch bark pitch of Friesack, find 1977:7/P4. Betulin, lupeol and lupenone, biomolecular markers of birch bark. Additionally plasticizers in form of Phtalatestern, Dibutylphthalat (DBP) (Courtesy of Baumer and Dietemann, 2008, Doerner Institute, Munich). .................................................................................................................. 111

Figure 4.9. Suggested workflow of method sequence for efficient residue AMS dating...... 113

Figure 5.1. Gas-Chromatogram from birch bark pitch of Friesack, find 1977:7/P4. Betulin, lupeol and lupenone, biomolecular markers of birch bark. Additionally, plasticizers in the form of Phtalatestern, Dibutylphthalat (DBP) (Courtesy of Baumer and Dietemann 2008, Doerner Institute, Munich). .................................................................................................................. 139

Figure 5.2. Stages of potential non-use related contaminants transfer on archaeological artefacts .............................................................................................................................................. 147
LIST OF ABBREVIATIONS

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>AAA</td>
<td>Australian Archaeological Association</td>
</tr>
<tr>
<td>AAA</td>
<td>Acid-Alkali-Acid</td>
</tr>
<tr>
<td>AINSE</td>
<td>Australian Institute of Nuclear Science and Engineering</td>
</tr>
<tr>
<td>AMS</td>
<td>Accelerator Mass Spectrometry</td>
</tr>
<tr>
<td>ANSTO</td>
<td>Australian Nuclear Science and Technology Organisation</td>
</tr>
<tr>
<td>AWRANA</td>
<td>Association of Archaeological Wear and Residue Analysts</td>
</tr>
<tr>
<td>BP</td>
<td>Before Present</td>
</tr>
<tr>
<td>CRC</td>
<td>Collaborative Research Centre</td>
</tr>
<tr>
<td>DCM</td>
<td>Dichloromethane</td>
</tr>
<tr>
<td>GC/MS</td>
<td>Gas Chromatography Mass Spectrometry</td>
</tr>
<tr>
<td>ICCAS</td>
<td>Isotopes in Climate Change and Atmospheric Systems</td>
</tr>
<tr>
<td>μgC</td>
<td>Micrograms of Carbon</td>
</tr>
<tr>
<td>pMC</td>
<td>Percent Modern Carbon</td>
</tr>
<tr>
<td>SEM-EDX</td>
<td>Scanning Electron Microscope Energy Dispersive X-ray</td>
</tr>
<tr>
<td>SCU</td>
<td>Southern Cross University</td>
</tr>
<tr>
<td>UQ</td>
<td>University of Queensland</td>
</tr>
</tbody>
</table>
CHAPTER 1

Introduction
INTRODUCTION

This PhD thesis offers an innovative contribution to the field of radiocarbon dating plant residues recovered from stone artefacts by means of Accelerator Mass Spectrometry (AMS). The relatively new technology enables the direct dating of small ancient use-related traces on artefacts. A prerequisite for this is a sound residue identification. Residue and use wear analysis on stone tools and AMS radiocarbon dating represent the research directions involved in this study. Subsequently, brief background information to both research fields is provided, while further data can be found in the literature review (Chapter 2) and in Chapters 3, 4 and 5.

1.1. Background

1.1.1. Residue and use wear analysis on stone artefacts

Stone tools from archaeological contexts can contain a suite of residues that might result from ancient task-related use of the artefact. Residues of both organic and inorganic nature can be observed microscopically and characterized using established diagnostic criteria (e.g. Fullagar, 2006a; Hardy and Garufi, 1998; Haslam et al., 2009; Lombard, 2008), reference materials (e.g. Field, 2006; Kononenko, 2011) and increasingly, biochemical methods (e.g. Monnier et al., 2013). Accurate residue characterization is crucial to ascertain if the residues on stone artefacts are use-related or are contaminant remains. Use-related residues can be subdivided into animal residues (including lipids, collagen, bone, hair, tissue, fibres, blood, feather barbules and scales), plant residues (including plant tissue, wood fiber, starch, phytoliths, raphides, adhesive materials such as resin, tar, gum and wax) and inorganic residues (including ochre, vivianite, aragonite and mica) (e.g.
FULLAGAR, 2006A; LOMBARD AND WADLEY, 2007; ROBERTSON, 2005, 2011). Non-use-related or contaminant residues can be transferred incidentally to a stone tool surface. This can occur during the time of use, for instance, if a tool comes into contact with organic material in a way that is not the result of using the tool for a task or from the time of artefact deposition onwards. These contaminants include fungi, lipids, skin scales and fabric fibre (e.g. Langejans, 2012b). Combined use wear and residue analyses have shown to be helpful in evaluating and discerning use-related from contaminant residues. This allows for a more confident interpretation of traces resulting from working or using the tool in a particular manner. Typical marks on stone tools include striations, rounding, polish and edge damage (e.g. Fullagar, 2006a; Robertson, 2009, 2011). When residues are found in context with these wear traces, the probability of their use-relatedness increases. Microscopically based use wear and residue analyses on Australian artefacts was first conducted by Fullagar (1986) and included the development of interpretation criteria. The method’s significance lies in its ability to solve functional questions in lithic research, as well as providing information about subsistence behaviour.

In the last two decades interest in residue analysis has intensified, especially in the Americas, Southern Africa, Oceania and Europe. This is related to increasing recognition of the method’s validity and ability to address particular research questions. For instance, such an approach might allow for the determination that stone artefacts were used for various tasks, regardless of their shape (e.g. Robertson, 2009, 2011). Particular areas of research interest include residue analyses of starches, residue preservation and residues associated with specific stone tool types including backed artefacts (Attenbrow et al., 2009; Robertson, 2009) and grinding stones (Fullagar et al., 2015). Experimental based analyses raised the awareness of contamination on stone tools (e.g. Haslam, 2004;
Langejans, 2011; Loy and Barton, 2006). A suite of further experimental studies helped in understanding complex residue patterns including wood (e.g. Hardy and Garufi, 1998) and blood residues on stone tools (e.g. Cattaneo et al., 1993; Gurfinkel and Franklin, 1988; Hyland et al., 1990; Shanks et al., 2004). Furthermore, issues around residue preservation were explored (Barnard et al., 2007; Barton, 2009; Jones, 2009; Langejans, 2010; Wadley and Lombard, 2007).

1.1.2. Preservation

Use-related residues were found preserved on stone artefacts in South Africa for up to two million years old (Loy, 1998; Jones, 2009). While the residue preservation on artefacts from the the Oldowan industry of Sterkfontein is exceptionally old, there are other sites with preserved residues of considerable age. For example: Tanzania has associated ages from 1.5–1.6 million years (Dominguez-Rodrigo et al., 2001) and the South African Sibudu cave, from 60,000 to 50,000 years BP, where lithic points containing organic residues from hafting (Lombard and Wadley, 2009) have been found. In Europe, residues on stone tool surfaces have been preserved from 48,000 years ago placing them in the Middle Paleolithic Mousterian period (Hardy et al., 1997). Further residue preservation was observed in sites of the early Upper Paleolithic Aurignacienn period (Hardy, 2009; Hardy et al., 2008) and in Mesolithic sites (Hardy and Svoboda, 2009; Pawlik, 2004). In Australia, residues were found preserved on lithics with age ranges between 32,000—37,000 years BP (Dodson et al., 1993; Fullagar and David, 1997) on backed artefacts with age determinations from 1,500—8,500 years BP (e.g. Attenbrow et al., 2009; Robertson et al., 2009), while a site in Tasmania yielded residues dating to 6000 years BP (Fullagar and Jones, 2004). Furthermore, 28,000-year-old starch residues were found on stone artefacts
from the Pacific region (Loy et al., 1992) and 90,000-year-old blood residues were identified on lithics from Israel (Loy and Hardy, 1992).

For most sites, both relative (typology and stratigraphic) and absolute (including radiocarbon dating on associated organic material) dating was conducted. The ability to directly date use-related residues from stone tools by AMS would expand numerical dating possibilities to artefacts lacking find context associated organic material. Though, of course the dateable time range for such an approach would be constrained to the last 50 ka.

1.1.3. Previous radiocarbon dating of stone tool residues

Previous attempts to apply radiocarbon dating to residues on stone tools was limited to blood residues (Loy, 1987, 1993; Nelson et al., 1986). This initial research was carried out on two Canadian stone tools: one contained 3 mg and the other 50 μg of carbon derived from blood residues and yielded radiocarbon dates of 1060 ± 160 and 2180 ± 160 years BP respectively. The age measurements obtained were in agreement with the reference ages (Nelson et al., 1986) – represented by dates from a hearth for the first and typology for the second stone tool. Further evaluation suggested that required blood residue quantities should range from 1 mg to 50 μg of carbon (Loy, 1987:62, 1993:46; Vogel et al., 1989:608). Loy (1987, 1993) also pointed out the important issue of contamination affecting AMS dates when using small sized residue samples. He emphasized that many methods able to separate contaminants from the sample unfortunately contain carbon-based chemicals (Loy, 1993:48), which could distort age measurements. Furthermore, he explained the difference between the initial sample size and the purified carbon mass sample.
To date, no systematic attempts have been carried out to test radiocarbon dating of use-related residues from stone tools, other than on blood with microgram sized carbon samples, or to clearly assess the problem of introduced carbon affecting residue samples processed for dating. This is of particular importance as recent residue analyzed lithics from archaeological contexts revealed that more than 50% of the analyzed stone tools contained residues (Hardy and Svoboda, 2009:165; Hardy et al., 2008:652; Robertson, 2009:300, 302). These high percentages of residue containing stone tools indicate their significant potential to be radiocarbon dated by AMS. As will be elaborated on further, this technology’s recent advances permit the dating of ‘ultra-small' samples containing carbon in the microgram scale. Until now, there has been insufficient understanding about the impact of contamination and carbon transfer which adversely affect the dating results. Furthermore, contaminant mitigation measures and a lack of adequate sampling protocols are also limiting factors.

1.1.4. Radiocarbon dating by Accelerator Mass Spectrometry (AMS)

Radiocarbon dating is a nuclear technique based on the measurement of the radioactive decay of the isotope $^{14}\text{C}$. The decay rate is measured by the half-life of $^{14}\text{C}$. Two values are commonly reported: the conventional ‘Libby’ value of 5568 ± 30 years BP (Anderson and Libby, 1951) and the ‘Cambridge’ value with a half life of 5730 ± 40 years (Godwin, 1962; Bronk Ramsey, 2008:254). The latter value is the revised version of the former and is thought to be the more accurate estimate (Godwin, 1962).

The radiocarbon method was developed by Willard Libby who first understood the development of radiocarbon in the atmosphere and that organic material could be dated by measuring the concentration of the isotopic carbon in ancient remains (Libby, 1946). Soon
after the first samples of known ages were successfully radiocarbon-dated (Arnold and Libby, 1949). Since then the technique has reformed archaeological chronologies by enabling independent dating possibilities for sites that lack chronological markers. As originally conceived, radiocarbon analysis relied on radiometric (beta) counting methods, which monitor the decay of $^{14}$C atoms by detection of the associated beta particles (maximum energy 154 keV). Due to the comparatively long half-life of $^{14}$C, achieving high accuracy with this technique required either very active or very large samples (or both) in order to achieve sufficient counting statistics and to maximize the noise to counts ratio. Accelerator Mass Spectroscopy (AMS), on the other hand, is an ultrasensitive technique based on atom counting, where individual atoms are identified only on the basis of their mass and atomic number. For this reason, accurate measurements can be made rapidly (20–30 min) on very small (as little as a few micrograms of carbon) and relatively inactive samples (backgrounds down to ~50 ka BP). This represents a considerable improvement over the original beta counting method and the AMS technique has been widely applied in numerous fields including archaeology, environmental science and climatology.

Muller (1977) demonstrated that by accelerating particles, a mass spectrometer could detect miniscule traces of $^{14}$C. Key to the success of $^{14}$C AMS are factors including that; (i) the isobar, $^{14}$N, is metastable and is thus eliminated when tandem accelerators are used; (ii) charge stripping in the central positive terminal destroys molecules ($^{12}$CH$_2^-$ and $^{13}$CH$^-)$, thereby eliminating ‘molecular ambiguity’; and finally; (iii) the high energies obtained permit the use of nuclear-physics ionization detectors, permitting single-atom detection on the basis of Bragg energy loss. Most importantly, AMS has the capacity to directly measure isotope ratios (e.g. $^{14}$C:$^{12}$C or $^{14}$C:$^{13}$C ratios). After ten half-lives or ~50 ka, the
amount of $^{14}$C left in the sample becomes so small that detection limits and background contamination generally prevent reliable dating. Measurement uncertainties always increase with sample age (e.g. Waterbolk, 1971) because the number of $^{14}$C atoms that remain for measurement is limited.

The inception of AMS radiocarbon dating swiftly led to a significant reduction in sample size from 1 g to ca 1 mg (e.g. Harris et al., 1987). This allowed multiple sample measurements from a site and thus enabled higher resolution dating. It also allowed precious and rare organic objects to be dated (the minute material amounts needed prevents complete destruction of such artefacts) and a wider range of materials: from blood residues from stone tools (e.g. Nelson et al., 1986; Loy, 1987, 1993) to organic substances in rock art pigments (Loy, 1993; Stasack et al., 1996; Taçon et al., 1997; Watchman and Cole, 1993; Watchman et al., 1997) to cooking residues from ceramic pot sherds (e.g. Stott et al., 2001). Constant technological advances allowed the dating of small samples having as little as 10 μg of carbon (μgC) (e.g. Hua et al., 2004) extending the material range, for example, to even smaller rock art pigments (e.g. Zoppi et al., 2004).

It is important to discern between the original sample weight and the yielded carbon mass size. The actual weight of the original sample typically ranges "between 4 to 10 times that of the final amount of graphitic carbon used for the measurement itself" (Taylor et al., 2014:117). The amounts required depend further on the homogeneity of the material, the carbon content of the material, the sample pre-treatment required and the sample age. For instance, charcoal contains generally between 50% and 60% carbon, marine shell has 12% carbon and the carbon content of bone is dependent on diagenetic or biogeochemical factors and can vary between 1% and 30% (Taylor et al., 2014:117).
More recent developments in AMS dating have further reduced the required carbon size to 5 μg (Smith et al., 2007, 2010a, 2010b; Yang et al., 2013). This significant size reduction and the above described advances in residue analysis instigated this thesis to further examine the feasibility of AMS for dating, in particular, plant residues recovered from stone tool surfaces. Logically, the smaller the sample the bigger the role contaminants play in the dating process. Therefore, this study concurrently investigates contamination that interferes with accurate dating processes.

1.2. Definition of the problem

From the former paragraphs it is clear that direct residue dating – once an established method – will provide an accurate chronology of archaeological artefacts, especially for those lacking firm stratigraphic context.

Archaeologists will have an extended range of dateable material, which could result in higher resolution chronologies being developed. The significant value here is that the last use of objects can be dated directly by their inherent use-related organic remnants. This direct dating may not only be more accurate than dating surrounding material, but also could allow a validation for age determinations achieved by relative dating, typology and dating stratified organic material. For instance, it could provide information on artefact movement through stratigraphic layers. Tool movement could be indicated when residue dates are not in accordance with organic material dated from a sediment layer. Conversely, the date of the strata could be questioned if several residue dates suggest a different age. Furthermore, the method holds the potential to provide archaeologists with an important independent dating tool for the majority of archaeological sites found worldwide: open
surface assemblages. Commonly these sites lack other datable organic material associated with the find context. Even if further organic cultural remnants (e.g. hearths or bones) are present on a site, their temporal association with an assemblage is uncertain. Theoretically, the remains may only represent a momentary snapshot of the site occupation, while the open site may have been visited repeatedly for hundreds or thousands of years (e.g. Bond, 2012). Therefore, the refinement of direct residue dating could provide a valuable tool to recreate site chronology and time depth of past human visits.

The first challenge that must be recognized and then overcome is contamination on small sample size radiocarbon dating. As contamination can occur in any of the stages (e.g. post-depositional, fieldwork, storage, handling), how to mitigate or prevent contamination transfer needs to be researched. The establishment of precise step-by-step protocols that build on previous research (Keeley, 1980; Loy, 1987, 1993) was one intention of this research in order to achieve accurate dating of stone tool residue. Furthermore which residue types are suitable for dating initially needs to be determined.

1.3. Knowledge gaps – the role of contaminants and suitable residue types

The discussion above, which is more extensively presented in the literature review (Chapter 2), reveals several key knowledge gaps relevant for residue radiocarbon dating. These issues must be carefully investigated in order to develop satisfactory dating protocols: (i) contamination pathways, sources and transfer; (ii) contaminant identification, both optical and biochemical; (iii) contaminant isolation and treatment; (iv) residue type suitability; (v) residue pre-treatment; (vi) requirements of ultra-small residue samples from
lithic tools (vii); testing and comparison to samples with reference ages; and (viii) adequate sampling strategies.

1.4. Research limitations

While solving these problems requires long-term research, this thesis commences the task step-by-step, in this direction, by investigating six interconnected issues as described in the research aims and objectives.

1.4.1. Financial and instrument access limitations

Firstly, one of the limitations in this research is economical. AMS radiocarbon dating of small sized residue remains is an expensive enterprise: it requires the application of costly equipment (the accelerator). Furthermore, specific treatments of residues from archaeological substrates need to be considered and factored in as an additional economic aspect. Secondly, high precision AMS instruments are not available everywhere. Access to specific scientific facilities is required.

The work presented here was undertaken at the Australian Nuclear Science and Technology Organisation (ANSTO) in Sydney, following the obtainment of an AINSE research grant (award no. ALNGRA11032), which enabled the AMS dating of ten residue samples. For the preliminary study, the AMS results were obtained from ancient plant samples deliberately transferred onto replicated stone tools, then extracted in the laboratory. The successful age agreements of this first study and the inspiring collaboration between Andrew Smith (ANSTO Physicist and Principal Research Scientist) and Fiona Bertuch (ANSTO AMS Chemistry Research Officer) and the PhD candidate resulted in
securement of funding to date a further ten stone tools from archaeological contexts. The second study was generously financed by the ICCAS project from the Institute for Environmental Science – part of ANSTO.

1.4.2. Time limitations of a PhD research project

Time was obviously another limiting factor. Some aspects will require several years of careful study before reaching acceptable results. Therefore, the aims for this study were formulated in a tight, restricted and coherent manner as outlined in the research aims.

1.5. Aims and objectives

The objectives of this investigation were to evaluate the feasibility of AMS radiocarbon dating of plant residues on stone tools, to expand knowledge about contaminant impact on obtained dates and to find mitigation measures when preparing ultra-small carbon samples. The aim of this thesis is to address six main issues related to radiocarbon dating plant residues from lithic artefacts.

1) Systematic testing of carbon introduction impact during sample preparation in the laboratory:

This initial study tests the hypothesis that if all known contamination sources are excluded, residue radiocarbon dating should be possible unless laboratory introduced contamination impacts age measurement agreements. The problem was addressed by excluding sources of contaminants from artefact retrieval, handling and storage, and factors including fungi.
influence (e.g. Barton, 2009:134), as well as post-depositional factors (Haslam, 2004; Langejans, 2011; Wadley and Lombard, 2007:1003). For the first time, an evaluation of laboratory-introduced contamination on dated residues from lithic artefacts was possible. This was achieved by using experimentally produced stone flakes in combination with a known-age plant sample (wood and peat from preceding environmental studies of the Far North Coast of New South Wales and South East Queensland [Boyd et al., 1997; Peters, 1990]). The scope of samples analyzed was tailored to the economic means (AINSE research grant for ten samples using AMS dating).

2) Feasibility of plant residue dating using archaeological stone tools with a known age:

To explore the feasibility and accuracy of AMS dating on archaeological stone tools with wooden and adhesive residues, a case study was conducted using previously dated artefacts. Use-related and contaminant residues were identified by microscopic analyses.

3) Contaminants identification and removal:

To identify and remove introduced contaminants from archaeological artefacts, a series of experimental and analytical tests were conducted. Initially, contaminants were identified by microscopic analyses in situ on the tool. Further contaminants were detected by using Scanning Electron Microscope-Energy Dispersive X-ray analysis (SEM-EDX). Residue type tailored removal strategies were elaborated.

4) Suitability of wood and adhesive residue types for AMS dating:

This evaluation was based on the assumption that various residue types by their nature contain different properties making them more or less suitable for radiocarbon dating. Constricting aspects may include susceptibility for contamination introduction, insufficient
homogeneity (intermix with multiple materials) and insufficient remnants of particular residues on a stone tool surface.

5) General assessment of dating residues from archaeological artefacts:

In particular, the interconnection between small sample size radiocarbon dating and residue identification, preservation, contamination and residue type was examined. This served the further objective to develop adequate sampling strategies.

6) Develop sampling protocol strategies to enhance residue radiocarbon dating:

This study section provides practical advice and easy-to-follow protocols for artefact retrieval, handling, storage, analysis and extraction in order to minimize contaminant and carbon transfer and thus to enhance future residue dating accuracy. This research builds on results gained from the previous study sections and on analysis of various aspects relevant for residue radiocarbon dating.

1.6. Research design

To meet the overall aims and objectives of this study, research strategies were planned in such a way that each research step consecutively built on previous results. For instance, before a method was applied on the residue of an artefact, a technique was first tested experimentally. Hereby significant experience was gained in sample processing and handling and at the same time challenges and problems were identified and could then be addressed. One important reason for this approach is that residues (or parts of them) are removed from the artefact and destroyed during the radiocarbon dating process. Therefore this step needed to be justified before being instigated. In addition, thorough microscopic
recording and documenting of an artefact’s residues was considered mandatory before residue removal. Experimental designs also allow the exclusion of certain parameters that are present in archaeological contexts (such as post-depositional contamination) and thereby permit the targeting of a specific research goal.

1.6.1. Phase 1: Pilot experiment

The first step tests the feasibility of residue dating under controlled laboratory conditions when contaminants from the field, storage and handling are excluded. This allowed evaluation of potential contaminant transfer and resulting age offsets when pre-treating residues in the laboratory. The assumption was that if carbon introduction in the laboratory could not be controlled, there would be little chance for success in applying the method on artefacts from archaeological contexts. An initial experimental study also permitted a suitability assessment of various residue types for AMS dating.

The experimental set up involved replicated stone flakes and contemporary plant and animal materials, as well as previously dated wood and peat from an experimental study. The materials were applied onto the artefacts by cutting and scraping the wood and peat material. Generated residues were then removed by using an ultrasonic bath and microscope monitored scalpel scraping. The samples containing the previously dated wood and peat were then further processed for radiocarbon dating in the ANSTO AMS pre-treatment laboratory.

The study has shown the technique’s feasibility for one important step in the residue dating process – in the laboratory environment. This was preparation for the next step: the further application on archaeological artefacts (Figure 1.1).
1.6.2. Phase 2: Case study

Keys for the second step in the research strategy were the use of stone tools that (i) have shown ample residue preservation and (ii) have a dated archaeological context that functions as a reference age for obtained residue dates. In addition, the use of residue types that promise success in radiocarbon dating required evaluation. Here the choice fell on wooden residues as they showed successful residue dating results in the previous experimental study, as well as possessing material homogeneity and general good preservation qualities. In addition, adhesive material was elected due to its toughness and frequently more abundant preservation on stone tools. This stage included further important steps in need of evaluation and relevant to successful dating, for example: potential contaminant introduction into the sample from the field, storage, handling and/or examination. Also, contaminant removal techniques were examined and identification methods were addressed.

1.6.3. Phase 3: Sampling strategies

The results from the experiments and case studies, along with further clues from the literature, were used to enhance the method’s applicability. The interconnected relevant
key issues – important in relation to small sample sized radiocarbon dating, which needed to be examined – were: (i) residue identification, (ii) residue preservation, (iii) contaminant introduction and (iv) interconnectedness of sample size, yielded carbon mass and residue type. An in-depth analysis of these topics provided further clues to enhance the accuracy of residue radiocarbon-dating while at the same time minimizing contaminant transfer. Investigated aspects were synthesized into new strategies for sampling in all stages of artefact treatment, designed to enhance small sample size radiocarbon dating.

1.7. Study materials and study sites

A comprehensive description of materials and methods used in Phases 1–3 are presented in chapters 3-5, which present each research step and its outcomes thoroughly. The outline below serves as a brief overview complementary to the research design section.

1.7.1. Phase 1

The experimental study component involved the use of replicated stone tools. For a first testing, 21 flakes were fabricated from a flint core (by Jürgen Junkmanns, owner of an archaeological experimental workshop [www.pfeil-bogen.de]). The flakes were used to conduct cutting and scraping activities on a range of organic materials such as bone, meat, fern, horn, antler and two kinds of hardwood (Figure 1.2). Subsequently, the flakes were air dried for two weeks and afterwards stored in sealed plastic bags. After six months, a microscopic analysis showed that wood and fern yielded the highest amount of residue preservation on the stone tools. Therefore, stone tools with these materials were chosen for
the succeeding experimental phase to increase knowledge with residue removal processes and to assess extracted residue quantities.

**Figure 1.2.** Initial trial: scraping and cutting movements with replicated stone flakes on contemporary materials.

In the second part of the experiment, replicate stone flakes fashioned in the laboratory were used to work ancient plant material in a laboratory environment. Five stone flakes, measuring 19 mm to 49 mm in maximum length, were struck from a chert core, using a copper headed stick. Wood and peat samples obtained from earlier environmental studies on the Far North Coast of New South Wales (NSW) and South East Queensland (QLD) (Boyd et al., 1997; Peters, 1990) and thus of known age were used to imitate plant processing with the flakes (Figure 1.3).
Laboratory equipment used during this phase included a sonicator (LEO Ultrasonic Cleaner, LEO-50); Milli-Q™ water; low and high powered microscopes (up to 100x and 1000x magnification, respectively); starch free gloves; clean unused scalpels; centrifuge tubes; centrifuge; and chemical and physical pre-treatment protocols (as outlined in Hua et al., 2001).

1.7.2. Phase 2

The challenge for the second research stage was finding stone artefacts from a well-dated context and on which sufficient organic residues were preserved. While obtaining such material was not an easy task, permission was finally granted to examine lithic tools from three different assemblages. These artefacts meet the terms mentioned above and derive from the German sites Friesack (five stone tools) and Wesseling (one stone tool) as well as the Australian site Yelgun (one stone tool) (Figure 1.4).
Ideally these stone tools would have been retrieved and stored with contaminant mitigation in mind. However, curation contamination was immediately visible in particular in the form of pencil graphite from labeling on the stone tools' surfaces. An experiment was set up aiming to find removal techniques for these contaminants, while leaving birch bark tar residues on the tool surface. The experiment involved ten replicated chert stone flakes, contemporary produced birch bark tar, various pencil types and a range of removal (cleaning) techniques as listed in Figure 1.5.

Figure 1.4. Map 1: showing the sites Friesack and Wesseling in Germany, Map 2: indicating the location of Yelgun, northeast NSW in Australia (map designed by Greg Luker, SCU GeoScience).
**Figure 1.5.** Experimental set up to evaluate cleaning techniques as preparation for sample pre-treatment for AMS dating. 1: proximal end of ventral surface of fabricated stone flakes before treatment, 2: fabricated flakes with attached contemporary birch tar and pencil graphite lines on proximal end of ventral surface, 3: removal technique, 4: proximal end of ventral surface of fabricated stone flakes after treatment. G = Graphite, BT = Birch tar; 1, 2 and 4 = microscopic images of 7 x to 10 x magnification ranges.
1.7.3. Phase 3

This study phase used the results from the previous research steps as well as re-evaluation of specific literature to synthesize the outcomes into adequate sampling strategies for lithic residue radiocarbon dating.

1.8. Archaeological study sites

1.8.1. Friesack 4

Friesack 4, a bog site located in Brandenburg (Germany), was excavated in the 1970s and 1980s and the site's published record contributed considerably to knowledge about the Early to Late Mesolithic periods.

Hunter-gatherer populations repeatedly visited the site for approximately 3200 years, which was then a lake shore. Excavation revealed about 100 Mesolithic layers identified in five different trenches. According to radiocarbon dates, Mesolithic settlement first began in the middle Preboreal period, around 9000 cal BC (Gramsch, 2001, 2006), and ended around 5800 cal BC during the Early Atlantic period. A hiatus of several hundred years was recorded during the middle Boreal period. Neolithic occupation was evident in the Later Atlantic period (Gehlen, 2009; Görsdorf and Gramsch, 2004). To date, Friesack represents the most comprehensive stratigraphy known from the Mesolithic period in Europe. The excellent preservation conditions allowed ample wooden and antler objects, countless bones and 140,000 stone artefacts to be unearthed (Gehlen, 2009; Gramsch, 1990, 2001, 2006, 2009/2010, 2011).
Use wear and residue analyses conducted on 306 stone tools revealed frequent preservation of hafting residues (adhesives), along with plant remains from wooden shafts (Pawlik, 2011a). In addition, Gas Chromatography Mass Spectroscopy (GC/MS) analysis on four samples (three pieces of tarry black material, two of which had chew marks, and one sample of tarry dark material adhering to a bone point) revealed the use of birch bark tar at the site (Baumer and Dietemann, 2008).

Five artefacts, three core axes, one scraper and one flake fragment with macroscopically visible dark residue concentration (Figure 1.6), all from dated stratigraphical contexts, were loaned for this study. Microscopic analysis revealed wooden residues in the form of wood fibers within retouch bows of the 'working edges' of the scraper and the three core axes (Figure 1.6, images 1–3). The latter showed working edges on both ends, confirming previous analyses by Pawlik (2011a). The flake fragments’ residue showed characteristic features for adhesive material such as droplet appearance and some plant structure; clear associated wear marks were, however, not recognized.

Figure 1.6. Mesolithic flint stone tools from Friesack 4 (from left to right): 1-3 core axes, 4 scraper and 5 flake fragment with macroscopically visible dark residue.
1.8.2. Wesseling

The open site of Wesseling is located within an old channel of the Rhine River in the western part of Germany. Excavations revealed six activity zones with typical Late Palaeolithic stone tools such as backed points, backed knives, scrapers and burins. The site also contains pebble plasters interpreted as working areas, several sandstone grinding plates and flat, geometrically-shaped, brown coal objects (Heinen, 2008; Heinen et al., 2010). Four AMS radiocarbon dates suggest an approximate date of ~11,500 BP for the site's occupation (AMS-Labor Erlangen, 2010). The chert flake used for this study had macroscopically visible dark residues present (Figure 1.7) and microscopic analysis showed droplet appearance and thus potentially classifies the deposit as adhesive material. Hafting resembling wear traces were not observed.

1.8.3. Yelgun

The open site Yelgun is located in the Byron Shire of northeastern New South Wales, Australia. Artefacts comprising 159 lithic tools and 60 ochre pieces were found as a surface scatter on a ridgeline overlooking a coastal shoreline and plain. This assemblage was held in a private collection and loaned for analysis, within this study. The stone tools were found prior to 1967. Therefore they were not required to be registered with the New South Wales’ (NSW) Office of Environment and Heritage (OEH). The assemblage is composed of cores, flakes, scrapers, ground edge hatchets, a grinding dish fragment and a burin-scraper combination tool (Figure 1.8). A Late Holocene use of the site or parts of the...
assemblage could be indicated by eight stone artefacts of the bungwall pounder type (e.g. Hall et al., 1989).

Figure 1.8. Lithic assemblage from Yelgun, NSW, Australia. From above left to bottom right: Grinding dish fragment, bungwall pounders, cobbles with use wear or pitting marks, ground edge hatchets, flat chalcedony piece with cortex, flakes, cores and tools sorted by raw material.

Raw materials consist mainly of diverse cherts, as well as a range of other materials including chalcedony, basalt, quartzite, quartz and silcrete (see Figure 1.9). Greywacke artefacts consist of ground edge hatchets, a few flakes, bungwall pounders and a fragmented grinding dish (Figure 1.9). The latter two artefacts indicate on-site food processing.
Further preliminary analysis of the assemblage also suggests on-site tool production. This is indicated by core preparation and core maintenance artefacts as well as cobbles with pitting marks. While these cobbles, and a further chalcedony pebble fragment with rolled cortex, point to procurement from river or creek sources, the coarse cortex of chert varieties suggest primary source provenance. Interestingly, the chalcedony pebble showed macroscopically visible, dark, patchy residue traces (Figure 1.10). A microscopic analysis showed a morphology suggestive of resin, such as a mud cracked structure (e.g. Fullagar, 2006a; Robertson, 2011) of the deposit. The amount of
residue present on the tool, coupled with a potential chronological allocation of the assemblage to the Late Holocene period (as indicated by associated bungwall pounders), made this artefact an ideal candidate for residue radiocarbon dating. Ample residue occurrence on the one tool enabled multiple dating procedures, such that the radiocarbon dating accuracy could be validated. It also allows for replication of dating in the future. In addition, insights into dating residues from open site artefacts might be deductible.

At the beginning of the study it was tried to obtain Australian stone tools with use related residues which also have a better provenance and dated context. Obtaining such artefacts was hampered by the fact, that residue dating was not an established method. Consequently, there was reluctance to use these stone tools' residues for dating. - The mostly small amounts of residues on these stone tools would have been irreversibly destroyed by the method, without guaranty of a correct age measurement. Therefore the Yelgun stone tool with ample adhesive residue offered an opportunity to use the method with concurrently allowing duplication of dating this residue in the future. Assuming that the entire assemblage is of one age, the presence of bungwall pounders could point to a Late Holocene Age.

In the following chapter 2, a literature review provides specific background information relevant for residue radiocarbon dating. This includes the wider field of residue and use wear analyses, term definitions, methods, contamination and preservation issues as well as a research history about radiocarbon dating residues from stone tools.
CHAPTER 2

A review of residue analyses aspects relevant to AMS radiocarbon dating
CHAPTER 2 A REVIEW OF RESIDUE ANALYSES ASPECTS RELEVANT TO AMS RADIOCARBON DATING

2.1. Introduction

Research in archaeological residue analyses has gained significant ground since Briuer (1976) provided evidence for organic residue survival and demonstrated that plant and animal residues on stone tools can be discerned. His examinations, undertaken in a crime laboratory, involved microscopic morphological identification supported by chemical techniques. Briuer's approach was further examined and developed by a number of researchers from the 1980’s onwards and has subsequently become a rapidly advancing field in archaeological science.

In this literature review the broad field of residue and use wear analyses will be explained, definitions will be determined and methodologies will be presented. Secondly, key issues related to AMS radiocarbon dating will be elaborated in. These include contamination transfer to archaeological artefacts and preservation. Furthermore, examples of previous attempts to radiocarbon date residues from stone tools will be presented. After these aspects are reviewed and the knowledge gaps are identified, directions, strategies and methods for present investigations can be defined.

2.2. Definitions, methodology and case studies

2.2.1. Definitions

2.2.1.1. Residue analysis

Archaeological residue analysis is a method that seeks to interpret use traces left on an artefact. As Briuer (1976:478) clarified, this approach goes back to forensic science research, which observed that contact between two materials always results in the
transference of material from one object to the other. These traces may be of a use-related nature, or may have been transferred incidentally by post-depositional processes or after artefact retrieval. To discern the former from the latter, in this study, 'use-related residues' on stone tools are defined as "functional traces" that "refer to materials that are transferred and adhere to an artefact" (Fullagar, 2006a:208). Conversely, 'non-use-related residues' are defined as those which can be transferred to an artefact incidentally, either during the use life of the artefact, post-depositional or after artefact retrieval.

To validate that residues are of functional origin, analysts use a combination of residue and use wear analyses. The importance of this validation was already recognised by Briuer (1976:478), who incorporated in his analyses microscopic edge wear patterns associated with residue occurrence. This approach was further developed by elaborating interpretation keys and was introduced by Fullagar (1986) as 'integrated' microscopic residue and ‘use-wear analysis’, and was applied to Australian artefacts. From this time onwards, this advance has been used in numerous research projects worldwide (e.g. Fullagar and Jones, 2004; Haslam, 2006; Robertson, 2002, 2005, 2006, 2009, 2011; Robertson et al., 2009; Rots and Williamson, 2004; Hardy et al., 2008; Hardy and Svoboda, 2009).

A 'multi-stranded approach' was introduced (Lombard and Wadley 2007; Wadley and Lombard, 2007) with the requirement to view residues not only in association with use wear, but also in their relation to: each other; distribution patterns; orientation; frequency; layering and how they adhere to a tool. The development of this approach was influenced by significant finds of residue preservation in South African Middle Stone Age sites such as Rose Cottage Cave and Sibudu Cave (e.g. as summarised in Lombard and Wadley, 2009).
2.2.1.2. Use wear analysis

Use wear analyses of stone artefacts is a method that describes wear attributes resulting from a task that has been carried out with the tool. Features of wear, therefore, can provide information on tool function. Semenov’s (1964) research was pioneering in establishing systematic characteristics for use wear traces. He recognised the correlation between work motion and striation and interpreted specific activities such as wood-working. While his interpretations were debated, further research accumulated on his work (Bamforth, 1988; Fullagar, 1986; Hurcombe, 1992; Kamminga, 1982; Keeley, 1982; Vaughan, 1985) and described various forms of use wear-causing stone tool surface alterations including edge rounding, scarring, polishing, soothing, striations and sickle gloss. Fullagar (2006a) gave interpretation explanations to both wear traces and residues.

2.2.1.3. Use-related residues

Use-related residues found on stone tools can be of an organic and inorganic nature. While organic residues on stone tools were recognised by Semenov (1964), discernment between animal and plant residues was attempted by Briuer (1976), employing a combination of use wear and residue analyses as well as the use of chemical reagents. In this study Briuer also cautioned that residues can be removed by artefact cleaning. The interpretation and description of use-related residues were further refined by successive studies.

Animal residues include animal lipids, collagen, bone, hair, tissue, fibres, blood, feather barbules and scales. Plant material observed is composed of plant tissue and fibre, wood fibre, starch, phytoliths, raphides and adhesive materials (resin, tar, gum, wax) (e.g. Fullagar, 2006a; Kealhofer et al., 1999; Langejans and Lombard, 2015; Robertson, 2005,
2011). An exception is residues of bitumen, a viscous or solid mixture of hydrocarbons (Collins English Dictionary, 2015), which is well known for its use as an adhesive, sealing, or mummification agent (e.g. Aufderheide et al., 2004; Boëda et al., 1996, 2008, 2009; Monnier et al., 2013).


In a publication that demonstrated the multiple use and versatility of Australian backed artefacts, by applying residue and use wear analyses, Robertson (2011) compiled previous work on residue types related to task and function traces. Information for this work was originally derived from her PhD thesis (Robertson, 2005): an overview is presented in Table 2.1 for animal residues [Robertson (2011:87)] and Table 2.2 for plant and inorganic residues [Robertson (2011:88)]. These tables provide great insights about how multiple residues can occur combined with several use wear marks, from performing a single task.
Table 2.1. Residue and use-wear criteria for hypothesized task associations and functions (adapted from Robertson, 2011:87).

<table>
<thead>
<tr>
<th>Task association</th>
<th>Description of functions</th>
<th>Residues</th>
<th>Use Wear</th>
</tr>
</thead>
<tbody>
<tr>
<td>Primary animal processing</td>
<td>Butchery Cutting skin and disarticulating bone, removing meat from bone.</td>
<td>Blood*, red blood cells*, proteinaceous film*, fibrous collagen*, muscle tissue, other connective tissue (collagen), fat, vivianite, hair, feather.</td>
<td>Small bending and step flake scars, slight edge rounding. Intersecting striae and striae parallel to the cutting edge.</td>
</tr>
<tr>
<td>Hunting</td>
<td>Impact/spearing, stabbing—mammal, reptile, bird.</td>
<td>Blood*, proteinaceous film*, tissue fragments*, collagen, hair, scales, feather, hafting resin.</td>
<td>Damaged tip—possible high energy impact shattered, striations parallel or sub-parallel to the long axis, major damage to retouched edges/large microflake scars. Finely striated polish.</td>
</tr>
<tr>
<td>Bone-working</td>
<td>Primary stage Scraping and cutting to remove flesh, sinew and periosteum.</td>
<td>Blood*, bone collagen* (sheet—periosteum, fibrils), bone fragments, connective tissue (collagen), fat and oil droplets, vivianite.</td>
<td>Edge rounding and fracturing, striations at 45–90° to the edge, possible retouch.</td>
</tr>
<tr>
<td></td>
<td>Secondary stage Cleaning of periosteum by scraping or cutting, smoothing and shaping/modified bone.</td>
<td>Sheet collagen* (periosteum), collagen fibrils*, bone fragments, granular bone collagen, proteinaceous film*, vivianite.</td>
<td>Edge rounding and fracturing, striations perpendicular and/or parallel to the edge.</td>
</tr>
<tr>
<td></td>
<td>Tertiary stage Working dry bone, engraving, smoothing, polishing, drilling, possibly sawing, adding fine detail, possibly decorating with ochre.</td>
<td>Granular bone collagen*, sheet collagen, bone fragments, ochre*, vivianite.</td>
<td>Sawing: continuous distribution of bending and step flake scars, feather fractures, rounding/smoothing, protein film or “polish” on ventral and dorsal aspects, striations parallel to the worked edge, but rare; engraving: edge rounding, “polish”, small step and bending scars; drilling: rounding of apex and associated lateral margins, bending and occasional step fractures on lateral margins, frequent tip snapping but continued use, and polish.</td>
</tr>
</tbody>
</table>
Table 2.2. Plant and inorganic materials: Residue and use-wear criteria for hypothesized task associations and functions (adapted from Robertson, 2011:88).

<table>
<thead>
<tr>
<th>Task association</th>
<th>Description of functions</th>
<th>Residues</th>
<th>Use-Wear</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wood-working</td>
<td>Scruping, chopping, cutting, incising, adzing.</td>
<td>Woody plant tissue*, cellulose*, plant cells visible in tissue, cellulose fibres*, resin, plant sap or exudate*, bordered pits, cells with helical wall-thickening, charcoal, charred sap/resin.</td>
<td>Scrapping: edges, both acute and obtuse, exhibit moderate rounding with an almost continuous distribution of bending, and occasional feather and step flake scars, striations are generally broad and shallow if present; polish, incising/engraving: some edge rounding on the tip and lateral margins at the tip, with small step fractures, striations parallel to the working edge.</td>
</tr>
<tr>
<td>Plant processing</td>
<td>Cutting, shredding, removing bark (scraping).</td>
<td>Plant tissue*, amorphous cellulose fragments*, fibres, resin, sap or exudate*, small starch grains*, chlorophyll, phytoliths.</td>
<td>Edge-scarring rare and small, usually only bending flake scars, slight to moderate rounding and few or no striations.</td>
</tr>
<tr>
<td>Starchy plants and seeds</td>
<td>Scruping, chopping, cutting.</td>
<td>Starch grains*, cooked starch, plant tissue*, raphides, amorphous cellulose, plant fibres*, phytoliths.</td>
<td>Edge-scarring rare and small, usually only bending flake scars, slight to moderate rounding and few or no striations.</td>
</tr>
<tr>
<td>Ceremonial and Decoration</td>
<td>“Crimping” or cutting to form cicatrices on the body.</td>
<td>Blood/human*, connective tissue, lipids/fats, vivianite.</td>
<td>None.</td>
</tr>
<tr>
<td>Miscellaneous</td>
<td>Decorating churinga or other object or body.</td>
<td>Ochre, feathers, blood, muscovite mica or none.</td>
<td>Unknown, possibly none.</td>
</tr>
</tbody>
</table>

Key
- Essential residues
  - Hurcombe, 1992:148
  - Akerman et al., 2002
  - Fullagar, 1986:172–191
  - Anderson, 1980
  - Fullagar, 1992
  - Briuer, 1976
  - Kamminga, 1982
  - David, 1993:77–79
  - Kay, 1996
  - Dockall, 1997
  - Kooyman et al., 1992
  - Loy, 1994
  - Hardy & Garufi, 1998
  - Tringham et al., 1974:189–191

- Unknown, possibly none.
- Fullagar, 1986:172–191
- Fullagar, 1992
- Kamminga, 1982
- Loy, 1994
- Tringham et al., 1974:189–191
2.2.1.4. Non-use related residues

Non-use related residues or contaminants refer to traces incidentally transmitted to a stone tool surface. This can occur any time after the artefact was deposited. These contaminants include post-depositional fungi, insect remains and plant fibre as well as artefact retrieval and curation contaminants such as finger grease, skin scales and fabric fibre (Langejans, 2012b).

2.2.2. Methodology

2.2.2.1. Optical interpretation

Residues are differentiated by morphological characteristics. By monitoring the \textit{in situ} location of residues under a microscope their association with use wear can be ascertained. Various lighting conditions help in the understanding of residue morphology and use wear patterns. In addition to Briuer's (1976) methodological approach, Odell (2004) suggested determining the distribution of residues in use areas of the artefact and to analysing the surrounding sediment to ascertain the use-relatedness of these residues.

Fullagar (2006a) presented detailed interpretation keys for morphological residue identification and use wear analysis. This work offers protocols and forms to standardise interpretation and aims to provide comparability. The ideal methodological sequence was described as starting with microscopic analyses at low 10x to 50x magnification and then continuing with up to high 1000x magnification. At the beginning of the sequence is residue analysis without prior cleaning of the tool. After residues and use wear traces are documented, it can be determined whether cleaning is required to see edges and further wear-traces more clearly. Cleaning of adhering sediment can involve gentle rubbing in a
plastic bag or using an ultrasonic bath with distilled water and floating boats. Removed residues (and sediment particles) can be captured in capped tubes and from there transferred to glass slides, which can be then observed by transmitted light microscope under high magnification. Also, further in situ residue and use wear analyses can follow and can be extended to high magnification microscopic observations. This approach was used by a number of residue and use wear analysts globally (e.g. Hardy et al., 2008; Hardy and Svoboda, 2009; Haslam, 2009; Lombard, 2008; Robertson, 2008, 2009, 2011; Robertson et al., 2009). Subsequent work further contributed to elaborate clues for morphological residue interpretation (Langejans and Lombard, 2015; Lombard, 2008; Robertson, 2005, 2011). For example, microscopic interpretation clues can include a mud-cracked structure, droplet appearance and occasional occurrence of plant tissue for resins (Fullagar, 2006a) or for plant tissue, bordered pits (Robertson, 2009, 2011). There is a general consensus, that residues are ideally observed in situ and removal recommended mainly when the artefacts' size does not permit microscopic observation (e.g. Hardy and Garufi, 1998; Langejans and Lombard, 2015).

2.2.2.2. Chemical identification

Chemical classification may confirm the nature of the material. Gas Chromatography Mass Spectrometry (GC/MS) is able to characterize and discern multiple constituents of a material. GC/MS has been applied on various residues successfully (an extensive overview is given by Evershed, 2008). However, the method requires relatively substantial amounts of ~ 0.5 mg pure organic material (Baumer 2014, personal communication), which exceeds the amount of most residues found on stone artefacts. In addition, the technique also destroys the sample during analysis. In contrast, SEM-EDX — previously helpful for the
characterization of birch bark tar (Pawlik, 1997, 2004; Pawlik and Thissen, 2011) — is non-destructive. The technique allows for the elemental composition and imaging of residues and this helps in validating the organic nature of residues. Recent studies show non-destructive chemical residue characterization is important in quantifying the substance to be dated and in confirming optical residue identification (Cesaro and Lemorini, 2012; Daher et al., 2013; Monnier et al., 2013; Prinsloo et al., 2014). For instance, microscopic identification of blood residues is not always straightforward and there is potential for misidentification with other residues such as resins. The use of biochemical reaction methods, such as the Hemastix™ test, help in the identification of blood residue, although other substances can cause false positives (Manning 1994). Forensic methods may help in identifying blood residues on lithic tools thousands of years old (Lombard, 2014).

2.2.2.3. Experimental studies to understand residue behaviour on lithic artefacts

Experimental residue studies contributed to the understanding of residue behaviour. Gurfinkel and Franklin (1988) examined the survival of blood in two test series: first, by burying blood stained slides for six weeks and second, for one year. By using urinalysis test strips it was suggested that four out of five samples after the six weeks still contained blood, as did two out of three samples from the one-year samples.

A further study investigated the feasibility of species identification (Hyland et al., 1990). For this, deer, dog and human blood dilutions were applied on 40 stone artefacts, which were dried at different temperatures. The samples were later tested with enzyme immunoassays with polyclonal antibodies, which could identify the species correctly, except for the species that were allocated to the boiled sample group (Hyland et al., 1990:109).
In order to test the extent to which blood on stone could be detected and identified immunologically to species level in buried material, four outdoor experiments were set up by Cattaneo et al. (1993). For time periods from one month up to two years, human and bovine blood was deposited on replicated flint artefacts and within bone samples. By using a specifically developed "enzyme-linked immunosorbent assay (ELISA) with monoclonal antibodies to detect blood in ancient contexts" (Cattaneo et al., 1993:29), the study aimed to identify specific human or bovine blood proteins albumin and IgG. The results showed some success. For instance, one scraper, after one year's burial, tested positive for bovine albumin, while bone fragments tested positive for human albumin after two years burial. It was concluded that blood can survive on flint flakes for at least two years. The authors further pointed out that that drying of blood prior to burial, and the type of burial sediment, could be critical for lasting survival of blood residues. In relation to this, the authors refer to Loy's (1987:58) observation, that soils containing a high clay-silt content (greater than 20 per cent) could be significant for long-lasting blood residue survival.

Further experimental work on 13-year-old blood residues applied on stone tools showed that they can survive several washes (Shanks et al., 2004). Sonication yielded significantly more residues than were visible on the tool surface. It was argued that tool surface residues degraded faster, while residues in micro-cracks were protected from degradation (Shanks et al., 2004:669–670).

Experimental studies on plant residues are not that common. Yet, a residue and use wear study on experimental woodworking on stone tools (Hardy and Garufi, 1998) helped in the recognition of such traces in the archaeological record. In the study, 100 test series of wood processing were carried out using replicated, unmodified flint flakes to work both hard- and softwoods with six different use-actions: whittling, slicing, incising, scraping,
planing and boring. Besides recognizable use wear patterns in accordance with use-actions, the experiments showed that wood fragments adhering to tool surfaces can be identified by microscopic analysis. The authors highlight that wood, due to its structure, has likely better preservation conditions than other plant residues. They also noted that wood has been found preserved under various environmental conditions, including waterlogged or desiccated situations. Therefore wood (and other plant residues) might be more commonly preserved than previously presumed.

Also, for microscopic morphological interpretation, experimental work was important. For instance, a study by Lombard and Wadley (2007), using modern plant and animal material applied on replicated stone flakes of various rock types as well as incorporating blind tests, helped in addressing difficulties in morphological identification such as discerning animal from plant residues. The authors suggested that some faunal residues could have been previously misinterpreted as plant residue. In their test series they revealed that birefringence, the double refraction of incident microscopic light, is not a characteristic feature of plant residues as presumed previously. Furthermore, morphological similarities, including rectangular and ordered cell structures and the angle at which residues are monitored, could lead to misidentification (Lombard and Wadley, 2007:156). In addition, it could be demonstrated that wood, bark and plant fragments were found incorporated in hafting residues, such as gum that was scraped from a tree (Lombard and Wadley, 2007:158). This important observation needs to be considered when assessing residue types. As in the case of adhesives, the simultaneous presence of wood does not unambiguously indicate wooden hafts or wood working. The authors were further able to discern use-related from contaminant residues. The latter was found to adhere only loosely
on the tool surface, and occurred "on top of use-related residues" (Lombard and Wadley, 2007:158).

In relation to contaminant discrimination, further experiments demonstrated that residue distribution is associated with use-wear patterns (e.g. Kononenko, 2008, 2011) and that there is a differential distribution of use-related and contaminant residues (Langejans, 2011). Use-related residues were described to have generally a more "macerated or smeared appearance" on the stone tool surface and they "can be trapped or jammed into cracks or crevices" (Kononenko, 2008:33). Furthermore, residues "are likely to be use-related when they" are associated with other use-related residues such as "allied residue types" (Langejans, 2011:998). "For example, fat, bone, blood and muscle tissue are all animal residues" and when they have similar distribution patterns, they "are likely to be use-related" (Langejans, 2011:998). Langejans (2011) also found a noticeably lesser amount of non-use-related residues compared to use-related residues and that non-use-related residues have an unsystematic distribution on stone tools.

In the last decade residue preservation and contaminant issues (Barnard et al., 2007; Barton, 2009; Haslam, 2004; Jones, 2009; Langejans, 2010, 2011; Loy and Barton, 2006; Wadley and Lombard, 2007) were subjected to increasing attention to experimental studies. Relevant aspects will be discussed in more detail in subsequent chapters.

2.2.3. Case studies

The here presented case studies serve as examples to illustrate how residue and use wear analyses have contributed to solving research questions.
2.2.3.1. Multiple tool use

It has been shown that Australian backed artefacts were used for many different tasks, regardless of their shape (e.g. Robertson, 2009, 2011), and frequently left different types of residue on the same artefact.

2.2.3.2. Subsistence

While Briuer (1976:483) saw the potential for residue analysis to shed light on "subsistence and procurement strategies," subsequent research contributed to resolving several aspects. For instance, early plant use was identified by two types of taro starch residues found on stone tools in the Solomon Islands dating to 28,000 BP (Loy et al., 1992). Furthermore, the analyses of 30,000- to 37,000-year-old stone tools from Cuddie Springs in New South Wales, Australia, reveal animal meat and plant food processing occurred at this site. Grass seed grinding, identified by use wear and residues on grindstones from Cuddie Springs, indicated a response to climate change 30,000 years ago as humans adapted to a harsher environment and the extinction of megafauna (Field et al., 2006). Further evidence for Pleistocene seed grinding was demonstrated in a recent functional study of 17 stone artefacts from Lake Mungo, south-eastern Australia, 14 of which indicate likely seed grinding (Fullagar et al., 2015). Also, much earlier evidence than expected, about the onset of plant domestication, was demonstrated by the extraction of manioc and arrowroot starch granules from stone artefacts from Panama (Piperno and Pearsall 1998., 1998; Piperno et al., 2000).
2.2.3.3. Open sites surface stone tools versus excavated artefacts

A study by Cooper and Nugent (2009) investigated the potential of applying microscopic residue and use wear analysis to surface collected stone artefacts from Camooweal, Queensland, Australia. A total sample of 23 stone artefacts comprised of four cores, 11 blades, four hand-axes and four tulas from surface and excavated contexts was subjected to analysis. Evidence of residues was observed most frequently, in varying quantities, on all tulas. Further use-related residues include blood, bone collagen and gelatinised starch, while non use-related residues such as sponge, spicules and algae were discerned; their occurrence was related to regular inundation of the site. The authors concluded that a large range of residues preserve on artefacts from both surface and subsurface sites. This study is important, because it provides evidence for use-related residue survival in open site assemblages. This has significant implications for potential future residue radiocarbon dating studies at surface sites, which represent the majority of archaeological assemblages found worldwide.

2.3. Contaminant sources and transfer to archaeological artefacts

2.3.1. Post-depositional

In his residue discrimination research, Briuer (1976) also recognized that, in particular, plant residues can originate from taphonomic processes and thus might not be use-related.

In order to address the issue of post-depositional contamination, Barton et al. (1998) compared the frequencies of starch grains and their occurrence on used and unused artefacts, as well as sediments in which they were deposited. Use wear analysis to
CHAPTER 2 A REVIEW OF RESIDUE ANALYSES ASPECTS RELEVANT TO AMS RADIOCARBON DATING

determine the function was conducted without knowledge about the outcome of the residue analysis. The results demonstrated that the occurrence of starch grains was strongly linked to used edges. This outcome allowed the inference that no great amount of contamination had occurred. The authors recommended the integration of residue analysis into surrounding sediments in addition to detailed studies of use wear.

To control this aspect of potential contamination, the inclusion of surrounding sediment has been integrated in residue studies (e.g. Hardy and Garufi, 1998; Kealhofer et al., 1999). The argument behind such sediment analyses is that if residues present on an artefact are not found in the sediment, then there is a high probability of the use-relatedness of the residue.

Cooper and Nugent's (2009) Australian study suggested that lithics of surface assemblage were affected by downwards movements though cracks in the soil surface, which occurred in dry seasons. Furthermore, it was inferred that sponge spicules, algae and plant tissue were probably transferred to stone tool surfaces "from regular inundation of the site by the Georgina River, rather than use of the artefact for aquatic plant processing". (Cooper and Nugent, 2009:208, 209). The study also raised awareness about environmental additives including "algae, fungal spores and hyphae, pennate, diatoms and insect remains" (Cooper and Nugent, 2009:217).

2.3.2. Starch

The latter aspect was further addressed by Haslam (2004) in a study about the decomposition of starch grains in soils. He also explored the relationship between starches on artefacts and in associated soils. This work refined the problem of use-related residues
versus non-use-related residues in so far that it could be demonstrated that starch grain frequencies in soils are much lower than quantities observed on artefacts. In addition, soil that immediately surrounds the artefact can have higher starch quantities (Haslam, 2004:1726). According to this study, starch survival in the soil is also dependant on soil aggregates, pores and composition that either promote or hinder starch preservation. As implications for archaeological residue analysis, it was deduced that sediment in the immediate artefact environment or adhering to the tool can also be an indicator of the former use of the tool. The latter notion was also observed in a residue and use wear study by Fullagar et al. (1998) on starch grain occurrence in sediments and stone tools from Papua New Guinea.

2.3.3. Fungus

Fungus (Fungi, Pl.) is a collective term for a variety of "eukaryotic single-celled or multinucleate organisms that live by decomposing and absorbing the organic material in which they grow", including molds, mildews, smuts, rusts, yeasts and mushrooms (Collins English Dictionary 2015).

In a study by Haslam (2006) the "typical fungal morphology" was described as consisting:

... of filaments known as hyphae (singular hypha), which as a mass may be referred to as the mycelium. Hyphal cell walls are strengthened by the polysaccharide chitin, as opposed to the cellulosic composition of plant structural components. Some fungi possess macroscopically visible fruiting bodies (for example, the mushrooms), however, the majority are individually microscopic, producing spores for either sexual or asexual reproduction. Spore and fruiting structure morphologies remain one of the most useful classificatory mechanisms for fungi, along with structural aspects such as the presence of septa in the hyphae (Haslam, 2006:116 citing Watanabe, 2002).
Conida, a form of fungal spores and a common soil component, was the subject of a study that sought to raise awareness about potential misidentification with starch grains when performing *in situ* analysis of residues ≤5 μm in size (Haslam, 2006). Confusion between the two residue types can occur, because conida may exhibit extinction crosses (Haslam, 2006:116), which is also the most significant diagnostic attribute used by starch residue analysts to determine starch presence under a cross-polarized light microscope. Differences in the rotation motion of the extinction cross and the use of iodine potassium iodide stain, which stains starch red-brown to blue-black, were suggested as useful in distinguishing starch from conida (Haslam, 2006:118–119). The author concluded that fungal activity may contribute to additional residues on a tool surface, which may lead to misidentification. Furthermore, while fungi add to the decomposition and removal of use-related residues, fungi presence on a tool surface in a concentrated pattern may indicate former residue locations.

As demonstrated by some studies, the actual decay of organic material, once incorporated in soil, happens within a relatively short period of time, such as three days (Haslam, 2004:1721; Loy, 1991, 1993). Also, "fungi and bacteria are the dominant decomposers responsible for at least 95-99% of organic breakdown" (Barton 2007:1753). Morphological identification of (ancient, post-depositional) fungi, in particular when present in clusters, can also be interpreted as an additional strand of previous use-related residue presence (Lombard and Wadley, 2007:162).

When ancient organic residues preserve, some of which possibly are associated with ancient fungi presence on an artefact, both trace remnants should have the same age. Therefore, an attempt to radiocarbon date either of these remnants, ancient residues and ancient fungi, should result in the same age or, in other words, should reflect the residue
Theoretically, this means that ancient fungi would not distort radiocarbon dating ancient residues. How the situation looks when modern fungi gets into contact with the sample will be elaborated further on in this thesis.

2.3.4. Excavation, retrieval and storage of artefacts

Wet sieving has the potential to distort residue analysis. This is illustrated by a residue use wear study on backed artefacts from Turtle Rock, a rock shelter in Queensland, Australia, which showed high frequencies of aquatic plant residues, often associated with used edges (Robertson, 2006). Ethnographic reports supported the inference that the tools could have been used to harvest aquatic plants. Yet, after researching the excavation procedures, revealing that the artefacts had been wet-sieved in a nearby waterhole, the author questioned this interpretation. Consequently, it was inferred that the diagnosed plant residues might have also been transferred to the artefacts from the waterhole. The plant residues therefore may possibly be the result of post-excavation contamination. This concept was supported by the presence of freshwater diatoms, sponge spicules and occasional algal tissue, which were found associated with some plant residues (Robertson. 2006:136).

Food was described as offering risk of contamination in the field (Loy and Barton, 2006). Therefore, the authors suggested washing hands after a meal, but that wearing starch free gloves was ‘overkill’. While the authors' latter notion may be understandable, and a constant wearing of starch free gloves may appear highly impracticable during fieldwork, caution may still be advisable with regard to artefact contact. This view is supported by evidence that cosmetics or sunscreen can be transferred into archaeological samples. For instance, synthetic Camphor, a component of many cosmetics, sunscreen and lip balm was
found incorporated in archaeological lipid residues (Buonasera, 2007:1387). The study investigated the presence of ancient absorbed residues in the milling surface of groundstone tools from California. Gas chromatography (GC) and gas chromatography-mass-spectrometry (GC-MS) analyses were used to identify lipids. Results demonstrated ancient residue presence in milling tools. While measurable amounts of azelaic acid were present on most milling surfaces, the material was not found on broken surfaces (Buonasera, 2007).

As described above, it was theorized that ancient fungi should not distort radiocarbon dating ancient residues, even if they are incorporated in the to-be-measured sample. However, the situation is presumed to be entirely different when, after artefact retrieval, recent (modern) fungi are incorporated onto a tool surface, for instance, by storing archaeological samples in a humid climate. Closed plastic bags are in general too warm for storage. Fungi incorporation as a possible cause for age measurement offsets needs to be considered.

This note of caution is substantiated by a study, in which pollen radiocarbon measurements resulted in too young dates (Neulieb et al., 2013). The authors argued that long storage in a warm environment, inviting fungi and other microbe development and decay, introduces $^{14}$C-rich CO$_2$ into the sample. Attempts to remove these contaminants by pollen preparation with standard treatment might be of limited success, as it is thought that the porous structure of pollen walls allows absorbed materials to remain inside the pollen grain (Kilian et al., 2002:25). To date, it is unknown whether recent fungi activity can be incorporated in ancient residues or affect radiocarbon measurements in other ways. The like sponge and diatoms transferred in archaeological samples by wet sieving (Robertson,
2006) could have the potential to distort radiocarbon dates towards too young age measurements.

### 2.3.5. Post-excavation

Post-excavation contamination was addressed by Loy and Barton (2006), who described contamination transfer in the laboratory by air conditioning (or by an open window with the same effect). The results showed that starch was found on clear nail polish on artefacts (from artefact labeling). Furthermore, feathers, hair, fragments of silt or soil particles, pollen granules and phytoliths can be transported this way. The authors further pointed out that tap water contains potential contaminants and recommended while working with starch (and other residues) to always use Ultrapure water, e.g. Milli-Q water. Gloves could be another source of contaminant. When starch powdered gloves are used they may well contribute significant amounts of airborne starch to the rooms. Therefore, the authors advised that they should not be used in areas where residue studies are conducted. If powder free gloves are not at hand, large forceps with cling wrap wrapped ends were recommended. In addition, the use of cling wrap or clean plastic sandwich bags to cover hands might be an alternative (Loy and Barton, 2006:167).

The extent of contamination by airborne starch became apparent by a study of Crowther et al. (2014). Starch occurrences were tested in two different ancient starch labs and relevant consumables. The authors found that airborne, modern starch grains landing on laboratory surfaces were omnipresent. The results demonstrated that starch contamination occurs in most material in the laboratory, including powder-free gloves, microslides, parafilm, pipette tips, sodium polytungstate, paper towels, lens tissue, sample bags, centrifuge tubes, cling film, and plastic weighing trays. Furthermore, the majority of starch granules were
found occurring in the fume hood, even after cleaning all surfaces and removing consumables. Starch granules were also found on environmental samples such as garments, wall paint, carpets, mats and shoe soles, as well as floor and ceiling tiles. Of decontaminates tested, only 5% NaOH and 5% KOH destroyed three tested starch species immediately. Incineration maintained for 30 s was able to decontaminate metal tools. The authors suspected gloves as being responsible for contamination of numerous consumables tested. They suggested reducing starch contamination from gloves by using brands routinely tested for starch contaminants, using sterilized forceps for grasping samples, and using gloves that can withstand long autoclave cycles of sterilization. The test results further indicated that covering samples with petri dishes was preferable to parafilm, foil, or cling film, due to abundant starch found on their surfaces. In addition, ideal lab conditions include HEPA air filters with controlled airflow and restricted access (Crowther et al., 2014).

### 2.3.6. Case study

Langejans (2012b) reported on a residue study of Early Stone Age artefacts from Sterkfontein, South Africa of which 33 artefacts were analyzed. The artefacts belonged to the around two million-years-old Oldowan and the 1.5 million-years-old Acheulean industries. Previous residue identification was questioned due to the age of the samples. By reviewing first the taphonomic history of the stone tools, and then analyzing the samples, the author concluded that recorded traces were not use-related, but the result of contaminant residues likely transferred from post-excavation handling and curation. Contaminants identified include pollen, hyphae, spores, insect remains, plastic particles, coloured fibres, manganese stains, ink stains, nail varnish, brightly coloured fat deposits
and rootlets (Langejans, 2012b:203). Additional contaminants were identified by their micro-stratigraphy, for instance: when a residue overlay site sediment. A total of 117 potentially use-related residues were observed on the sample. The majority consisted of plant residues, while also some animal residues in the form of fat and bone were present (Langejans, 2012b:207). However, the typical patterned occurrence for use-related residues could not be confirmed for these remains, which raised questions about their antiquity. It was concluded that the residues from the Sterkfontein artefacts were post-excavation contaminants.

2.4. Residue Preservation

2.4.1. Conditions

In a study concerned with starch preservation, Haslam (2004) reported that heavy metals such as lead, copper, aluminium, iron, and zinc are known to lower organic decomposition rates. He noted that, according to studies, the presence of such metals in sediments restrain enzyme and microorganism activity and disrupt the normal interaction between soil, substrate and enzymes. Because metals are capable of forming stable complexes with proteins, they affect the enzyme’s activity. Therefore, starch degradation is decelerated by the reduction of the contact amount between enzymes and starch grains (Haslam, 2004:1725).

Furthermore, it is known that clay-rich soils, an alkaline pH and cation exchange capacity, water and oxygen-free and nutrient-depleted environments are beneficial for residue conservation (Gurfinkel and Franklin, 1988; Jones, 2009; Loy, 1987:58; Langejans, 2010). Desiccation can also conserve residues very well and prevent microbial growth (Evershed,
2008), even though the latter's contaminating impact is still not fully understood. Artefacts buried in waterlogged conditions such as bogs, and generally characterized by a low pH, also demonstrated exceptional residue preservation (e.g. Langejans, 2010). Furthermore, anoxic conditions and nutrient limitation, especially of nitrogen (N) and phosphorus (P), prevent microbial activities and thus limit organic degradation (Evershed, 2008). The susceptibility of biomolecules to structural modification and degradation is described in the order of: “lipids < carbohydrates ≈ lignin < protein < nucleotides” (Evershed, 2008:910). This sequence can change as influenced by environmental conditions. A hydrophobic condition, preventing leaching and microbial decay, is seen as the primary reason for lipid survival (Evershed, 2008). On the other hand, it was found that water in the form of rainfall on surface stone tools did not promote decay of inherent starch residues (Barton, 2009) or phytolith residues (Fullagar, 1993).

An experimental study by Langejans (2010) provided more detail on different residue type preservation. The study revealed bone, fat and wooden plant tissue to be more resistant to decay, while blood, muscle tissue and starch showed noticeably weaker conservation.

It is known that preserved residues can build a protective hydrophobic barrier preventing microbial impact (Barton, 2007, 2009; Barton and Matthews, 2006; Loy, 1987, 1990) and this explains why residues can withstand several washes and are often resistant to extraction (Barton et al., 1998:1233; Fullagar, 1986, 1993; Fullagar et al., 1996; Shanks et al., 2004). In relation to residue protective conditions, Barton (2007:1757) suggested that once residue deposit forms and dries on a tool surface, a proportion of organic material is protected and remains shielded to some extent from microbial activity.
2.4.2. Proportions of stone tools with residue preservation in lithic assemblages

Until 2006, the ratio of stone tools with residues compared to those without was uncertain (Haslam, 2009) because there was a lack of transparency of sample amounts analyzed. More recent research indicates that several assemblages comprise >50% of lithics with residues (Hardy and Svoboda, 2009:165; Hardy et al., 2008:652; Robertson, 2009:300, 302). Three recent case studies are here presented in more detail to illustrate exceptional proportions of residue preservation in lithic assemblages of various time periods and locations.

1) Two Mesolithic sites in northern Bohemia, Czech Republic were selected for functional microscopic residue and use wear and analysis (Hardy and Svoboda, 2009). The study aimed to examine differences in site type in regard of subsistence strategies and economic behaviour. Previous investigations had delivered evidence that one of the sites represented long-term (Pod zubem) occupation and the other short-term (Pod křížlem) occupation. A total of 70 artefacts were analysed after they had been freshly removed from both sites during excavation. The random samples were minimally handled and left unwashed. The results revealed that 45 (64.3%) had some form of functional evidence (Hardy and Svoboda, 2009:165). Residues identified were comprised of plant, wood (particularly conifers/gymnosperms), feathers, hair, resin and starch grains. It was demonstrated that stone tools were used on a wide range of materials at the long-term occupation site of Pod zubem, while short-term occupied Pod křížlem yielded foremost evidence for plant processing and handheld tool-use. Animal processing and evidence for frequent hafting were characteristic features at the long-term occupation site. Furthermore, hazelnut residues were found at both sites.
2) The use of Australian backed artefacts was investigated by employing an integrated residue and use wear analysis (Robertson et al., 2009). The authors reviewed previous concepts on the use of these tool types such as with armatures on projectiles or barbs on composite spears. The study showed that backed artefacts were used for multiple tasks on diverse materials. Evidence of use was found on 33 used backed artefacts from the Deep Creek shelter (out of 41 artefacts), 60 from Emu Tracks (out of 65 artefacts) and 66 from Mussel (out of 93 artefacts) (Robertson et al., 2009:298—299). Overall, 63 per cent of used artefacts gave evidence of both task association and function. Residues found include bone, blood, collagen and muscle fibres, lipids and feathers, resin, plant cells, fibres, tissue with bordered pits, cellulose, ochre, mica and vivianite. Differential frequencies of task association found on the tools revealed inter-site variability. Considerable numbers of stone tools had more than one function.

3) In order to better understand the function of Aurignacian period stone tools, a residue and use wear analyses of 109 stone tool from three sites in south-western Germany was undertaken: Hohle Fels, Vogelherd and Geißenklösterle (Hardy et al., 2008:649). The sample included three handling categories: freshly excavated artefacts, water screened (water immersed) artefacts and lightly washed artefacts from a previous excavation (Geißenklösterle). The artefacts encompassed 39 different tool types and examination revealed that 64 (58.7%) had identifiable residues (Hardy et al., 2008:652). Overall, residues observed include hair, feathers, bone, antler, plant tissue and fibres, starch grains, wood, phytoliths, pollen and resin. All tool classes revealed evidence of use on multiple materials indicating both animal and plant processing. The widest range of use was found on unmodified flakes. Hafting evidence was only possible to be allocated to a maximum of six stone tools. Compared to the otherwise exceptional residue preservation, hafting
absence could suggest primarily handheld tool use (Hardy et al., 2008:658). Residues were found preserved on all three handling groups, including the washed specimens.

2.5. Previous radiocarbon dating of residues from stone tools

Previous research concerned with dating residues from lithic artefacts are limited to attempts on dating blood residue (Loy, 1987). A requirement for dating blood residue was that enough mass needed to be present on the artefact; in general 1 mg of carbon (Loy, 1987:62). The first successful dating of blood residue was conducted on two stone artefacts from northern British Columbia, Canada. These artefacts contained significant amount of blood residues and obtained results were in agreement with radiocarbon dated reference material (Nelson et al., 1986). The first stone tool contained 3 mg of carbon and was found in the same pit with a hearth. Charcoal from the hearth was dated 1060 ± 160 years BP, while the blood residue yielded an age of 1010 ± 90 years BP. The second artefact, a bifacial retouched chert knife, had only 50 μg of carbon and was found along with 23 other stone tools as part of a cache. While there was no possibility to obtain a radiocarbon date from the find context, the obtained residue date of 2180 ± 160 years BP was found to be in agreement with those for other artefacts of this tool type in the Yukon territory region. In subsequent studies, Loy (1987, 1993:48) pointed out the difference between the initial sample size and the purified carbon mass sample. For example, he estimated that 10 μm thick blood residues spread over more than 1 cm² would contain 0.5 mg of carbon, while larger residues such as animal hairs, bird feathers or plant remains should contain higher carbon amounts (Loy, 1987:62). Furthermore, Loy (1993:48) subsequently suggested that the original sample size for blood would be around ten times
larger than the yielded carbon mass for an AMS date, e.g. 500 μg sample to obtain 50 μg carbon.

2.6. Previous contaminant mitigation strategies

Keeley (1980:10–11) lists a series of cleaning steps for stone tools and suggested adjusting the degree of cleaning individual artefacts. His cleaning methods included the use of white spirit or methylated spirits to remove finger grease followed by washes or soaking in hot water and detergent. For the latter he considered grit-free ammonia-based household cleaners as useful. To remove lime and other mineral deposits an immersion in warm HCl (10% solution) was suggested, while extraneous organic deposits can be removed with NaOH (20% to 30% solution). Keeley cautioned that leaving tools longer than 20 to 30 minutes in the cooling solution can cause a light, white patination on the stone tool surface. Ultrasonic cleaning with detergent followed by clear water was recommended to remove sand, silt, dust and clay particles, while iron oxide can be removed by prolonged immersion in hot HCl. It is important to understand that Keeley's cleaning suggestions aimed to improve visibility on use wear traces on stone tools. In order to retain residues, but free them from contaminants, adjusted strategies are required.

To remove post-depositional contamination from blood residues, Loy (1987:62) reported that the sample was made soluble by using CHAPS, a non-ionic, non-denaturing detergent known to isolate membrane-bound proteins. After various filter actions and rinses with de-ionized water, to remove contaminants and CHAPS, sample age measurements were performed. The extracted blood residue was further treated by using a quartz tube
containing 1 g wire from CuO$_2$, used to evaporate the pipette solution, followed by heating to ~ 900°C for combustion.

The author also reported about radiocarbon dating attempts that resulted in a modern age, likely due to contamination (Loy, 1987, 1993). Loy (Loy, 1987, 1993) addressed contamination on AMS dates and raised awareness about their impact when using minuscule sized residue samples. For instance, he pointed out that many methods able to separate contaminants from the sample contain carbon-based chemicals (Loy, 1993:48), whose application could impact residue radiocarbon dates. Probably because of the unsolved problem of contamination on residue samples, to date,¹ no further endeavors have been conducted other than blood microgram sized carbon samples from stone tools residues, or to address contamination issues in various sampling steps of AMS dating.

### 2.7. Identified aspects relevant for research directions for this study

From the above review we can infer the following aspects relevant to AMS dating:

1. Use-related residues on lithics preserve in some assemblages in high proportions in various regions and within different ancient time periods.

---

¹ ‘To date’ refers here to the beginning of this study and includes the time in which the first publication (Yates et al., 2013 online, 2014 print), incorporated in this thesis, was published. After this time, a publication by Lentfer et al. (2013) published a study in which nut residues were AMS radiocarbon dated. This study (cited in Chapter 5 of this thesis) however, did not discuss or indicate the amount of mass used for dating (e.g. micrograms of carbon mass) and therefore offers only limited comparison to the results presented in the present study. A lack of sample size indication is also true for a study by Zarillo et al. (2008) on dating maize (this research cited and further discussed in Chapter 5).
2. Residues were found preserved in sheltered cave sites and to a lesser degree in open sites. Less evidence for the latter might be related to the fewer assemblages that have been analyzed.

3. Experimental studies also show exceptional residue preservations in open sites.

4. Animal and plant residues were found to survive tens of thousands of years worldwide.

5. Use-related residues can be discerned from non-use-related residues and contaminants.

6. Some information on contamination transfer into residue samples exists; however, further systematic analyses is required to assess contamination impact on small sample size radiocarbon dates.

2.8. Identified knowledge gaps relevant for research directions for this study

Several key knowledge gaps in the field of lithic residue dating were identified from the literature research. These are related to poor understanding or shortcomings in the following areas:

1. Contaminant transfer during various sampling steps for residue AMS dating.

2. Clear contaminant mitigation and avoidance measures required for radiocarbon dating in extremely small sized residues.

3. Contaminant type identification – microscopically and biochemically.

4. No record indicated the dating of residues treating microgram carbon samples other than blood.
5. Suitability of various residue types for radiocarbon dating (estimations about amounts of preserved residues).

6. Validation of obtained residue dates in correlation with sound reference chronology.

7. Connection between residue identification, preservation, contamination and AMS radiocarbon dating.

8. Identification of suitable sampling strategies in the field, in storage and in the laboratory.

9. Sample size needed nowadays to achieve an AMS date.

10. Carbon content of various residues types.
AMS dating of ancient plant residues from experimental stone tools: a pilot study

Unformatted published paper is shown in Appendix 3

Residue analyses on stone artefacts have contributed to resolving functional questions in stone tool research. Although identifying the function of tools through the analysis of their micro-residues is possible, the establishment of a sound numerical chronology for stone tools lacking a clear stratigraphic sequence, such as surface scatters, remains a challenge. While radiocarbon dating of blood residue on stone artefacts has been published previously (Loy 1987, 1990, 1993; Loy et al., 1990; Nelson et al., 1986), this paper reports on an experiment designed to assess the possibility of directly dating residues on stone artefacts by accelerator mass spectrometry (AMS) based radiocarbon measurements. Innovative with this approach is (1) the use of mid and late Holocene pre-dated plant material (wood and peat), processed with contemporarily manufactured stone flakes under controlled laboratory conditions and (2) the use of very small carbon masses (less than 22 mg) for radiocarbon dating. The 14C results of the wood residues are in excellent agreement with the original sample, whereas the 14C results of the peat residues yield a wider age variation as expected due to the inhomogeneity of the material, but nevertheless, provided dates within an expected age range. Preliminary results demonstrate the feasibility of dating very small amounts of plant residue on lithics directly when contaminants are confined.

Keywords: Residue dating, Residue analysis, AMS, 14C, Radiocarbon dating, Micro-remains, Contaminants, Preservation, Conservation, Experimental archaeology, Stone tools
3.1. Introduction

This study examines the feasibility of dating plant residues on stone artefacts by AMS radiocarbon dating. The majority of stone tools, both in Australia and abroad, are found in open sites and are referred to as ‘surface scatters’. For most of these artefacts, it is difficult to achieve a sound chronology in the absence of datable organic material or cultural markers (e.g. characteristic typology). Even in the best scenario, where additional parameters can be identified, only a relative dating with a broader time frame can be deciphered. Recent results of organic residues analyses on stone tools from open sites (Barton, 2009; Cooper and Nugent, 2009; Langejans, 2010) have identified their potential for direct dating of artefacts.

Previous research focusing on dating stone tools residues was limited to dating blood residue (Nelson et al., 1986; Loy, 1987, 1993). The first attempt was conducted on two stone artefacts containing sufficient amounts of blood residue. Results were consistent with their radiocarbon dated stratigraphic position (Nelson et al., 1986). Some analysts experimentally tested the practicability of detecting blood components, such as the proteins haemoglobin, immunoglobulin G and albumin on artefacts and found it difficult, but possible (e.g. Cattaneo et al., 1993; Gurfinkel and Franklin, 1988). Others doubt the plausibility of preservation of protein and haemoglobin after several hundred years’ burial, and in particular, the possibility to differentiate between species based on blood residue (Smith and Wilson, 1992). The survival of proteinaceous residue, however, appears to be related to burial conditions. Clay rich soils, along with other conservative conditions such as an alkaline pH and cation exchange capacity have shown to be beneficial for their survival (Jones, 2009; Loy, 1987: 58; Gurfinkel and Franklin, 1988). Further dating of
blood residue on stone tools has established the perturbing effect of contaminants on the AMS dates and possibilities for their chemical removal were discussed (Loy, 1987, 1993). An additional limitation for blood residue dating was the actual quantity required: between 50 μg and 1 mg carbon was required to achieve high enough precision (Loy, 1987: 62, 1993: 46; Vogel et al., 1989: 608).

We are aware of no further attempts to either date residues other than blood or to address the problem of contamination of non-use-related residues on the samples extracted for dating. In this experimental study, we investigate the potential of directly dating plant residues from stone tools.

Potential sources of contamination resulting from artefact environmental exposure, retrieval, handling and storage were addressed by using experimentally produced stone tools and predated plant materials. In a real-artefact context factors such as fungal activity (e.g. Barton, 2009: 134), the precipitation of calcite on the artefact (or hard water/freshwater reservoir effect) (Fischer and Heinemeier, 2003; Long et al., 1992), insect remains (e.g. Cooper and Nugent, 2009: 217), post-depositional related soil components (Wadley and Lombard, 2007: 1003; Langejans, 2011) all must be addressed. Care must also be taken with sample preparation (e.g. Barton and Matthews, 2006; Fullagar, 2006b: 191). By excluding these factors we have focussed on testing the possible influence of introduced modern carbon during sample preparation, the impact on dated results, and consequently, the significance of applying the method on dating residues.
3.1.1. Identification and preservation of residue on stone artefacts

Although use-related residues on stone artefacts have been reportedly preserved for up to 2 million years in Africa (Loy, 1998; Jones, 2009; Dominguez-Rodrigo et al., 2001; Lombard and Wadley, 2009) and tens of thousands of years abroad (Hardy et al., 1997, 2008; Hardy and Svoboda, 2009; Loy and Hardy, 1992; Pawlik, 2004) in this study we focus on the 40,000 year dating time frame of AMS radiocarbon dating.

In this context, in mainland Australia, residues have been found preserved on stone tools dated between 30,000 and 37,000 years BP (Dodson et al., 1993; Fullagar and David, 1997), on backed artefacts, dating from 1500 to 8500 years BP (e.g. Attenbrow et al., 2009; Robertson et al., 2009) and in Tasmania 6000 years BP (Fullagar and Jones, 2004). In the Pacific region, 28,000-year-old starch residues were found on stone artefacts (Loy et al., 1992).

While not all residues found on stone artefacts are use-related, residue and usewear analyses aim to identify and interpret ancient residues. Pioneering research on, use wear and residue analyses was conducted by Semenov (1964), Kamminga (1977, 1979, 1982), Fullagar (1986) and Loy (1987, 1993). Thomas Loy also started integrating interdisciplinary methods and initiated several new directions in residue analyses e with detection of ancient blood residue on stone artefact surfaces and attempted species and DNA identifications (Loy, 1983; Loy and Remington, 1994). However, the reliability of species identification through the analysis of blood residues has been debated (Fiedel, 1996; Smith and Wilson, 1992) and in the case of immunological techniques, such as protein radioimmunoassay on ancient samples, is still subject to misidentification caused by taphonomic and diagenetic alterations of proteins (Potter et al., 2010).
Haslam (2009) reviewed published microscopic residue analyses over the 30-year span from 1976 to 2006. He pointed out the differences in sample sizes examined within assemblages and the lack of transparency indicating the ratio of artefacts with no residue to artefacts with residue. The need to refine sampling protocols for residue analysis was identified. Relevant to this are the recent finds of residue analysed stone artefact assemblages from archaeological contexts which show that more than half of the examined inventories contain residues (Hardy et al., 2008: 652; Hardy and Svoboda, 2009: 165; Robertson, 2009: 300, 302).

Research over the last decade has focussed on the occurrence of residues on particular implements, such as backed artefacts (Attenbrow et al., 2009; Fullagar et al., 2009; Hardy et al., 2008; Robertson et al., 2009; Robertson, 2011), bondi points (Robertson, 2011), pieces esquillée (Langejans, 2012a) or the detection of hafting locations on lithic points (Lombard and Wadley, 2009; Parr, 2006). Further analyses concentrated on preservation (Barton, 2009; Cooper and Nugent, 2009; Field et al., 2009; Fullagar et al., 2009; Hardy and Svoboda, 2009; Jones, 2009; Langejans, 2010), contamination of residues (Barton, 2009: 134; Cooper and Nugent, 2009: 209, 217; Haslam, 2004, 2006: 1717; Kononenko, 2008: 33; Langejans, 2011; Wadley and Lombard, 2007: 1003) and starch residue research (Barton, 2009; Haslam, 2004; Lentfer, 2009; Torrence and Barton, 2006; Torrence, 2006).

While many earlier experimental studies researched blood residues (e.g. Cattaneo et al., 1993; Gurfinkel and Franklin, 1988; Hyland et al., 1990; Shanks et al., 2004), recent experimental residue analyses have increasingly focused on preservation and contaminant issues (Barnard et al., 2007; Barton, 2009; Haslam, 2004; Jones, 2009; Langejans, 2010, 2011; Loy and Barton, 2006; Wadley and Lombard, 2007).
In the context of residue preservation previous studies have illustrated that use-related residues, such as plant residues, can be very resistant to extraction and even withstand several washes (Barton et al., 1998: 1233; Fullagar, 1986, 1993; Fullagar et al., 1996; Shanks et al., 2004). The observations of some analysts indicate that residues can build a shield (Barton, 2009; Loy, 1987: 58, 1990: 650) once preserved. This protective barrier is considered to be hydrophobic and defiant to microbial attacks (Barton, 2007; Barton and Matthews, 2006; Loy, 1990).

The studies discussed above outlining the recent improvement in residue identification coupled with recent advances in AMS dating which have reduced the carbon size limit down to a 5 μg of carbon (e.g. Smith et al., 2007, 2010; Yang et al., in press) has inspired the objective of the current study to further examine residue dating.

The aim of this study is to test the feasibility of dating residues on stone tools, having excluded post-depositional related contaminants and e for the first time e to systematically investigate the influence of possible introduced contaminants during sample preparation for AMS radiocarbon dating. It was decided to use experimentally produced stone flakes, so avoiding the physical alteration of ancient artefacts by residue extraction and the possibility of extracting any ancient remaining residues on the tools. This also permitted duplication of the experiments (Loy, 1993: 46; Fullagar et al., 1996: 741).
3.2. Materials and methods

3.2.1. Materials and procedure

The experimental design had two parts. The aim of the first part of the study was to gain experience with residue extraction procedures and to quantify the amount of residue collected. In the initial trial, stone flakes were produced from flint cores using an antler as a percussion stick and a leather leg/lap protection. Organic residue was applied to stone flakes in the form of fresh plant materials of wood, and fern (Fig. 3.1.). The flakes with the applied residue were dried in a closed clean room for 2 weeks before being stored in re-sealable plastic bags. Residues were then extracted to estimate the amount of organic material that can be retrieved from a single flake. Extraction was conducted in two ways: by physically removing residue with a scalpel whilst monitoring it under microscope, and by using a sonic bath (Fullagar, 2006a: 213) (Fig. 3.1).

We elected to use a jewellery sonicator (LEO Ultrasonic Cleaner, LEO-50), rather than an industrial sonicator, because the sonication would be gentler and less likely to dislodge parts of the stone itself. Following this preliminary procedure we were prepared for the second part of the experiment, involving the application and extraction of ancient residue under controlled laboratory conditions on stone flakes produced in the laboratory.
Organic material, comprising peat and wood, was chosen from previous environmental studies of the North East Coast of New South Wales and Southeast Queensland (Boyd et al., 1997; Peters, 1990). Samples from various cores from these studies were previously radiocarbon dated by ANTARES AMS facility at ANSTO. One wood sample and three peat samples, originating from the same survey, but not radiocarbon dated yet, were chosen for this pilot study. The samples retrieved from various depths in three different sediment cores, were utilized. Peat was chosen as a possible and accessible plant material on which plant processing with stone tools could be simulated.

Five chert stone flakes, measuring between 19 mm and 49 mm in length were produced in the laboratory on a prepared plastic surface whilst wearing starch-free gloves and using a copper headed percussion stick (Fig. 3.2). Prior to conducting the experiment, the tools were sonicated for 2 min in order to remove all possible sources of contamination that may have resulted from previous handling. The tools were then used to cut through and scrape the wood and peat material. Starch-free gloves were worn at all times and exchanged for fresh gloves each time a new stone flake or a different material was handled. All tool surfaces were examined by light microscopy to monitor residue distribution in preparation for subsequent extraction.

**Figure 3.2.** Production of stone flakes using metal headed percussion stick, plastic-coated production surface and wearing starch-free gloves, cutting into wood, sample pre-treatment with the acidic-alkali-acid method.
Extraction was conducted immediately after working the stone flake. A drying period was not considered necessary because the organic material was already in a desiccated state and long exposure of the artefacts, even under controlled conditions in the laboratory, might bring them in contact with contaminants, e.g. ubiquitous starch (Loy and Barton, 2006). Each stone tool was used twice to process the same plant sample. Adhering residues were extracted by both ultra-sonication for 120 s and by scraping the residue with the scalpel after having located its distribution under microscopy. The aim of using this double extraction approach applied respectively on the same stone flake was to identify a more reliable method for the purpose of residue yield and for dating the residue. During sonication the stone flakes were placed into plastic floating boats filled with Milli-Q water. The original wood piece measured 73 mm in length, 41 mm wide and 19 mm deep. Cutting and scraping activities on the wood piece with stone tools were performed on the same location, on the outer margin of the wood piece (Fig. 3.2) to enable highest possible comparability of the data. Peat samples consisted of dried plant material in which cutting movements with stone flakes were carried out. The sequence of the experiment (part two) is illustrated in Fig. 3.3.
3.2.2. Preparation for AMS radiocarbon dating

Table 3.1 summarises the samples prepared for this experiment at ANSTO. From the original wood sample, duplicate residue samples were taken by scraping under a microscope and by sonication for 120 s in Milli-Q water, whereas single residue samples were taken by the two extraction methods from each of the three original peat samples. The five dry samples resulting from scraping of residue were transferred into centrifuge tube using a Milli-Q water rinse, while the five sonicated samples, already suspended in Milli-Q water, were placed in centrifuge tubes including their plastic float boats in order to capture as much residue as possible. All samples were chemically pre-treated with the acidic-alkali-acid method to remove carbonaceous contaminants such as carbonates and soluble organics (fulvic acids and humic acids). Any plant rootlets that were present in the
peat samples were removed and then samples were ground with a mortar and pestle. In order to remove carbonate contaminants the samples were treated with 2 M HCl at 60 °C for 2 h. Alkali washes were then performed to remove soluble organic contamination (fulvic and humic acids). Washes of increasing molarity between 0.1 and 2.5M NaOH were performed for 2 h at 60 °C until all contamination was removed (i.e. the solution was clear at the end of the 2 h wash). The samples were then treated with 2M HCl at room temperature for 2 h to remove any carbonates that precipitated from modern atmospheric CO2 during the alkali washes. Samples were then rinsed with distilled water and dried in a 60 °C oven (Fig. 3.2). The pre-treated samples were then converted to CO2 by combustion using the sealed-tube technique (Vandeputte et al., 1996) and graphitised as outlined in Hua et al. (2001). Since its inception, AMS has permitted radiocarbon measurements to be made on samples containing small amounts of carbon, and continual refinement of the technique currently enables researchers to work with samples containing just a few micrograms of carbon. But with decreasing sample size, researchers are increasingly at risk of extraneous carbon, which may be ‘modern’ or ‘dead’ or of intermediate age, being introduced to the sample carbon and so corrupting an age determination. The ANTARES AMS facility at ANSTO (Fink et al., 2004) Australia, routinely measures the 14C/12C ratio for samples containing as little as 5 μg of carbon (e.g. Smith et al., 2007, 2010; Yang et al., 2013).
Table 3.1. Radiocarbon ages from original wood and peat samples and associated residue samples extracted by sonication and scraping, amount of carbon mass used for AMS dating, time range of calibrated radiocarbon ages (using CALIB 6.0. after Stuiver et al., 2012).

<table>
<thead>
<tr>
<th>Sample ID</th>
<th>ANSTO Sample ID</th>
<th>Material Kind of extraction</th>
<th>Carbon Mass [μg]</th>
<th>Rad Age ±</th>
<th>Calibrated Rad Age Range BP cal</th>
</tr>
</thead>
<tbody>
<tr>
<td>NA5 Wood 95 – 39-1</td>
<td>OZ 0652</td>
<td>Wood Original</td>
<td>2060</td>
<td>6415</td>
<td>35 7275 7420</td>
</tr>
<tr>
<td>NA5 Wood 95 – 39-1</td>
<td>OZ 0650</td>
<td>Wood Sonication</td>
<td>170</td>
<td>6520</td>
<td>60 7312 7519</td>
</tr>
<tr>
<td>NA5 Wood 95 – 39-1</td>
<td>OZ 0651</td>
<td>Wood Scraping 1</td>
<td>19.76</td>
<td>6410</td>
<td>210 6842 7670</td>
</tr>
<tr>
<td>NA5 Wood 95 – 39-1</td>
<td>OZ 0648</td>
<td>Wood Scraping 2</td>
<td>10.50</td>
<td>6590</td>
<td>290 6844 8000</td>
</tr>
<tr>
<td>NA5 Wood 95 – 39-1</td>
<td>OZ 0649</td>
<td>Wood Sonication</td>
<td>2.54</td>
<td>Sample did not proceed to AMS</td>
<td></td>
</tr>
<tr>
<td>NA2 Peat 1 ES 92 - 59-60</td>
<td>OZ 0640</td>
<td>Peat Original</td>
<td>1920</td>
<td>5845</td>
<td>35 6557 6743</td>
</tr>
<tr>
<td>NA2 Peat 1 ES 92 - 59-60</td>
<td>OZ 0642</td>
<td>Peat Scraping 1</td>
<td>21.10</td>
<td>5060</td>
<td>200 5449 6278</td>
</tr>
<tr>
<td>NA2 Peat 1 ES 92 - 59-60</td>
<td>OZ 0643</td>
<td>Peat Sonication</td>
<td>2.68</td>
<td>Sample did not proceed to AMS</td>
<td></td>
</tr>
<tr>
<td>NA3 Peat 1 ES 92 - 66-67</td>
<td>OZ 0653</td>
<td>Peat Original</td>
<td>950</td>
<td>5155</td>
<td>35 5885 5991</td>
</tr>
<tr>
<td>NA3 Peat 1 ES 92 - 66-67</td>
<td>OZ 0644</td>
<td>Peat Scraping 1</td>
<td>16.31</td>
<td>6500</td>
<td>240 6851 7837</td>
</tr>
<tr>
<td>NA3 Peat 1 ES 92 - 66-67</td>
<td>OZ 0645</td>
<td>Peat Sonication</td>
<td>7.66</td>
<td>Sample delivered modern age</td>
<td></td>
</tr>
<tr>
<td>NA4 Peat E2 92-90-91</td>
<td>OZ 0641</td>
<td>Peat Original</td>
<td>730</td>
<td>1155</td>
<td>30 979 1171</td>
</tr>
<tr>
<td>NA4 Peat E2 92-90-91</td>
<td>OZ 0647</td>
<td>Peat Scraping 1</td>
<td>11.91</td>
<td>1810</td>
<td>230 1299 2208</td>
</tr>
</tbody>
</table>

3.3. Results

In the first part of the experiment in which modern flakes were used on modern residue, it was found that using a jewellery sonicator for only 60 s did not extract sufficient residue, whereas after 120 s there was macroscopically visible residue. Accordingly, we used a sonication time of 120 s for the second part of the experiment. Despite the preliminary investigation with modern residue, we found that sonication yielded just 2–3 μg of carbon for one of the wood samples and for two of the peat samples. We elected not to proceed to
AMS with such small samples yields. Furthermore, the remaining peat sonication sample (7.66 μg of C) yielded a modern age.

### 3.3.1. Wood samples

The original wood sample OZ 0652/NA 5 95 39-1 delivered a radiocarbon age of 6415 ± 35 years (Table 3.1).

One residue sample extracted by sonication provided a radiocarbon age of 6520 ± 60 years, whereas another extracted in a sonic bath wooden residue sample did not yield enough carbon mass (2.54 μg) to proceed to AMS dating. The two residues extracted by scraping have radiocarbon ages of 6410 ± 210 years for sample OZ 0651 and 6590 ± 290 years (OZ 0648) (Table 3.1). These dates all fell within a narrow range of each other, demonstrating that the extracted residues reflect the age of the original sample (Fig. 3.4).

### 3.3.2. Peat samples

The original peat samples derive from different sediment cores. Sample OZ 0640/Peat NA2 ES 92 retrieved from a depth of 59–60 cm delivered a radiocarbon age of 5845 ± 35 years (Table 3.1). Residue of this sample extracted by scraping yielded 5060 ± 200 years.
while the residue extracted by sonication did not deliver enough dateable carbon mass (2.68 μg). The age gap between original sample and extracted residue is larger than is the case for the wooden samples discussed above (Fig. 3.5).

![Radiocarbon Ages derived for NA2 Peat and extractions](image)

**Figure 3.5.** Radiocarbon age of NA2 original peat and corresponding date of extracted residue.

Similarly there is a gap in age between the original sample OZ 0653/peat NA 3 ES 92 from 66 to 67 cm depth with a radiocarbon age of 5155 years ± 35 years and the residue extracted by scraping with a radiocarbon age date of 6500 ± 240 years. With around 1000 year’s difference, the extracted residue does not reflect the date of the original sample (Fig. 3.6). The equivalent residue extracted by sonication delivered a modern age with carbon mass 7.66 μg carbon mass.

![Radiocarbon Ages derived for NA3 Peat and extractions](image)

**Figure 3.6.** Radiocarbon age of NA3 original peat and corresponding date of extracted residue.

Sample OZ 0641/peat NA4 E2 from 90 to 91 cm depth is the third original peat sample and resulted in a radiocarbon age of 1155 ± 30 years (Table 3.1). The corresponding sonicated residue again gave a low carbon mass of 1.90 mg which was insufficient for
dating, whereas the scraped residue yielded a radiocarbon age of 1810 ± 250 years. This date for the extracted residue also deviates from the original sample (Fig. 3.7).

![Figure 3.7. Radiocarbon age of NA4 original peat and corresponding date of extracted residue.](image)

From the results illustrated in Figs. 3.4–3.7 we find that all extracted samples have a larger error than the original (because they all contain less carbon and so did not produce the same counting statistics) and that with the exception of the NA 2 peat sample they are providing dates older than the original sample. Only the extracted NA 5 wood sample dates overlap the expected value.

### 3.4. Discussion

We have conducted an experiment under laboratory conditions to assess the recovery and reliable radiocarbon AMS determination of residues extracted from stone (chert) tools. One wooden sample of radiocarbon age 6415 ± 35 and three peat samples of 5845 ± 35, 5060 ± 200 and 1155 ± 30 radiocarbon age were used for these tests. The age span of the carbon in these samples was previously unknown. Two methods of extraction were trialled: scraping with a scalpel under a microscope and sonication in Milli-Q water. In most cases, sonication did not produce adequate yield of the plant residues for measurement.
We have shown that wooden residues gave radiocarbon ages with errors that spanned concurrent measurements on the original wood sample age, although two of them shifted to older ages. The radiocarbon ages and errors derived from the residues extracted from the three peat samples are at variance with the radiocarbon ages of the original material. In particular, the only sonicated sample that proceeded to AMS yielded a modern age, rather than the expected 5155 ± 35 radiocarbon years. Two of the scraping samples were significantly older than the originals, whereas the third was significantly younger than the original. Such variable results could be explained by greater sample (age) inhomogeneity for the peat over the wood, although it is difficult to see how the observed disparity of greater than a 1000 radiocarbon years could be supported by this explanation. It is more likely that the smaller scraped samples were shifted towards an older age by the addition of older extraneous carbon, as appears to be the case for the wood. We have no explanation for the shifts towards modern for NA3/OZO645 and scraped NA2/OZO642.

The larger variability in the dates of peat-extracted residues might originate from inhomogeneity of the material itself. Inhomogeneity may result from root growth, and since the material is porous and penetrable, organic material (such as pollen) possibly travelled downwards during its genesis. Since the material consists of an accumulation of organic matter, decayed plant material from different sources may have been penetrated while slicing and cutting with stone flakes into the material. As the peat residue samples, which are shifting to an older age, have rather small carbon masses of 11.91 and 16.31 (Table 3.1), the possibility of introduced contamination is higher and might have contributed in the age variation. Samples this small are more susceptible to background contamination introduced during the chemistry preparation. In retrospect, the use of peat to simulate plant processing with stone tools, proved to be not an ideal choice for this
experiment, more homogenous material such as nuts and seeds in subsequent studies would eliminate some of these uncertainties.

It needs to be noted that the only extracted residues which did not proceed to AMS stemmed from 120 s of sonication, while all residues retrieved by scraping revealed sufficient residue that was able to be dated. Another residue sample extracted in sonic bath yielded a modern age, while the corresponding scraped residue dates 6500 ± 240 years BP. As a matter of fact, from the five sonicated residues, only one of the wooden samples provided an AMS date, whereas all scraped residues provided enough material to date (Table 3.1, Fig. 3.3).

These findings point to the following:

(1) More than 120 s of sonication is likely required to extract enough dateable residue. Sonication of residues from stone tools is attested in some studies as the most efficient extraction method (e.g. Shanks et al., 2001), especially when longer exposure in the sonic bath was carried out to achieve sufficient yield (Pearsall et al., 2004: 428).

(2) Scraping is the more efficient way to extract residue for dating purposes. While scraping appears to be the more efficient method for residue extraction to radiocarbon date the sample, this method is likely to be more invasive and may alter the stone surface. For example, scraping the artefact can cause surface markings, which could possibly be confused for use wear patterns (Fullagar, 2006b: 198). Hence scraping to extract residues should only be applied after thorough documentation of use wear traces on the artefact.

Other extraction methods (e.g. Fullagar, 2006a: 213; Loy and Fullagar, 2006: 197, 198) might be employed with consecutive experiments. The fact that chert was used as raw
material for the experimental flakes might prevent the residues from adhering to the rather smooth tool surface in the same way that they would remain on coarser grained raw materials, hence resulting in less extractable residue. Having said that, previous studies have demonstrated that residues do adhere on even smoother material, such as obsidian (e.g. Barton et al., 1998; Shanks et al., 2001; Weisler and Haslam, 2005). For this pilot study we decided to utilize only one kind of raw material to allow thorough investigation in one particular material. Using only one type of stone enables more reliable comparability of other parameters, such as extraction method. Whether other rock types yield higher amounts of residue in their coarser grain needs to be investigated in subsequent research. In addition it should be taken into account that larger stone tools may contain more residues, e.g. due to their longer working edge. Limitations of this study lie in a small dataset and consequently limited possibilities to combine singular parameters such as extraction method, residue yield and raw material type.

Since the results demonstrate the feasibility of the method when post-depositional contaminants are confined, it seems plausible to review the applicability of the method on artefacts from archaeological contexts. For the application of the method on archaeologically retrieved tools, the challenge consists of finding well-dated artefacts with sufficient organic residues preserved and on which contaminants are discernible. Ideally, where stone tools were retrieved from a context where they were incorporated in a site with well-dated stratigraphy, and are excavated, handled and stored with consideration of contaminant exclusion. Fullagar (2006b: 189) suggests a general limiting in handling, in situ removal of artefacts with adhering soil (rather than from sieves) by wearing starch-free gloves or using plastic instruments and storage in plastic bags. Obviously, a limited amount of suitable material is expected, nevertheless, several studies have demonstrated
the feasibility to discriminate contaminant residues from use-related residues (e.g. Barton et al., 1998; Cooper and Nugent, 2009: 209, 217; Fullagar, 2006a; Langejans, 2010, 2011; Wadley and Lombard, 2007: 1003). Most non-use-related residues are considerably smaller and have a more irregular distribution on the tool surface, whereas use-related residues often have distribution patterns and are trapped in micro cracks or crevices (e.g. Barton and Matthews, 2006; Fullagar, 2006a; Kononenko, 2008: 33; Langejans, 2011). By taking control soil samples, it was concluded that if residues found on artefacts are not present in the soil, then the residue are likely to have resulted from use of the artefact (Haslam, 2004: 1717). With regards to residue preservation, examining tool-adhering sediment proved valuable in retaining use deposited residue (Barton et al., 1998). These circumstances give added rationale for further study on the dating of stone artefact residues.

High preservation rates of residues in some assemblages and advanced differentiation ability of use-related and non-use-related residues on tools provides a potential for further analyses. In particular, there is a potential to differentiate between contaminant residues from use wear residues on tool surfaces. Additionally the presence of residue protecting shields may allow the removal of modern contaminants, but this needs to be tested.

Such protective barriers might possess different qualities: First for individual residues; and secondly in the way the barrier has developed in various environments, e.g. dry climate, fast desiccation, open or sheltered site, degree of oxygen and water exposure and surrounding sediments. These conditions are important to consider as they may affect the amount of yield extractable from a stone tool as well as its quality in terms of conservation and penetrability.
Importantly, residue resistance to washes and to some forms of extraction, as well as their protective shields, most likely guards them from removal when associated contaminants are detached. Microwear analysts suggest a range of procedures to remove contaminants from a tool surface (Fullagar, 2006a: 216; Loy, 1987: 62, 1990, 1993). The elimination of non-use-related residues from artefacts significantly reduces contamination of carbon mass. Additionally, attempts to counter contaminant issues during the procedure of AMS dating and to refine the techniques are in progress (e.g. Liebl et al., 2010).

After the removal of non-use-related residues, a partial extraction of use-related residue can be conducted. Required, this step should only be carried out after a detailed microscopic examination and documentation of each individual artefact with regards to identifiable residues and signs of use wear. Subsequently, a partial residue extraction might then be carried out, allowing experimental repetition from the remaining residues on tools (Loy, 1993: 46; Fullagar et al., 1996: 741). As many use wear and residue analysed stone tools showed evidence of multiple uses, often several organic and inorganic residues are present on one tool. For the residue dating process dividing these residues might not be required because the age of the tool’s use life is the goal here, not its function as an implement. It might be of advantage having more than one organic residue on a tool as combined these may increases the carbon mass, and hence, the possibility of dating the residue. Some stone flakes may have never been used but may still contain residues resulting from the percussion act (Byrne et al., 2006) to produce the artefact. If an organic implement such as antler was used as pounder, the dated remaining residue would indicate the birth moment of the flake.
Once established on stratified dated artefacts, the method is applicable on stone tools from open sites, with little or no further dating possibility. Residue analyses of stone artefacts from some open sites show considerably high preservation rates compared with buried tools and preservation (e.g. Cooper and Nugent, 2009). A number of experimental studies demonstrated favouring preservation of some residue types on surface tools (Barton, 2009; Langejans, 2010). Accordingly, if contaminants could be discriminated and removed from surface tools, previously difficult to date open site artefacts might be able to be dated by their residues. Further, from several open sites of a region, a fine chronology can be established to a degree that enables us to reveal movement of settlement patterns. The method may also allow us to ascertain the time period when some open sites have been revisited. For stratified finds, it is a technique that confirms previously conducted or associated dates, and ideally, contains the ability to establish finer chronologies within sediment layers.

3.5. Conclusion

The preliminary results of this study point to the feasibility of dating plant residues on stone tools directly when contaminants are confined. While AMS radiocarbon dated wooden residues have delivered highly corresponding dates with the original sample, dated peat residues showed a wider deviation range, possibly due to the materials inhomogeneity. On the example of 14C results of original wood and corresponding extracted samples we demonstrated that the influence of introduced contaminants during sample preparation for AMS can be small. The potential of the technique presented in this study, once refined, provides not only the possibility to deliver additional dates to
excavated and already age-determined assemblages, it might also be used as an independent dating technique, especially for finds with limited dating possibilities (such as open sites).

As the results of this project illustrated the applicability of AMS dating on residues, the necessity of constructive research becomes apparent. As discussed, the recognition of contaminants by residue analysts has become standard practice. Although discernible by specialists, research into ways to remove non-use-related residues from artefacts, without compromising the integrity of use-related residues is required.

Further test series would be beneficial prior to conducting new tests on artefacts from archaeological contexts. Although, residues have been described to adhere to raw materials with smooth surfaces, such as obsidian, an investigation of materials such as silcrete, quartz and quartzite for residue dating appears to be the logical next step.

Testing additional extraction methods, such as brush, wash, pipette and peel extraction, would aid in comparing previous and new extraction methods. Because three of the experimental samples did not yield enough residues using sonication as extraction means, longer exposure of the artefact in the float boat in order to potentially yield more residue needs to be tested. As the examined peat sample dates vary from original sample, extra vigilance for contact with contaminants during the extraction process is indicated. Sonicated wood sample, however, delivered a suitable date with the corresponding original sample, which could indicate that peat sample might be unsuitable. Other plant material, such as nuts or seeds, could potentially offer suitable material for residue dating.
3.6. Acknowledgements

AINSE grant (award no ALNGRA11032) enabled us to date 10 residue samples extracted from stone tools and the dating of associated original organic plant samples. We owe thanks to Professor Bill Boyd, Southern Cross University, who kindly provided wood and peat samples from an environmental study for this experiment. Many thanks goes to Dr. Jürgen Junkmanns, Erftstadt-Bliesheim, Germany, www.Pfeil-Bogen.de for assisting in the first part of the experiment by producing stone flakes and working them on organic material. Thanks to Fiona Bertuch, ANSTO, who supervised sample preparation in ANSTO laboratories.
CHAPTER 4

Radiocarbon-dating adhesive and wooden residues from stone tools by Accelerator Mass Spectrometry (AMS): challenges and insights encountered in a case study

Unformatted published paper is shown in Appendix 4

ABSTRACT

In this study we present and assess a process to enhance archaeological residue AMS dating by focusing on contaminant confinement. The sequence of methods applied consists of: 1) optical residue and use wear analyses, 2) experimental designs addressing cleaning treatments to mitigate impact of contaminants, 3) preparation and extraction of residues from (mostly) previously dated stone artefacts, and 4) establishing the elemental characteristics of residues by using SEM/EDX as a final step to avoid sample contamination during analyses. We found the alkaline surfactant Decon 90 is a useful solution for removal of skin scales and fabric fibre but has limited effect on graphite contamination introduced by pencil lead. Adhesive residues were not affected by Decon immersion, however, wooden residues from bog sites were partly dislodged. While the methodological sequence was in general successful and some artefact residues were dated within the anticipated age range, difficulties were encountered with other lithic residues. Some artefact residues attained AMS dates which appear to be affected by modern contaminants and other residue radiocarbon dates were seemingly affected by fossil shell derived from flint stone, plasticisers or from a fixative substance older than the fabrication and use of the artefact. One outcome from this study is that performing chemical residue identification earlier in the method sequence using non-destructive and non-contaminating methods would guide the choice of residue treatment and improve reliability of age determination.

Keywords: Residue dating, Residue analysis, AMS, 14C, Radiocarbon dating, Resin, Birch bark tar, Bitumen, Adhesives, SEM-EDX, Plasticisers, Mesolithic Friesack
4.1. Introduction

Direct dating of artefact residues has the potential to provide archaeologists with new chronological information. Accelerator Mass Spectrometry (AMS) radiocarbon measurements can be made on samples containing as little as 5 µg or less of carbon (‘µgC’) (Smith et al., 2007, 2010a; 2010b; Yang et al., 2013) and in principle this allows very small amounts of residues from stone tools to be radiocarbon dated. In a pilot study, we demonstrated the feasibility of direct residue dating under laboratory conditions with only 10.5 µgC obtained from wooden residues (Yates et al., 2014). One of the key limitations, however, was the impact of contamination due to the extremely low mass used for the dating. Fungus and soil components can be easily transferred into archaeological residues during handling and storage (e.g. Barton, 2009: 134; Wadley and Lombard, 2007: 1003; Langejans, 2011). This highlights the need to develop preparation and removal protocols for accurate AMS dating.

In our pilot study, we also suggested that a next step after experimental residue dating should be to date well-preserved residues from artefacts that are stratified and well dated, so as to verify the methodology. For this study, we obtained seven stone tools with associated age determinations from three archaeological assemblages. Five artefacts from Friesack, Germany were selected for two reasons: first, a use-wear residue analyses conducted on 306 stone tools revealed frequent preservation of hafting residues (adhesives) along with plant remains from wooden shafts (Pawlik, 2011a); and second, Gas Chromatography Mass Spectroscopy (GC/MS) analyses on four samples (three pieces
of tarry black material, two of which had chewing marks, and one sample of tarry dark material adhering to a bone point) revealed birch bark tar was used at the site (Baumer and Dietemann, 2008). This provided an indication of the likelihood of finding this hafting fixative on lithic tools. Two stone tools, one from Wesseling, Germany, and one from Yelgun, Australia, were chosen because they exhibited macroscopically visible dark residues resembling adhesive residues. This case study aimed to test the feasibility of radiocarbon dating adhesive and wooden residues from archaeological stone tools and at the same time to reduce contamination by finding adequate removal techniques.

Adhesive residues have the advantage of having a tough texture, and wooden residues have been dated successfully in the above mentioned study. Both residues have been found preserved under various conditions. However, adhesive residues are commonly preserved in higher quantities than other residues, allowing replicate radiocarbon dating. This fact is significant because different kinds of adhesive residues are preserved worldwide and the possibility of directly dating minute amounts would provide archaeologists with a new way of determining age. By choosing adhesive residues for radiocarbon dating, we aimed to establish protocols which then can be transferred and adjusted for other scarcer residues.

Archaeological adhesives function to a large extent as fixatives for lithics to hafts of wood, antler or bone. In Europe, birch bark tar (also known as birch pitch) is reported as a common hafting fixative. So far the oldest examples have been found at Middle Paleolithic sites (Mania and Toepfer, 1973; Hedges et al., 1998:229; Grünberg, 2002; Grünberg et. al., 1999; Koller et al., 2001; Mazza et al., 2006; Pawlik and Thissen, 2011), followed by Mesolithic sites (e.g. Aveling and Heron, 1998; Clark, 1954; Pawlik, 1997, 2004) and
from sites from the Neolithic period onwards (Charters et al., 1993; Müller-Beck, 1965; Regert et al., 1998, 2003; Urem-Kotsou et al., 2002).

In the Near East, tar or pitch produced from naturally occurring bitumen is reported as hafting cement and is also evident from the Middle Paleolithic period (e.g. Boëda et al., 1996, 2008, 2009; Hauck et al., 2013; Monnier et al., 2013). The use of *Podocarpus elongatus* (Yellowwood) with regionally different hafting technologies has also been documented in South African sites (Charrié-Duhaut et al., 2013). In the Diepkloof Rock Shelter, the adhesive was associated with the Howiesons Poort and was mixed with bone and quartz grains while in the Sibudu Cave, ochre was found as an additive (Lombard, 2006, 2007; Wadley et al., 2009). Underneath plant residues found on segments in the Sibudu Cave, 67% consisted of resin or gum (Lombard, 2008). Furthermore, there is variability in haft materials through time. These analyses suggest the oldest tools have been hafted to bone and the younger ones hafted to wood (Lombard and Wadley, 2009).

In Southeast Asia, resinous residues probably from *Shorea* spp., *Agathis* spp., or *Canarium* spp. were found on lithic implements from terminal Pleistocene layers at Ille Cave, Palawan (Pawlik, 2012), and have also been identified on stingray spines used as hafted projectile points in the terminal Pleistocene at Niah Cave, Borneo (Barton et al., 2009).

In Australia, the use of various resin types is reported. Two of the more common ones were derived from a grass tree (*Xanthorrhoea*) (e.g. Cribb and Cribb, 1981:89; Leiper, 1982; Zola and Gott, 1992:59) and from Spinifex (*Triodia pungens*) (Gamage et al., 2012; Mondall et al., 2012), (*Triodia iiritans*), (e.g. Boot, 1993:5). Other Australian resin types reported as cement for hafting stone implements to wooden handles include beefwood.
(Grevillea striata), sugarwood (Myoporum platycarpum), cypress pine (Callitris collumellaris), and kurrajong (Brachychiton populneus) (e.g. as described in Boot, 1993:5).

4.1.1. Archaeological study sites

4.1.1.1. Friesack 4

Friesack 4, a bog site located in Brandenburg (Germany), was occupied for approximately 3200 years and contains 100 Mesolithic layers identified in 6 different trenches. According to radiocarbon dates, Mesolithic settlement first began in the middle Preboreal period around 9000 cal BC and ended around 5800 cal BC during the Early Atlantic period (Gehlen, 2009; Görsdorf and Gramsch, 2004), with a hiatus of several hundred years during the middle Boreal period. To date, this represents the most detailed stratigraphy known from the Mesolithic period in Europe. The excellent preservation conditions revealed numerous wooden and antler objects as well as thousands of bones and 140,000 stone artefacts (Gehlen, 2009; Gramsch, 1990, 2001, 2006, 2009/2010, 2011).

4.1.1.2. Wesseling

The open site of Wesseling is located within an old channel of the Rhine River in the western part of Germany. Excavations revealed 6 activity zones with typical late Paleolithic stone tools such as backed points, backed knifes, scrapers and burins. The site also contains pebble plasters interpreted as working areas, several sandstone grinding plates and flat, geometrically-shaped, brown coal objects (Heinen, 2008; Heinen et al.,
Four AMS radiocarbon dates suggest an approximate date of ~11,500 BP for the site’s occupation (AMS-Labor Erlangen, 2010).

4.1.1.3. Yelgun

Yelgun, in north-eastern New South Wales, Australia, is located on a ridgeline overlooking a coastal shoreline and plain. The site consists of a stone artefact scatter of 159 lithic tools and 60 ochre pieces. Artefacts include cores, flakes, scrapers, ground edge tools and are held in a private collection. Eight stone artefacts of the bungwall pounder type (e.g. Hall et al., 1989) possibly suggest a late Holocene age for the site.

4.2. Materials and Methods

Seven stone tools from the three sites were analyzed in this study. Table 4.1 provides information about the lithic type, inherent residues and methods applied. In the following sections, these methods are described in the sequential order they were applied to the stone tools. An exception is the experimental design (section 2.3.) in which only modern fabricated artefacts were utilized.

4.2.1 Remarks on conditions of GC/MS analyses previously carried out on Friesack samples

From the four Friesack samples small fractions were gradually extracted using the solvents isooctane, methanol, chloroform and methanol-oxalic acid. The extracts were then injected directly and as a derivative (methylation with TMSH) into the gas chromatograph
mass spectrometer combination and subsequently analysed (Baumer and Dietemann 2008).

An Agilent GC 6890 N gas chromatograph was coupled with an Agilent MSD 5975 quadruple mass spectrometer; the GC was equipped with J&W capillary column (DB5-ht,

<table>
<thead>
<tr>
<th>Archaeologic sample/type</th>
<th>Origin/ID</th>
<th>Anticipated Age</th>
<th>Optical res. interpret.</th>
<th>Pre-treatm.</th>
<th>Extraction/Removal by</th>
<th>ANSTO ID of AMS dated samples</th>
<th>SEM-EDX remain. residues</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water rolled cobble with dark residue patches</td>
<td>Yelgun, NSW, Australia</td>
<td>Late Holocene</td>
<td>Adhesive material</td>
<td>2% Decon90, 2M HCl</td>
<td>Scalpel scrape off</td>
<td>OZQ 696-U1</td>
<td>Yes</td>
</tr>
<tr>
<td>Flake with dorsal dark residue concentration</td>
<td>Wesseling, Germany (NW 2008/1001)</td>
<td>~ 11,500BP</td>
<td>Adhesive material</td>
<td>2% Decon90</td>
<td>Scalpel scrape off</td>
<td>OZQ 695-U1</td>
<td>Yes</td>
</tr>
<tr>
<td>Flake fragment with dark residue concentration</td>
<td>Friesack, Germany (C3/S_9b)</td>
<td>9,640 ± 60 BP</td>
<td>Adhesive material (also Pawlik 2011a)</td>
<td>2% Decon90</td>
<td>DCM immersion, AAA</td>
<td>OZQ 694-U1</td>
<td>No</td>
</tr>
<tr>
<td>Core axe with use-wear associated wood fibres on both working ends (Further wood and adhesive-wood mix discovered after Decon treatment).</td>
<td>Friesack</td>
<td>between 9,000 and 9,250 BP</td>
<td>Adhesive material clinging to wood</td>
<td>2% Decon90, AAA</td>
<td>Sample was tweezer picked from tool, immersed in DCM dried down in combustion tube</td>
<td>OZQ 689-U1</td>
<td>Yes</td>
</tr>
<tr>
<td>Core axe with use-wear associated wood fibres on both working ends</td>
<td>Germany (DS/7)</td>
<td>between 9,000 and 9,250 BP</td>
<td>Wood</td>
<td>2% Decon90, AAA</td>
<td>tweezer picked from beaker, after Decon cleanse</td>
<td>OZQ 689-U2</td>
<td></td>
</tr>
<tr>
<td>Core axe with use-wear associated wood fibres on both working ends</td>
<td>Friesack, Germany (B2/7)</td>
<td>~ 9,200 BP</td>
<td>Wood, (also Pawlik 2011a)</td>
<td>2% Decon90, AAA</td>
<td>Pick out wood from Decon solution</td>
<td>OZQ 690</td>
<td>No</td>
</tr>
<tr>
<td>Core axe with use-wear associated wood fibres on both working ends</td>
<td>Friesack, Germany (25_10b)</td>
<td>9-10,000 BP</td>
<td>Wood, (also Pawlik 2011a)</td>
<td>2% Decon90, AAA</td>
<td>Scalpel scrape off</td>
<td>OZQ 691</td>
<td>No</td>
</tr>
<tr>
<td>Scraper with use-wear associated wood fibre</td>
<td>Friesack, Germany (CO15/8a)</td>
<td>9,640 ± 60 BP</td>
<td>Wood</td>
<td>2% Decon90, AAA</td>
<td>Pick out wood from Decon solution</td>
<td>OZQ 692</td>
<td>No</td>
</tr>
</tbody>
</table>
Helium was used as the carrier gas at a flow rate of 1.5 ml/min. Samples were introduced via splitless mode in the injection port at a temperature of 250 °C. The column temperature was initially held at 55 °C for 2 min then increased to 360 °C at a rate of 10 °C/min. The GC/MS interface temperature was set at 280 °C. The ionisation energy was 70 eV and the ion source was set at 250 °C under electron ionisation (EI) conditions. The scan range was from 40 to 500 m/z. The GC/MS interface temperature was set at 280 °C. Output files were analyzed using NIST98 Mass Spectral Database (further analysis details in Koller and Baumer, 2010).

GC/MS requires small amounts of pure organic material (~ 1- 0.5 mg) (personal communication Baumer) that will be dissolved in the analyses process. The method is therefore considered destructive.

4.2.2. Optical residue and use-wear analyses

Initially, the seven stone tools were examined by using a low-power light microscope (stereo microscope, Olympus, CX40) with microscope photography (Olympus DP12) at 7x to 115x magnification ranges and high power light microscope (Olympus BX51) with microscope photography (MicroPublisher5.0 RTV) up to 1000x magnification ranges.

Edges, dorsal and ventral surfaces from flakes and at least two surfaces from core tools were examined. The occurrence of residues and use-wear traces was mapped, recorded, imaged and described.

After chemical treatment and removal methods were applied, each artefact was examined again to assess the quantity of residues left and potential changes to the stone tools’ surfaces.

87
Visual residue and use-wear classification was conducted using established analytical criteria and compared with published material (e.g. Fullagar, 2006; Haslam et al., 2009; Hardy and Garufi, 1998; Lombard, 2008).

### 4.2.3. Experimental design to establish decontamination protocols

The optical analyses showed that besides archaeological residues, some artefacts exhibited pencil graphite markings that could potentially contaminate sampling for AMS dating.

To overcome graphite contamination an experiment was set up to test graphite removal methods which simultaneously preserve birch bark tar residue. This experiment involved the production of ten stone flakes and birch bark tar as well as the use of several cleaning methods.

Modern birch bark tar was produced by using sealed steel-sheet containers (5 cm diameter, 10 cm length), also known as retorts (e.g. Weiner, 1988, 1991), filled with birch bark rolls. The containers were placed in a charcoal fire for 15 min at 300–350 °C which transformed the bark into a viscous substance (~50% yield from original bark material). The still warm tar was then chewed to eliminate charcoal remnants and to homogenize the substance. The so processed substance was formed into lumps which were stored for further processing. This production procedure relied on inferred prehistoric conditions inspired by Mesolithic and Neolithic birch tar pieces with chewing marks and birch bark tar production experiments (Aveling and Heron, 1998, 1999; Charters et al., 1993; Palmer, 2007). Hardened modern birch tar was made viscous by holding it under a flame. The viscous mass was then deposited on the right margin of the proximal ventral surface of ten replicated chert flakes. In addition, five graphite marks were sketched onto the fabricated
chert flakes - two from a 2B pencil, two from an HB pencil and one from a 2H pencil (using Faber Castell and Staedtler pencils). Pencil types were selected according to specifications given by stone tool graphic artists.

Decon 90 (2% + 5% diluted), acetone, 2M hydrochloric acid (HCl), 2M sodium hydroxide (NaOH) and an ultrasonic bath (LEO Ultrasonic Cleaner, LEO-50) were trialed for cleaning the artefacts prior to sampling. Decon is in common use in the ANSTO laboratories for cleaning equipment used in AMS radiocarbon sample preparation. With the exception of acetone (known for its degreasing properties, e.g. for removing finger grease), the remaining agents were chosen in accordance with research undertaken by Keeley (1980) and Loy (1987, 1990). While this previous research aimed to remove mineral, carbonate and extraneous organic deposits, our study aimed to understand the efficiency of each agent on graphite contamination as well as the potential damage to the deposited birch bark tar and to the stone surface. For each method, two chert flakes were utilized. They were immersed in the respective liquid for two hours at room temperature, or sonicated in intervals up to 30 min.

**4.2.4. Methodological approach on artefacts - preparation for AMS dating**

**4.2.4.1. Cleaning pre-treatment with Decon 90, 2% diluted**

Because Decon 90 (2% diluted) delivered the best results in keeping birch tar and removing graphite in the above described experimental procedure, this agent was used on the seven archaeological stone tools to remove contamination. Each artefact was immersed in solution at room temperature for two hours.
The other methods applied to the archaeological samples are presented in Table 4.1.

4.2.4.2. Residue Removal

Three removal methods were carried out: (1) removal by scraping with a fresh, unused scalpel under a microscope; (2) penetrating adhesive residues from Mesolithic and Late Paleolithic artefacts with dichloromethane (DCM), a known solvent for birch bark tar (Urem-Kotsou et al., 2002), and capturing the solution for later evaporation; and (3) some residues already dissolved in the Decon 90 (2% diluted) cleaning solution were centrifuged several times and rinsed with Milli-Q™ water.

4.2.4.3. AMS dating

Table 4.1 shows samples prepared with the acid-alkali-acid method (AAA) (2M HCl, 1% NaOH, 2M HCl) to remove carbonaceous contaminants such as carbonates and soluble organics. For a detailed description of the method, see Yates et al. (2014).

Residue samples were converted to CO₂ by combustion using the sealed-tube technique (Vandeputte et al., 1996) and then graphitized as outlined in Hua et al. (2001). Samples were then radiocarbon dated at the ANTARES AMS facility at ANSTO, Australia (Fink et al., 2004).

4.2.5. Establishing elemental characteristics of residues

A Zeiss EVOLS/15 scanning electron microscope with attached EDX was used as a means to further interpret adhesive residues. Although considered non-destructive, the analyses had to be carried out after partial residue extraction for AMS dating on the remaining
residues. This was required because of potential contamination from oil vapor or carbon particles in the SEM chamber.

X-ray analyses were performed in High Vacuum SEM mode whilst using a Back Scatter Detector (BSD). The high vacuum analysis was preferred to enhance accuracy. The BSD allowed interpretation of the various residues present because the differential atomic weights of materials appear in varied shades of grey. The contrast is based on detecting areas with different atomic numbered elements, e.g. more carbon-based (organic) materials look darker, while silica (from a stone tool) appears lighter.

All samples were left uncoated to avoid destruction and to allow possible further analyses using other methods. Therefore, images were taken in Variable Pressure SEM to overcome the absence of a coating. Due to the size of some artefacts, residue samples had to be extracted for analysis and were placed on aluminum stubs. Other residues attached to smaller stone tools (OZQ695) were analyzed in situ.

4.3. Results and Discussion

4.3.1. Optical residue and use-wear analyses

Three stone tools, OZQ695 (Late Paleolithic, Wesseling, Germany), OZQ694 (Mesolithic, Friesack Germany) and OZQ696 (Yelgun, Australia) showed macroscopically visible dark-blackish residues (Figure 4.1, images 1–3). These were interpreted as a form of adhesive because of their mud-cracked and smooth-droplet appearance, some of which suggested embedded plant tissue (e.g. Fullagar, 2006:218; Lombard, 2008) (Figure 4.1,
images 4–15). OZQ695 showed a black residue with a partial droplet-like appearance (Figure 4.1, images 4–6). Non-use related residues on OZQ695 were interpreted as graphite overlaying putative adhesive residues, modern fabric fiber and a white round mass, possibly fungus (Figure 4.1, images 7–9). Dark residue on artefact OZQ694 varied in color between dark brown and black. The brown colored residues often appeared to have a plant tissue structure, while the darker residues had more droplet patterns (Figure 4.1, images 10–12). OZQ696 showed a homogenous black residue with a smooth surface and a mud-crack structure, in this case with sediment attached (Figure 4.1, images 13–15).

Examination of OZQ694 showed wear-related scars which could indicate hafting (e.g. Odell, 1994; Rots, 2010), whereas OZQ695 and OZQ696 showed no such patterns.
Figure 4.1. Macroscopic and microscopic images of stone tools containing putative adhesive residues: 1 Late Paleolithic flake (OZQ695), 2 Mesolithic flake fragment (OZQ694), 3 undated cobble fragment (OZQ696), 4–9 OZQ695: Microscopic images of 4 adhesive concentration 32x mag., 5 partially droplet appearance of adhesive, 6 white mass overlays adhesive, possibly bone collagen, 115x mag., 7 pencil graphite overlaying adhesive residue, 8 modern fabric fiber, 9 possible fungus contamination. 10–12 OZQ694: Microscopic images of 10 dark droplet appearance of adhesive with brown (plant) tissue, 11 edge area with fibrous (plant) material and dark droplet like adhesive spots 100x mag., 12 right margin edges damaged and fibrous (plant) material 20x mag., 13–15 OZQ696: Microscopic images of 13 presumably adhesive chunk mixed with sand 25x mag., 14 adhesive patch cross section at 20x mag., 15 adhesive chunks and mud cracked appearance of adhesive parts at 7x mag.
Wooden residues in the form of wood fiber were observed within retouch bows of the ‘working edges’ (Figure 4.2) of the Mesolithic stone tools from Friesack. The three core axes, (OZQ689, OZQ690 and OZQ691) showed working edges on both ends, confirming previous analyses by Pawlik (2011a). Wooden residues were identified predominantly in the retouched working edge of OZQ692 (scraper), and only occasionally on other parts of the tool edge. Traces of pencil graphite were observed on the edges and tool surfaces of all four artefacts. Modern fabric fiber and hair was also found on OZQ690 and OZQ692.

**Figure 4.2.** Macroscopic and microscopic images of Mesolithic stone tools with identified wooden residues associated with use-wear traces. 1st row: OZQ689 (Friesack, D5/7), Core axe, (Scale 1 =1000 μm, 2 = 500 μm, 3 = 200 μm). 2nd row: OZQ690 (Friesack, B2/7), Core axe (Scale, 1 and 2 = 500 μm, 3 = 2000 μm). 3rd row: OZQ691 (Friesack, F25/10b), Core axe, (Scale 1 and 3 = 1000 μm, 2 = 200 μm). 4th row: OZQ692 (Friesack, CO15/8a), Scraper, (Scale 1 and 3 = 500 μm, 2 = 1000 μm).
4.3.2. Experimental design to establish decontamination protocols

A 2% Decon 90 solution proved the most effective cleaning agent for maintaining the birch tar and for weakening all degrees of graphite hardness. However, a light wipe was necessary to complete the removal of graphite from the stone surfaces. The graphite marks in the 5% Decon 90 solution were visibly faded and some smaller parts of the birch tar were removed. Sonication removed graphite marks as well as all birch tar residues (on a stone flake lying face down in the floating boat). Sonication of another stone flake, lying face up and covered with Milli-Q™ water, had little effect on removing the graphite and no effect on the birch tar. Immersion of flakes in acetone resulted in the partial dissolution of both graphite lines and birch tar. The use of a 2 M HCl solution slightly weakened the graphite lines, while removing ~ 40% of the birch tar. Treatment with 2 M NaOH had no effect on the graphite marks and only small amounts of birch tar were removed on one flake, while on another flake graphite marks were weakened with no effect on the birch tar.

The limitations of these experimental procedures lies in the fixation of the birch tar deposit to the tool. The reproduced fixative appeared less solid and less strongly attached to the contemporary flakes than adhesive observed on archaeological stone tools. Therefore, the cleaning treatment effects on adhesives may be regarded as indicative only.

4.3.3. Methodological approach on artefacts - preparation for AMS dating

4.3.3.1. Cleaning pre-treatment with Decon 90, 2% diluted

Loose graphite traces, skin scales, fabric fiber and fungi were removed from the tools, whilst ink writing was unaffected by the Decon 90 treatment. Faint graphite lines could be
wiped off using a nylon cloth followed by Milli-Q™ water rinses. No changes on the stone tool surface were observed.

Putative adhesive residues: Two samples (OZQ695 and OZQ696) showed no macroscopically observable change. Sample OZQ694 showed residue dislodgement after 7 min of immersion. The artefact was therefore removed from the solvent and then oven-dried at 35°C and freeze-dried at -52°C. The centrifuged and Milli–Q™ water-rinsed solution was retained. Examination by light microscopy revealed that most of the residues on the ventral and on large parts of the dorsal surface had been removed.

Wood residues: All four samples (OZQ689, OZQ690, OZQ691, OZQ692) showed partial detachment of wooden residues. Microscopic examination showed some wooden residues were still present in retouched working edges of OZQ690 and OZQ692. From one Mesolithic artefact, further residues were uncovered which were previously trapped in a cavity. The Decon treatment washed out the material previously filling the rock cavity. Two samples, 1) wooden residues (OZQ689-U2), and 2) wood mixed with dark substance residues (OZQ 689U1) (Figure 3) were taken and processed for AMS dating.

Overall, these results show that along with some cleaning effect, the wooden residues did get dislodged from the stone tool. While this may be a desired effect when extraction is the aim, in this situation it is possible that graphite particles may have been transferred into the sample. On the other hand, as a substitute for the ‘Alkali’ step in the Acid-Alkali-Acid treatment, the solution is believed to be effective at removing fulvic and/or humic acid contamination. Wooden residues captured in the solution were rinsed with Milli-Q™ water, centrifuged and further treated as described in Table 4.1.
4.3.3.2. Residue Removal

4.3.3.2.1. Scraping. Removal of residue deposits by scraping with a fresh scalpel blade was conducted on tools with putative adhesive residues. Two samples were taken from OZQ696, one from OZQ695, and one sample from OZQ691. The extracted residues were captured in centrifuge tubes. Powder-free gloves were worn at all times and changed between samples and intermittently.

4.3.3.2.2. Dichloromethane (DCM). On OZQ694 and OZQ695, extraction was trialed with dichloromethane (DCM) because the artefacts’ find contexts suggested birch bark tar as a likely adhesive residue type.

Using a syringe containing ~ 1 mL of DCM, the adhesive deposit on OZQ695 was sprayed in two consecutive steps. The collected DCM was drawn up into the syringe and repeatedly re-applied to the stone tool deposit. The DCM was then transferred into a combustion tube. No change in color was observed in the DCM solution. There was little effect on the deposit.

For OZQ694, two sample fractions were obtained by using DCM. First, the captured residues from the Decon immersion were centrifuged and rinsed with Milli-Q™ water. The organic material was then treated with DCM followed by drying in a combustion tube. Second, the remainder of the residue which was still adhering to the tool was separated by immersion in DCM, followed by 20 min of ultrasonification. The solution was then evaporated on a hotplate and dried in a combustion tube.

4.3.3.2.3. Decon 90, 2% diluted. OZQ690 and OZQ692 were extracted unintentionally by the Decon immersion. These samples were further prepared as described in the method.
section and Table 4.1. The wood-black substance mix from OZQ689-U1 (uncovered after Decon immersion) was removed from the tool using tweezers (Figure 4.3). A second sample, OZQ689-U2, consisted of the wooden remnants picked out from the beaker after the tool was Decon-cleaned and desiccated.

![Figure 4.3](image.png)

**Figure 4.3.** Detailed picture of OZQ689 wooden residues intermixed with dark deposit from an unknown substance (possibly adhesive residue) (200x magnifications).

In total, eleven residue samples were removed from seven stone tools. Three of these samples yielded less than 5 µgC, which was insufficient to proceed to AMS (Table 4.2).
### Table 4.2. Summary of residue sample dating by AMS. Radiocarbon ages are indicated in BP and previously obtained dates are from Görtsdorf and Gramsch (2004) and Gramsch, (2000, 2012). (Abbreviations: D = Decon90, 2% diluted, DCM = dichloromethane treated, AAA = Acid-Alkali-Acid, pMC = Percent Modern carbon).

<table>
<thead>
<tr>
<th>ANSTO ID</th>
<th>Residue Interpretation</th>
<th>Pre-treatment</th>
<th>Carbon Mass</th>
<th>pMC</th>
<th>Radiocarbon age BP</th>
<th>Anticipated Age</th>
<th>SEM-EDX interpretation</th>
</tr>
</thead>
<tbody>
<tr>
<td>OZQ 696- U1 Yelgun, NSW</td>
<td>Adhesive material</td>
<td>D, scrape extract., untreated</td>
<td>44.38µg</td>
<td>77.1</td>
<td>2089 ± 67</td>
<td>Late Holocene</td>
<td>Organic, substance</td>
</tr>
<tr>
<td>OZQ 696- U2 Yelgun, NSW</td>
<td>Adhesive material</td>
<td>D, scrape extract., 2M HCl</td>
<td>33.30µg</td>
<td>80.09</td>
<td>1783 ± 60</td>
<td>Late Holocene</td>
<td></td>
</tr>
<tr>
<td>OZQ 695- U1 Wesseling</td>
<td>Adhesive material</td>
<td>D, scrape extraction</td>
<td>35.30µg</td>
<td>338.9</td>
<td>- 9805 ± 14</td>
<td>~ 11,500BP</td>
<td>Inorganic substance, possibly Manganese Oxide/dendrites</td>
</tr>
<tr>
<td>OZQ 695- U2 Wesseling</td>
<td>Adhesive material</td>
<td>D, DCM penetration,</td>
<td>4.87µg</td>
<td>_</td>
<td>Insufficient carbon</td>
<td>~ 11,500BP</td>
<td></td>
</tr>
<tr>
<td>OZQ 694- U1 Friesack</td>
<td>Adhesive material</td>
<td>D, DCM imm, ultrasonication</td>
<td>0.23mg</td>
<td>17.4</td>
<td>14,130 ± 90</td>
<td>9,640 ± 60 BP</td>
<td></td>
</tr>
<tr>
<td>OZQ 694- U2 Friesack</td>
<td>Adhesive material</td>
<td>D, DCM, AAA</td>
<td>4.09µg</td>
<td>_</td>
<td>Insufficient carbon</td>
<td>9640 ± 60 BP</td>
<td></td>
</tr>
<tr>
<td>OZQ 689- U1 Friesack</td>
<td>Adhesive material /wood</td>
<td>D, tweezer picked from tool, DCM,</td>
<td>137.14µg</td>
<td>10.54</td>
<td>18,370 ± 140</td>
<td>between 9,000 and 9,250 BP</td>
<td>Wood, amorphous, organic substance and shell</td>
</tr>
<tr>
<td>OZQ 689- U2 Friesack</td>
<td>wood</td>
<td>D, tweezer picked from beaker, AAA</td>
<td>18.97µg</td>
<td>36.28</td>
<td>7,890 ± 180</td>
<td>between 9,000 and 9,250 BP</td>
<td></td>
</tr>
<tr>
<td>OZQ 690 Friesack</td>
<td>wood</td>
<td>D, Pick out wood, AAA</td>
<td>4.90µg</td>
<td>_</td>
<td>Insufficient carbon</td>
<td>~ 9,200 BP</td>
<td></td>
</tr>
<tr>
<td>OZQ 691 Friesack</td>
<td>wood</td>
<td>D, scrape extraction, AAA</td>
<td>9.20µg</td>
<td>79.45</td>
<td>1848 ± 124</td>
<td>9-10,000 BP</td>
<td></td>
</tr>
<tr>
<td>OZQ 692 Friesack</td>
<td>wood</td>
<td>D, Pick out wood, AAA</td>
<td>8.68µg</td>
<td>54.69</td>
<td>4848 ± 244</td>
<td>9,640 ± 60 BP</td>
<td></td>
</tr>
</tbody>
</table>

99
4.3.3.3. AMS dating

The residue radiocarbon dates delivered mixed results. There were some agreement and also some significant deviations from the anticipated age (Table 4.2, Figure 4.4).

![Residue radiocarbon ages (rounded) plotted against anticipated ages. From left to right: Both samples of OZQ696 date within expected Late Holocene age range, OZQ695 significantly too young, OZQ694 and OZQ689-U1 significantly too old, 6 OZQ689-U2 dates close to anticipated age, OZQ691 and OZQ692 too young.](image)

**Figure 4.4.** Residue radiocarbon ages (rounded) plotted against anticipated ages. From left to right: Both samples of OZQ696 date within expected Late Holocene age range, OZQ695 significantly too young, OZQ694 and OZQ689-U1 significantly too old, 6 OZQ689-U2 dates close to anticipated age, OZQ691 and OZQ692 too young.

4.3.3.3.1. Adhesive residues. Both adhesive residue samples from the Australian stone tool (OZQ696-U1/U2) dated within the expected Late Holocene time frame. The ages of ~ 2 ka BP correspond with dates reported for the ‘bungwall pounder’ type artefact (e.g. Hall et al., 1989:155 and McNiven, 1992:706 who suggest possible use from Mid Holocene), which were present at the site with OZQ696. As such, they provide a clue to the age of the assemblage. Discrepancies between the samples’ ages may be related to differential treatment — for instance only one sample was acid treated (OZQ696-U2).
CHAPTER 4  RADIOCARBON-DATING ADHESIVE AND WOODEN RESIDUES FROM STONE TOOLS BY ACCELERATOR MASS SPECTROMETRY (AMS): CHALLENGES AND INSIGHTS ENCOUNTERED IN A CASE STUDY

OZQ695-U1 yielded a surprising result. The sample measured a radiocarbon activity of 338.9 per cent of modern carbon (pMC) (corresponding to about 10 kA in the future) (Table 4.2). Even during the bomb pulse peak (~ 1963 AD), atmospheric CO₂ activity only doubled to about 200 pMC. Levels higher than this must be associated with nuclear technologies or ¹⁴C tracers used in industry and medicine. We conclude, therefore, that this sample inadvertently came into contact with such materials. Our investigations have not revealed any clues as to how this might have occurred.

The two remaining adhesive residues from Friesack dated significantly older than the anticipated age.

The OZQ694 (Friesack C3/S_9b) residue’s AMS date delivered an age of 14,130 ± 90 BP, which is at variance with the date of the stratigraphical position of the tool, 9640 ± 60 BP. In addition to the typological assignment of the residue-dated tool being a typical Mesolithic core-axe, the find circumstances within a bog site which developed after the Pleistocene and from the middle Preboreal Period onwards (Gramsch, 2001, 2006), somewhat exclude the occurrence of artefacts of this age on this site. Taphonomic processes or bioturbation as factors causing artefacts to move between soil layers would only be an explanation if, underneath the bog site layers, older Pleistocene archaeological deposits had been discovered. However, this is not the case. Therefore, one possible explanation is that the residue attached to the artefact consisted of a material that was older than organic material collected during the Mesolithic period. Although not previously reported in a Mesolithic context, the use of naturally occurring bitumen would be one potential explanation. Natural bitumen exists in the Nordhorn deposit, in the Northwest German basin (Clarke and Trinnaman, 2010:145), and reportedly is present e.g. in
4.3.3.2. Wooden residues. In contrast, the second sample fraction of the core axe (OZQ689-U2) was dated to 7,890 ± 180 BP, which is ~1000 years too young. The tool was excavated from layer 7 of section Z in Friesack. Layer 7 was not radiocarbon dated, however, layer 17, occurring underneath layer 7, delivered dates of 9180 ± 70 and 9240 ± 70 BP (Gramsch, 2001:61). Layer 6c, present above layer 7, yielded dates of 8980 ± 60, 9010 ± 70 and 9040 ± 70 years BP. In addition, layer 7 in section A was dated to 8850 ± 70 and 8975 ± 70 years BP (Gramsch, 2001:61). These dates suggest an age between ~9000 and ~9250 years BP for artefact OZQ689. One possible explanation for this might be that the tool moved down from higher (younger) layers (e.g. through bioturbation or taphonomic processes). Furthermore, it is possible that the wooden residues (or parts of
them) found in the cavity of the stone artefact were actually not use-related, but the result of root growth. In addition, contamination by modern carbon may be responsible for the age deviation of the small sample size consisting of 18.97 µgC.

Remaining wooden residues dated significantly too young compared with the anticipated age (Table 4.2, Figure 4.4). Residue from OZQ692 was removed unintentionally during the Decon clean and as such was intermixed with surface contamination including pencil graphite. Therefore, it is possible that contamination affected the radiocarbon date of the sample which was sized only 8.68 µgC. While graphite remains would have led to an older age, other unknown factors may have contributed to the younger age. The possibility of the tool having been moved through the sediment is unlikely as we are not aware of core axes typologically evident from ages as young as 4848 ± 244 years BP.

A microscopic image of wooden residue remains from OZQ691 suggested a possible precipitation of calcite on the artefact’s wooden residue. For this possibility to exist, an older age would be expected (e.g. Long et al., 1992). In general, it was found that the influence on artefacts of carbon atoms from water in bog sites usually results in an age that is too old (van der Plicht et al., 2004: 472, 473). However, any carbonate contamination deposited by groundwater should have been removed during the acid steps of the AAA pre-treatment. Long storage in a warm environment, inviting fungi and microbe development and decay, may introduce $^{14}$C-rich CO$_2$ into the sample and may also result in radiocarbon dates that are too young – as has been suggested for pollen age deviations (Neulieb et al., 2013). We find it unlikely that contamination occurred during the scrape extraction as only fresh, unused scalpel blades were used. The dates yielded for OZQ691 and OZQ692 were not congruently “false” but showed a ~3000 year difference between
them, while the anticipated age for both samples was between 9000 and 10,000 years BP (Table 4.2). For the above reasons, we suggest that the small sample size, and introduced atmospheric or modern carbon might be the more likely reason for the significantly younger age of the samples. The wide span of incongruence suggests an unknown amount of carbon as well as an unknown type of carbon introduced into the samples.

4.3.4. Establishing elemental characteristics of residues

Residues were still present on tools OZQ689, OZQ695, OZQ696 after the aforementioned treatment. This allowed the use of SEM-EDX analysis to establish the basic elemental composition. In addition, a modern Xanthorrhoea sample was analyzed for comparison.

4.3.4.1. OZQ689

On the one hand, we suspect that fossil bitumen might be responsible for the older radiocarbon age. On the other hand, however, SEM images also showed the presence of shell which was likely to have been formerly embedded in the flint stone matrix. This shell could also have contributed to radiocarbon age overestimations. In addition, wood fiber, silica and the putative adhesive (amorphous black substance) were observed by microscopy (Figure 4.5). Published EDX data on archaeological bitumen residue show different elemental composition compared to the sample data (absence of P, Ca and Fe, but presence of Na, S and Zn) (Monnier et al., 2013:3729–3732) (Table 4.3). However, elemental compositions of bitumen are known to vary from source to source (e.g. Brown et al., 2014). Also, addition of other materials to produce a fixative and the resultant
differential decay may contribute to the differences. Further methods are required to clearly identify the nature of the residues.

Figure 4.5. SEM images from OZQ689 residue: 1 Unknown substance possibly adhesive material with silica chunk attached right side, (Scale = 50 μm); 2 and 3 Unknown organic substance (Scale = 20 μm); 4 from left to right: silica chunk, partially unknown substance and wood piece (Scale = 200 μm); 5, 6 shell with unknown residue (Scale = 50 μm).
Table 4.3: Extracted residue from cavity of OZQ689: Weight Percentages determined from EDX data for elements >1% average atomic weight.

<table>
<thead>
<tr>
<th>ID</th>
<th>Description/Interpretation</th>
<th>Average atomic weight:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C</td>
<td>O</td>
</tr>
<tr>
<td>1</td>
<td>Silica particle</td>
<td>35</td>
</tr>
<tr>
<td>2</td>
<td>Black amorphous matter</td>
<td>22</td>
</tr>
<tr>
<td>3</td>
<td>Black amorphous matter</td>
<td>22</td>
</tr>
<tr>
<td>4</td>
<td>Black amorphous matter</td>
<td>40</td>
</tr>
<tr>
<td>5</td>
<td>Black amorphous matter</td>
<td>105.19</td>
</tr>
<tr>
<td>6</td>
<td>Black amorphous matter</td>
<td>99.9</td>
</tr>
<tr>
<td>7</td>
<td>Black amorphous matter</td>
<td>90.23</td>
</tr>
<tr>
<td>8</td>
<td>Black amorphous matter</td>
<td>52.67</td>
</tr>
<tr>
<td>9</td>
<td>Black amorphous matter</td>
<td>79.68</td>
</tr>
<tr>
<td>10</td>
<td>Wood fibre</td>
<td>52.38</td>
</tr>
<tr>
<td>11</td>
<td>Wood fibre</td>
<td>95.51</td>
</tr>
<tr>
<td>12</td>
<td>Shell</td>
<td>4.59</td>
</tr>
<tr>
<td>13</td>
<td>Shell</td>
<td>15.02</td>
</tr>
</tbody>
</table>

4.3.4.2. OZQ695

The high levels of Manganese and the occurrence of Barium (Table 4.4) indicate that the

Table 4.4: Residue and rock matrix of OZQ695: Weight Percentages determined from EDX data for elements >1% average atomic weight.

<table>
<thead>
<tr>
<th>ID</th>
<th>Description/Interpretation</th>
<th>Average atomic weight:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C</td>
<td>O</td>
</tr>
<tr>
<td>1</td>
<td>Black substance on tool tip</td>
<td>10.34</td>
</tr>
<tr>
<td>2</td>
<td>Black substance on tool tip</td>
<td>7.37</td>
</tr>
<tr>
<td>3</td>
<td>Black substance on tool tip</td>
<td>6.92</td>
</tr>
<tr>
<td>4</td>
<td>Black substance on tool tip</td>
<td>-2.99</td>
</tr>
<tr>
<td>5</td>
<td>Black residue streaks on tool</td>
<td>47.95</td>
</tr>
<tr>
<td>6</td>
<td>Black residue streaks on tool</td>
<td>2.48</td>
</tr>
<tr>
<td>7</td>
<td>Black residue streaks on tool</td>
<td>3.85</td>
</tr>
<tr>
<td>8</td>
<td>Black residue streaks on tool</td>
<td>34.48</td>
</tr>
<tr>
<td>9</td>
<td>Black residue streaks on tool</td>
<td>3.16</td>
</tr>
<tr>
<td>10</td>
<td>Black residue streaks on tool</td>
<td>35.99</td>
</tr>
<tr>
<td>11</td>
<td>matrix of rock</td>
<td>4.46</td>
</tr>
<tr>
<td>12</td>
<td>matrix of rock</td>
<td>2.83</td>
</tr>
<tr>
<td>13</td>
<td>matrix of rock</td>
<td>14.89</td>
</tr>
<tr>
<td>14</td>
<td>matrix of rock</td>
<td>8.72</td>
</tr>
<tr>
<td>15</td>
<td>matrix of rock</td>
<td>11.69</td>
</tr>
<tr>
<td>16</td>
<td>matrix of rock</td>
<td>10.22</td>
</tr>
</tbody>
</table>
black substance, mimicking the adhesive in appearance, might be a form of manganese dendrites (e.g. Potter and Rossman, 1979). A study analyzing manganese dendrites on different rock varieties by using SEM-EDX has identified Mn and Ba as major components (Xu et al., 2010). The non-organic nature of the dark residue is further suggested by the brighter appearance in the SEM images (Figure 4.6).

![Figure 4.6. SEM images of OZQ695 residue: 1 adhesive streaks (white); 2 adhesive streaks (white) and grey traces from scraping off adhesive; 3 residue on tip area of tool (Scale 1 + 2 = 1.0 µm, Scale 3 = 10 µm).](image)

4.3.4.3. OZQ696

Two residue samples (A and B) were analyzed with A consisting of a chip scraped off the tool, and B being a scraping with powdery consistency. The SEM images show that both samples had a consistent fan-like plant structure which in sample A shines through the sealed part (Figure 4.7, images 1-3). In contrast to this is the very dense and compact consistency and structure of the contemporary Xanthorrhoea sample. The beam caused cracks on the surface of the resin, probably due to the density of the material (Figure 4.7, image 6).
Further clues suggesting different materials are found in the elemental composition of contemporary *Xanthorrhoea* which is limited to high C and O fractions. By contrast, the black to dark green colored adhesive material in samples numbered 1 to 7 (Table 4.5)
shows high average weights of O, and sequentially decreasing weights of Si, C, Fe and K.

Except for samples 3 and 5, Mg is present in all other samples, while Al occurrence is limited to sample 7. Noteworthy are the clearly higher proportions of O, Si, K, Mg and Fe in sample B (6, 7) which had the more powdery consistency. This may indicate that the elemental composition within the material had changed because the outer layer, more exposed to weathering (sample A), contains less of these elements.

**Table 4.5** Residue and rock samples of OZQ696, in comparison with contemporary Xanthorrhoea samples: Weight Percentages determined from EDX data for elements >1% average atomic weight.

<table>
<thead>
<tr>
<th>ID</th>
<th>Description/Interpretation</th>
<th>Average atomic weight:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>C</td>
</tr>
<tr>
<td>1</td>
<td>black-green substance, sample A</td>
<td>7.37</td>
</tr>
<tr>
<td>2</td>
<td>black-green substance, sample A</td>
<td>2.28</td>
</tr>
<tr>
<td>3</td>
<td>black-green substance, sample A</td>
<td>15.08</td>
</tr>
<tr>
<td>4</td>
<td>black-green substance, sample A</td>
<td>5.53</td>
</tr>
<tr>
<td>5</td>
<td>black-green substance, sample A</td>
<td>12.39</td>
</tr>
<tr>
<td>6</td>
<td>black-green substance, sample B</td>
<td>4.44</td>
</tr>
<tr>
<td>7</td>
<td>black-green substance, sample B</td>
<td>4.32</td>
</tr>
<tr>
<td>8</td>
<td>Silica particle</td>
<td>3.1</td>
</tr>
<tr>
<td>9</td>
<td>contemporary Xanthorrhoea</td>
<td>78.7</td>
</tr>
<tr>
<td>10</td>
<td>contemporary Xanthorrhoea</td>
<td>66.97</td>
</tr>
</tbody>
</table>

Comparing images and data of the archaeological adhesive residue material with the contemporary *Xanthorrhoea* sample does not provide great insight. Chemical modifications, decay, the presence of other materials (e.g. sand, animal fur) (e.g. Gamage et al., 2012; Rots, 2010) and weathering may have altered, partially or completely, the SEM surface aspect as well as the elemental composition. The differences in data between archaeological and contemporary material could, however, indicate two different materials.
4.4. Discussion

The detection of several shells by SEM in OZQ689 residue, interpreted as a constituent of flint stone (e.g. Pellant, 2000:246), has important repercussion on inferences about the dates. The AMS age distortions obtained for OZQ689-U1 and OZQ694-U1 could well be explained by the presence of shells. In order to preserve the adhesive residue, both sample fractions were not AAA treated, the acid step of which would have removed shell remains and related age offsets. On the other hand, one cannot determine bitumen presence in the residue solely by SEM-EDX analyses and this could be another cause for age offsets.

Apart from the reasons described above, plasticisers may also be incorporated into the sample from storing the artefact in plastic bags. The GC/MS analyses on the four analyzed Friesack samples has shown every birch tar piece contained plasticisers (one example represented by Figure 4.8) (Baumer and Dietemann, 2008).

Due to the fossil origin of plastic materials, too old radiocarbon dates could be expected. We found diverse information about the effect of plasticisers on radiocarbon dates: First, direct contact with a wood sample caused a ~200 years age overestimations (Hyman and Rowe, 1997:64). Second, "...a surprising level of plasticizer (as phthalate)" was found in lipid samples, although it was considered "...not enough to invalidate the dates." (Hedges et al., 1992:911). - The obtained dates were ~ 5000 and ~ 10,000 thousands of years too old (Hedges et al., 1992:910). The authors suspect laboratory contamination as responsible for the age offsets, however more detail for this assumption was not provided. Third, "considerable" amounts of plasticizes found in birch bark tar sample Königsaue B may have contributed to a > 4000 age overestimation compared with the stratigraphical younger sample Königsaue A, which had "lesser impurities" (Koller et al., 2001:103). For the
latter study it needs to be considered that both samples stratigraphical ages are older (80,000 years and older) than the age range obtainable with radiocarbon dating. These examples indicate the potential effect of plasticisers on wood, lipid and birch bark tar samples. Yet, a more in-depth analyses, that demonstrates measurable evidence of plasticisers effect is missing. This shows that currently there are too many unknown variables possibly responsible for age offsets caused by plasticisers. These variables include residue type, length of storage, plastic type used for storage, heat exposure of sample in plastic bag. A specific radiocarbon dating series testing plasticisers effect with the combination of these variables would help to resolve the problem.

Figure 4.8. Gas-Chromatogram from birch bark pitch of Friesack, find 1977:7/P4. Betulin, lupeol and lupenone, biomolecular markers of birch bark. Additionally plasticizers in form of Phthalatestern, Dibutylphthalat (DBP) (Courtesy of Baumer and Dietemann, 2008, Doerner Institute, Munich).
Finally, graphite contamination needs to be taken into account for age overestimations, despite attempts to remove the material with Decon 90.

SEM-EDX analyses allowed discerning organic from inorganic residues, suggested the presence of manganese dendrites (OZQ695) and detected fossil shell (OZQ689) as a potential contamination source. The possibility to examine residues in situ can be applied as a preliminary step to validate their organic nature. In combination with optical light microscopy, the method demonstrated to provide mutual complementary results for understanding residue and use-wear (e.g. Pawlik, 1995, 2004; Jahren et al., 1997; Dinnis et al., 2009; Pawlik and Thissen, 2011; Borel et al., 2014). EDX results need to be considered as an indication of the basic elemental composition and the data as qualitative rather than quantitative. These results are further impaired by the uneven and rough surfaces of the samples.

Although accurate in results, sample amounts needed for GC/MS are larger than most lithic residues preserve. In addition, for our purpose, to radiocarbon date smallest residues we need to avoid sample destruction which is also desirable for replicate analyses. Vibrational spectroscopic techniques (e.g. Daher et al., 2013) might be the answer to assess the actual nature of residues. That said, a clear evaluation of the impact on the sample, such as damages by the beam or by pressure, the importance of plane surfaces as well as the need for reference material, is mandatory to validate its use (Cesaro and Lemorini, 2012:300; Matheson and McCollum, 2014:125; Prinsloo et al., 2014:738). In addition, the application of techniques such as UV light irradiation and chemiluminescence (e.g. Matheson 2014; Lombard 2014) may aid in residue detection and identification. A practicable workflow could then be as represented in Figure 4.9.
Figure 4.9. Suggested workflow of method sequence for efficient residue AMS dating.

In the context of dating, one can argue for the lesser role that residue type plays in the actual radiocarbon age as long as it is use-related. For example, stone tools are known to have been used for multiple tasks and therefore contain various residues. While this argument might be acceptable for certain types of organic residues (e.g. bone, blood, fat, protein, starch, or wood), adhesive residues might be intermixed with materials that distort the radiocarbon ages. We have pointed out the possibility of bitumen additions as one distorting factor, however, sand, dust, and ground shell might also lead to age deviations. This study has shown that the chemical signature is needed on such samples prior to AMS dating as it would have guided the choice of extraction and preparation protocol.

4.5. Conclusion

The aim of this study was to test the feasibility of radiocarbon dating residues of archaeological stone tools and at the same time to mitigate inherent contaminant impact. We have obtained modest, but encouraging AMS dates for one wooden residue and for one adhesive residue. This indicates the feasibility of residue AMS dating, but it also clearly shows we are still at an early stage of method development in this research field. In this study we demonstrate that successful residue dating requires a thorough identification of their type and the nature of potential contaminants. SEM-EDX and GC/MS were helpful in
detecting potential contamination sources, such as fossil shell, and plasticisers, for age deviations. Further sample analyses from Friesack may reveal whether bitumen, or other sources, could explain the age overestimations. Decon 90, 2% diluted, appears to remove graphite contamination while preserving birch bark tar, however, the effect on other adhesives is unknown and the agent is unsuitable for wooden residues. In general, the applied methodological sequence appears appropriate and led to successful outcomes. Additional characterisation methods at the beginning of the analyses, such as vibrational spectroscopic techniques or the application of biochemical substances, would be beneficial, especially for adhesive residues.

Future research needs to develop residue-specific sampling protocols for these non-destructive characterization methods to avoid contaminant transfer. In addition, a further protocol for artefact handling from excavation through to adequate storage is essential.

4.6. Acknowledgements

We wish to acknowledge ANSTO for supporting laboratory work and AMS measurement through the ICCAS project in the Institute for Environmental Science. We owe thanks and appreciate the Tweed Byron Local Aboriginal Land Council for permitting and endorsing residue radiocarbon dating from stone tools, which were kindly loaned for this purpose. Thanks to: Dr Martine Regert and Dr Thilo Rehren (JAS editors) for valuable comments on earlier drafts on the manuscript which improved this paper substantially; Ms Maxine Dawes, SCU, for her assistance provided with SEM-EDX analyses; Dr Andrew Rose, SCU, for introducing and providing access to Raman Spectroscopy equipment; Ms Ursula Baumer and Dr Patrick Dietemann, Doerner Institut, Munich, for providing Gas
Chromatograms and information about their GC/MS research on birch bark tar; Andreas Kurzweil, Museum Düppel, working group ‘Teerschweele’, Berlin, for discussion and information about bitumen in an archaeological context and experience with this material; and Dr Jürgen Junkmanns, www.pfeil-bogen.de, for producing birch bark tar used in our experiment.
Residue radiocarbon AMS dating review and preliminary sampling protocol suggestions

Unformatted published paper is shown in Appendix 5

ABSTRACT

Radiocarbon dating of microgram residues is a relatively new field in archaeological research and is currently limited by a lack of analytical protocols and instrumentation. Successful applications of Accelerator Mass Spectrometry (AMS) have demonstrated the potential of the technique on small samples but also revealed challenges and problems, especially with contamination. This paper reviews the literature on AMS radiocarbon dated residues using microgram sized carbon samples. Samples from archaeological studies are targeted, including residues from lithics, ceramics and rock art. We examine data helpful to avoiding contamination and to facilitating residue radiocarbon dating. As a result we present a preliminary sampling protocol to assist archaeologists in preventing contaminant transfer from fieldwork onwards.

*Keywords:* Residue; Radiocarbon dating; Contamination; Sampling protocol; Preservation; AMS; Age offsets; Plasticizers, fieldwork, storage
5.1. Introduction

Accelerator Mass Spectrometry (AMS) radiocarbon dating of archaeological residues on a carbon microgram scale is a relatively new field in archaeological chronometric research. Recent successful applications demonstrating the method’s feasibility (Yates et al., 2014, 2015), however, have revealed challenges with contamination when dealing with such small sample sizes. The refinement of the method is significant because it has the potential 1) to provide chronology for artefacts from archaeological contexts that lack other datable organic material (e.g. surface scatters), 2) to validate dates achieved by other means (e.g. within stratigraphy), and 3) to give insights not previously available into artefact movement through stratigraphic layers. In general, it provides archaeologists with an extended range of dateable material and thus could deliver higher resolution chronologies.

In this paper we provide a critical review of AMS radiocarbon-dated residues using microgram sized carbon samples. First, we look for clues in the literature that are relevant to improving residue radiocarbon dating. Second, we use the results and conclusions drawn from that review to design a preliminary sampling protocol for archaeologists to assist in avoiding the introduction of contaminants.

A major focus is on dated residues from stone artefacts. Data in this field is sparse, and therefore residues from rock art and ceramics are considered in order to expand our knowledge about concerns in residue dating. Studies of residue radiocarbon dating are closely linked to residue and use-wear analysis which helps to determine whether residues are use-related or contamination. In this context, it needs to be noted that in the process of
AMS dating, artefact inherent residues (or parts of them) are destroyed. Therefore, they need to be thoroughly recorded, imaged and described before removal in order to document their original state. With these considerations in mind, it is suggested radiocarbon dating of archaeological residues be performed when the study is linked to specific research questions and when other factors, which will be elaborated on further in this paper, promise a successful outcome.

In this study we will review four particularly important aspects of AMS radiocarbon dating: (1) residue identification, (2) preservation, (3) contaminant introduction, and (4) interconnectedness of sample size, yielded carbon mass and residue type. Two case studies illustrate the main points discussed in this evaluation. This approach aims to provide guidance for improving sampling strategies that avoid contaminants compromising AMS dates.

It is hoped this information will improve sampling strategies in the field as well as in storage and sample treatment in the laboratory and thereby improve the accuracy of residue radiocarbon dating. In pursuit of that goal, we present hands-on advice in the form of easy-to-follow sampling steps for the residue-dating of promising stone artefacts.
5.2. Aspects related to AMS radiocarbon dating

5.2.1. Residue identification

5.2.1.1. Microscopic residue interpretation

Microscopic residue interpretation enables us to identify what is to be dated and gives an idea of the amount and presence of various residue types. It also provides information on the degree of intermix with other materials and occurrence of contaminants. Optical residue observation uses diagnostic criteria which help in residue type classification (e.g. Fullagar, 2006a; Hardy and Garufi, 1998; Lombard, 2008), while associated use-wear traces can confirm that residues are use-related (e.g. Fullagar, 2006a; Robertson, 2009, 2011). Furthermore, residue distribution in a patterned way further allows differentiation from contaminant traces (Kononenko, 2011; Langejans, 2011).

5.2.1.2. Biochemical identification and interpretation

For adhesive residues, chemical characterization may confirm the nature of the material and assess the adhesive type. Of relevance to our aims is the understanding that chemical identification techniques are non-destructive and do not introduce contamination. Gas Chromatography Mass Spectrometry (GC/MS) is able to characterize and discern multiple constituents of a material and can also detect otherwise non-identifiable components such as lipids (an extensive overview is given by Evershed, 2008). In relation to residue radiocarbon dating, the technique is valuable for detecting material components when ample residues are present. The method also holds the potential to support dating lipids as separated compounds from cooking pottery (Stott et al., 2001, 2003). However, more
research is needed to assess the introduction of potential contamination during the chemical process when analyzing samples < 100 μg carbon.

Scanning Electron Microscope-Energy Dispersive X-ray (SEM-EDX) analyses, previously helpful in the characterization of birch bark tar (Pawlik, 1997, 2004; Pawlik and Thissen, 2011) allow the elemental composition and imaging of residues and this helps in validating their organic nature. Because SEM-EDX analyses can introduce contamination into samples (Yates et al., 2015), it is preferable to perform the analyses on the remaining portion of the residue, if still present after AMS dating. This is also desirable because, depending on the residue type, the EDX beam can leave a tiny crater or cracks on organic substances.

Recent studies show non-destructive chemical residue characterization is important in quantifying the substance to be dated and in confirming optical residue observation (Cesaro and Lemorini, 2012; Daher et al., 2013; Prinsloo et al., 2014; Monnier et al., 2013). For instance, microscopic identification of blood residues is not always straightforward and there is potential for misidentification with other residues, such as resins. The use of biochemical reaction methods, such as the Hemastix® test, helps in the identification of blood residue, although other residues (Robertson, 2011) and contaminants from handling and soil can cause positive reactions (Matheson and Veal, 2014). Chemiluminescence may possibly help in identifying blood residues on lithic tools that are thousands of years old (Lombard, 2014). Cooked or damaged starch granules can be identified with staining solutions such as Congo Red (Lamb and Loy, 2005). Additional staining agents and their reactions to materials such as lignin, collagen, lipids and further plant varieties are described in Fullagar et al., (2015). To allocate these staining protocols
to a place in the analysis order, they should be tested to determine whether they introduce carbon into residue samples. Depending on the outcome, staining agents could be placed either at the beginning or at the end of the analysis sequence.

For the reasons mentioned above, combining microscopic observation and non-destructive chemical residue identification is therefore a desirable approach at the beginning of the methodological sequence for residue radiocarbon dating.

5.2.2 Residue Preservation

5.2.2.1. Residue preservation in general

It is important to gain an understanding of the conditions under which residues are preserved as well as residue resilience to environmental degradation and laboratory treatment. Preservation of sufficient residue is a critical prerequisite for residue radiocarbon dating. Recent research indicates that several assemblages comprised > 50% of lithics with residues (Hardy et al., 2008: 652; Hardy and Svoboda, 2009: 165; Robertson, 2009: 300, 302). This highlights the potential of using a sample of such preserved residues for radiocarbon dating.

Residues have shown to be best preserved in depositional contexts with alkaline pH, a high cation exchange capacity (such as those containing clay-rich sediments), and water or oxygen-free environments which limit microbial growth (Jones, 2009; Loy, 1987: 58; Gurfinkel and Franklin, 1988; Langejans, 2010). Desiccation can also conserve residues very well and prevent microbial growth (Evershed, 2008). For instance, artefacts buried in waterlogged conditions such as bogs demonstrated exceptional residue preservation (e.g.
Langejans, 2010). Furthermore, anoxic conditions and nutrient limitation, especially of nitrogen (N) and phosphorus (P), prevent microbial activities and thus limit organic degradation (Evershed, 2008).

With regards to site type specific residue preservation, experimental studies found inconsistent results: while Langejans (2010) found that sheltered sites such as caves favor preservation, Barton (2009) reports good survival of starch residues from open sites. Furthermore, it was demonstrated that archaeological context artefacts from open sites also showed good preservation of various residue types (Cooper and Nugent, 2009). These results indicate that a combination of several variables might be responsible for residue survival in sites. Further work is needed to investigate variables favoring residue survival in open and sheltered sites. Overall, however, residue survival in these various site types and different environmental conditions shows the wide range of potential applications for their dating.

5.2.2.2. Specific residue preservation

The susceptibility of biomolecules to structural modification and degradation is described in the order of: “lipids < carbohydrates ≈ lignin < protein < nucleotides” (Evershed, 2008:910). This order can vary according to environmental factors and the history of the artefact. One experimental study found bone, fat and woody plant tissue to be more robust residues, while blood, muscle tissue and starch were more brittle (Langejans, 2010).

5.2.2.3. Conditions that protect preserved residues

The hydrophobic nature of lipids prevents leaching and microbial decay, aiding their survival in the archaeological record (Evershed, 2008). This notion is supported by high
percentages of lipid preservation on 10,000 year old artefacts, despite previous water washing and years of storage (Mazzia and Flegenheimer, 2015).

It was also observed that preserved residues can build a protective hydrophobic barrier preventing microbial attacks (Barton, 2007, 2009; Barton and Matthews, 2006; Loy, 1987, 1993). For starch it was noted that once a "residue deposit forms" and dries "on a tool surface, a proportion of organic material is protected" and "remains shielded" to some extent "from microbial activity" (Barton, 2007:1757). This was shown even for cooked starch granules, known for their "susceptibility to decay by enzymes" that survived in tropical contexts (Barton, 2007:1759). A fast drying process that shields part of the (starch) residues from decay (Barton, 2009) might be of significance. This concept is substantiated by the discovery that water in the form of rainfall on surface stone tools did not promote decay of inherent starch residues (Barton, 2009) or phytolith residues (Fullagar, 1993). Furthermore, this could explain why residues can withstand several washes and are often resistant to extraction (Barton et al., 1998: 1233; Fullagar, 1986, 1993; Fullagar et al., 1996; Shanks et al., 2004).

Important for our purpose is the possibility that the protective conditions for preserved residues could have important repercussions for artefact cleaning and pre-treatments linked to contaminant removal. However, further research is needed to investigate the hydrophobic nature of a protective shield and under what environmental conditions this barrier is created to maintain the integrity of residues. So far the concept of a protective shield, generated by a fast drying process, is of a theoretical nature. Therefore, a systematic examination of the variables that determines what residue types are preserved and under what circumstances is required.
5.2.3 Contamination

5.2.3.1. Radiocarbon dating small (<100 μg) samples.

It is rare to find quantities of residues larger than ~1 mg on a single artefact, although larger amounts of adhesive residues can sometimes be found due to their original function, their toughness and better resistance to decay. The major problem preparing very small sized samples is the addition of contaminants in the field, during storage and in the laboratory. The amount of contaminant addition becomes increasingly significant with a decrease in sample size. A comparison of two samples, (1) having 1 mg (1000 μg) and (2) the other 10 μg sample mass, both 10 ka old (i.e. 29.8 pMC), illustrates the problem.

While 1 μg modern carbon contamination results in an age offset of only ~10 years for the 1mg C sample (1), (which is negligible considering the error in the age might be larger than this), the same amount of contamination results in an age difference of approximately 1600 years for the smaller 10 μg C sample (2). With 3 μg modern contamination, sample (1) yields an age of 9950 years and sample (2) yields an age of 6420 years. These examples highlight the sensitivity of very small samples to minute quantities of contamination.

Furthermore, the accuracy of AMS measurements decreases with sample age as the impact of contamination increases (e.g. Waterbolk, 1971:18; Bird et al., 1999; Wood, 2015). This makes dating small amounts of ancient carbon difficult (e.g. Bird et al., 2014).

The ANTARES AMS facility at the Australian Nuclear Science and Technology Organisation (ANSTO) (Fink et al., 2004) has continuously refined radiocarbon measurements on small samples containing just a few micrograms of carbon. The current
background variability achievable with the microfurnaces at ANSTO that produce the iron/graphite for AMS measurement is equivalent to 0.05 ±0.01 μg of 100 pMC material when graphitizing CO₂ (Smith et al., 2010a; Yang et al., in press). Placed in the context of ages, a 20 μg sample of infinitely old age would measure 50 ka old if this amount of modern ‘extraneous’ carbon was added. Key factors which enabled the preparation of ultra-small carbon samples were choice of an appropriate catalyst, miniaturizing the graphitization furnace (Smith et al., 2010b), the development of reliable methods of handling the graphite/iron sample, and the way of mounting the samples in the ion source target holder (Smith et al., 2010a).

5.2.3.2. Contaminant management in the AMS pre-treatment laboratory

The sample type, its stability and condition determine the chemical pre-treatment method that is employed. AMS pre-treatment laboratories have established detailed chemical and physical pretreatment protocols for various sample types and equipment, a summary of which can be found for example in Hua et al. (2001). The pre-treated samples are converted to CO₂ by combustion using the sealed-tube technique (Vandeputte et al., 1996) and graphitised as outlined in Hua et al. (2001). Moreover, procedural blanks are used to assess the mass and 14C activity of extraneous carbon is added to a sample during all stages of processing in the laboratory (Hua et al., 2004).

5.2.3.3. Case study

In an experimental study (Yates et al., 2014), we tested contamination added in the laboratory while processing ultra small residue samples extracted from stone tools.
Previously radiocarbon dated wood and peat, obtained from an environmental study were applied on replicated clean chert flakes. The stone tools were then immediately processed under laboratory conditions where contaminants from field, storage and handling were excluded. The removal methods used were ultrasonic baths and the scraping off of residues. The latter was more successful in yielding sufficient sample mass to proceed to AMS dating. Sample amounts < 22 µgC were dated. Wooden residue age determinations were in agreement with the original wood age using carbon masses as little as 10.5 and 19.76 µgC (Yates et al., 2014:598).

The three radiocarbon dated residue samples yielded relatively consistent dates. This suggests that while analyzing lithic residues, contamination added in the AMS laboratory is probably low enough to allow dating of ultra-small Holocene age residue samples. Additional work is required to demonstrate this conclusively. Peat showed greater deviations (although in some parts close to the expected date) that may have partly been caused by the heterogeneity of the material.

Future work would benefit from assessing procedural blanks in the pre-treatment protocols. This would entail assessing the mass and radiocarbon activity of extraneous carbon added during this process (e.g. as demonstrated by Santos et al., 2010) and designing the protocols to be routine whilst keeping quantities as constant as possible. Such an approach permits in-principle correction for the carbon extraneous to the sample, however, it is expensive and time consuming.
5.2.3.4. Contamination added in the field/storage environments

Contact between organic material and the artefact during excavation can introduce extraneous carbon. This includes touching finds with leather gloves or bare hands. Commonly used sealable plastic bags or cardboard boxes for storage of archaeological artefacts facilitate contamination. Of course, this also applies to the find label that commonly consists of paper or plastic.

Depending on post-depositional factors, starch, pollen and phytoliths can offset the dating towards younger or older ages when not identified as non-use related residues. Not yet identified is the age distortion effect of synthetic Camphor, a modern contaminant, possibly introduced via cosmetics or sunscreens and found incorporated in archaeological lipid residues (Buonasera, 2007). The like sponge and diatoms transferred in archaeological samples by wet sieving (Robertson, 2006) and use of plastic excavation instruments also have the potential to distort radiocarbon dates.

Long storage in a warm environment, inviting fungi and other microbe development and decay can also introduce $^{14}$C-rich CO$_2$ into the sample (Neulieb et al., 2013). This can result in age estimations that are too young. Storage in plastic bags or containers and labeling with ink or pencil graphite may also cause measurement offsets due to their in-built ages.

5.2.3.5. Contamination added in artefact handling after retrieval

Besides touching artefacts with bare hands, several treatments and examinations may lead to carbon introduction. Examples are: cleaning with tap water and brushing dirt off by using animal hair brushes; examining the artefact under a microscope after placing it on
unclean surfaces; and moving it with bare hands and leaving it exposed for long periods of time. Sketching the artefact may also introduce graphite or ink contamination.

Table 5.1 lists Fullagar’s (2006b) suggestions for residue preservation during various actions from artefact retrieval onwards.

Table 5.1: Fullagar’s sampling suggestions for residue preservation during and following fieldwork (Fullagar 2006b: 189, 191, 195) and required sampling adjustments for AMS residue dating.

<table>
<thead>
<tr>
<th>Issue</th>
<th>Fullagar’s sampling suggestions for residue preservation</th>
<th>Adjusted sampling requirements for AMS dating</th>
</tr>
</thead>
<tbody>
<tr>
<td>Artefact handling</td>
<td>Limit handling,</td>
<td>Limit handling, no contact with any organic material (cardboard, plastic, wood, leaves, ink, graphite (pencil), clothing, skin).</td>
</tr>
<tr>
<td>Artefact retrieval</td>
<td>Ideally, undertake in situ with adhering soil.</td>
<td>Undertake in situ with adhering soil, however, soil needs to be removed after residue and use-wear analysis. For example, humic acid, fulvic acids and carbonates to be removed with AAA treatment in AMS pre-treatment laboratory.</td>
</tr>
<tr>
<td>Sediment sample</td>
<td>Sample surrounding sediment</td>
<td>Only required for preceding residue analyses.</td>
</tr>
<tr>
<td>Excavator clothing</td>
<td>Wear starch free gloves</td>
<td>Wear starch free gloves.</td>
</tr>
<tr>
<td>Excavator behavior</td>
<td>Avoid food and drinks during artefact retrieval.</td>
<td>No food or drinks during artefact handling.</td>
</tr>
<tr>
<td>Wet sieving</td>
<td>Not recommended as it destroys residues and possibly adds fungus varieties.</td>
<td>Not recommended as it destroys residues and possibly adds fungus varieties.</td>
</tr>
<tr>
<td>Dry sieving</td>
<td>Optional as it causes less damage than wet sieving.</td>
<td>Not recommended because contact with other materials in sieve may add contaminants.</td>
</tr>
<tr>
<td>Excavation instruments</td>
<td>Use plastic instruments.</td>
<td>Use clean metal instruments.</td>
</tr>
<tr>
<td>Storage</td>
<td>Use plastic bags.</td>
<td>Use sealed glass or aluminum containers. To seal a container, aluminum foil or glass dish can be used. Parafilm may be used with caution. Because it may contain materials of fossil origin. Direct contact with sample should be avoided.</td>
</tr>
<tr>
<td>Cleaning soil from artefacts</td>
<td>Rub artefact gently in a plastic bag to remove thick lumps of soil or clean with a soft dry nylon brush.</td>
<td>As a plastic bag is not a preferred storage container, the rubbing method is not recommended. This method may also transfer carbon carrying contaminants (e.g. humic acid, fungus, spores) from the soil onto the artefact. A nylon brush cleaned for each individual tool is safer. Because nylon potentially contains carbon of fossil origin, Milli-Q™ water rinses should follow. Preferably only Milli-Q™ water rinses should be used.</td>
</tr>
<tr>
<td>Further cleaning soil from artefacts</td>
<td>Apply 10–20 (or more) sec sonic cleaning. Use caution with friable materials containing paint or decoration.</td>
<td>Depending on residue type and find conditions, Apply Milli-Q™ water rinses or 10–20 s sonic cleaning with Milli-Q™ water.</td>
</tr>
</tbody>
</table>
To improve the accuracy of AMS dating, we used these suggestions and adjusted them to become the requirements needed to avoid introduction of unwanted carbon.

5.2.3.6. Contamination added during sample processing prior to arrival in the AMS laboratory

The influence of starch as a potential contamination source is relevant for ultra-small carbon samples and may currently be underestimated. As organic material, starch has the potential to bias age measurements in samples. Starch granules, proven to be ubiquitously present in laboratories, enter through air conditioning and were found on laboratory equipment (Loy and Barton, 2006). A recent analysis in ancient starch laboratories and relevant consumables found that "airborne, modern starch grains landing on laboratory surfaces" (Crowther et al., 2014:96) are omnipresent. The results demonstrated starch contamination occurs in most material in the laboratory, including "powder-free gloves, microslides, parafilm, pipette tips, sodium polytungstate, paper towels, lens tissue, sample bags, centrifuge tubes, cling film, and plastic weighing trays" (Crowther et al., 2014:96). Furthermore, the majority of starch granules were found occurring in the fume hood, even after cleaning all surfaces and removing consumables. Starch granules were also found on environmental samples such as garments, wall paint, carpets, mats, shoe soles, as well as floor and ceiling tiles.

Of decontaminates tested, "only 5% NaOH, 5% KOH butane torches (on metal tools) maintaining incineration for 30 s caused complete destruction of native starch granules" (Crowther et al., 2014:100). Crowther et al. (2014:101) suggest reducing starch contamination from gloves by using brands routinely tested for starch contaminants, "by using sterilized forceps" for grasping samples, and by "using gloves that can withstand
long autoclave cycles of sterilization”. The test results further indicate that covering samples with petri dishes is preferable to parafilm, foil, or cling film, due to the abundant starch found on their surfaces. In addition, ideal laboratory conditions include HEPA air filters with controlled airflow, and restricted access (Crowther et al., 2014:103).

Ideally, the entire sample handling process for radiocarbon dating should be undertaken in a specially equipped AMS laboratory. However, the reality of archaeological research, access availability and practicality do not always allow this. For researchers who use other laboratories for sample examination (e.g. microscopic) and treatment, it is therefore useful to adopt relevant practices used in AMS laboratories. This includes standard procedures for cleaning consumables to overcome contamination in the laboratory.

To begin with, only materials that reduce the risk of contamination should be brought into contact with samples such as quartz, glass, stainless steel, and Teflon. Glass and quartz materials ideally should receive thermal treatment for 3–4 h at 450-500°C immediately before use (e.g. Brock et al., 2010; Cao et al., 2013). Sealable glass and single-use aluminum foil containers can be utilized for the procedure and for storage. To avoid the development of fungus, granular silicate in an additional aluminum or glass container can be added to the artefact. If a desiccator is used, organic sealing agents (e.g. oil or rubber-based) placed between lid and container should be avoided. Cleaning procedures for glass containers can involve Decon, followed by Milli-Q™ water rinses, while aluminum containers should be used only once.

Because of the danger of contamination by gloves and clothing, frequent replacement is required. The exposure of uncovered samples should be kept to a minimum to limit the introduction of ubiquitous starch and other airborne contaminants.
Causes for residue radiocarbon date offsets that produce ages that are too young and too old, including potential removal strategies, are listed in Table 5.2 and Table 5.3 respectively.

**Table 5.2.** Causes for residue radiocarbon dates that are too young and potential removal strategies.

<table>
<thead>
<tr>
<th>Contamination cause</th>
<th>Reference</th>
<th>Suggested removal strategy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sponge and diatoms</td>
<td>Robertson, 2006; Cooper and Nugent, 2009:209;</td>
<td>Apply acid wash followed by Milli-Q-water rinses (Yates et al. – current paper)</td>
</tr>
<tr>
<td>Starch, non-use related</td>
<td>Crowther et al., 2014; Loy and Barton, 2006</td>
<td>For equipment: Apply 5% NaOH and 5% KOH (Crowther et al., 2014). On samples: apply alkali step of Acid Alkali Acid Method (AAA) treatment (Yates et al. – current paper).</td>
</tr>
<tr>
<td>Touch with bare hands</td>
<td>Yates et al. – current paper</td>
<td>Apply acetone wash.</td>
</tr>
<tr>
<td>Loose soil</td>
<td>Yates et al. – current paper</td>
<td>Apply Milli-Q™ water rinses.</td>
</tr>
<tr>
<td>Radio tracers</td>
<td>Yates et al., 2015</td>
<td>Avoid contact with medical or industrial radio tracers.</td>
</tr>
<tr>
<td>Coprolites or manure</td>
<td>Reber and Hart, 2008</td>
<td>Apply Decon90 (2-5% diluted immersion) and Milli-Q™ water rinses (Yates et al. – current paper).</td>
</tr>
<tr>
<td>Insect remains</td>
<td>e.g. Cooper and Nugent, 2009:217</td>
<td>Apply an Acid wash, followed by Milli-Q™ water rinses (Yates et al. – current paper).</td>
</tr>
<tr>
<td>Post-depositional related</td>
<td>Wadley and Lombard, 2007; Langejans, 2011</td>
<td>Apply Acid Alkali Acid Method (AAA) followed by Milli-Q™ water rinses (Yates et al. – current paper).</td>
</tr>
<tr>
<td>soil components</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Paper/cardboard</td>
<td>Yates et al. – current paper</td>
<td>Apply Milli-Q™ water rinses.</td>
</tr>
<tr>
<td>Fungus</td>
<td>Neulieb et al., 2013</td>
<td>Apply Acid wash followed by Milli-Q™ water rinses (according to Yates et al. – current paper).</td>
</tr>
</tbody>
</table>

5.2.3.7. *In-built ages affecting radiocarbon determinations*

Age overestimations caused by water can occur when older carbon is incorporated through organisms living in a marine environment — the radiocarbon marine reservoir effect — or where old carbon was introduced, e.g. by waters passing through or over limestone (containing ‘dead C’), or possibly by vegetation growing near old groundwater or volcanic emissions. These effects have been linked with radiocarbon dating artefacts associated with bog bodies (van der Plicht et al., 2004:472, 473), pottery cooking residues containing older carbon incorporated in bones and shells from freshwater fish and mollusks (e.g. Fischer and Heinemeier, 2003; Roper, 2013, 2014; Hart and Lovis, 2007, 2014) and pollen
(e.g. Long et al., 1992). (Table 5.3). Radiocarbon age excesses also occur when older material, such as fossil bitumen, was used as embalming or possibly as a hafting agent (Aufderheide et al., 2004; Yates et al., 2015). Recent research also points to the occurrence of radiocarbon date overestimations when modern petroleum-based paint was used in Australian rock art (McDonald et al., 2014).

Table 5.3. Causes for residue radiocarbon dates that are too old and potential removal strategies.

<table>
<thead>
<tr>
<th>Contamination Cause</th>
<th>Reference</th>
<th>Suggested removal strategy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calcite precipitation</td>
<td>Long et al., 1992</td>
<td>Apply Acid Alkali Acid Method (AAA) or only acid treatment (2M HCl) (Yates et al. – current paper).</td>
</tr>
<tr>
<td>Carbonate influence</td>
<td>Neulieb et al., 2013; Li et al., 2014</td>
<td>Apply cold 10% HCl (Neulieb et al., 2013), then apply 10% HCl at 80˚C for 30 min (Li et al., 2014).</td>
</tr>
<tr>
<td>Carbon introduced by water (reservoir effect)</td>
<td>Fischer and Heinemeier, 2003; Long et al., 1992; van der Plicht et al., 2004:472, 473.</td>
<td>Apply acid steps of AAA to remove any carbon deposited by groundwater. However, removal is not possible if incorporated in fish bones, shells and molluscs (Yates et al. – current paper).</td>
</tr>
<tr>
<td>Graphite from pencil</td>
<td>Yates et al., 2015</td>
<td>Apply Decon90 (2-5% diluted immersion) followed by Milli-Q™ water rinses.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>In-built-ages causing age offsets</th>
<th>Reference</th>
<th>Suggested removal strategy or action (Yates et al. – current paper).</th>
</tr>
</thead>
<tbody>
<tr>
<td>Petroleum based products (carbide or kerosene) incorporated in (modern) paint mixture for rock art</td>
<td>McDonald et al., 2014</td>
<td>Removal might not be possible because the material might be incorporated in the residue. Suggest chemical identification of residue to assess reason for age deviation.</td>
</tr>
<tr>
<td>Bitumen</td>
<td>Aufderheide et al., 2004; Reber and Hart 2008:135; Yates et al., 2015</td>
<td>As bitumen can be mixed into a hafting adhesive used to seal pottery or can be part of an embalming ingredient, use chemical identification of residue to assess reason for age deviation.</td>
</tr>
<tr>
<td>Fossil shell from rock type, e.g. flint, used to produce stone tool</td>
<td>Yates et al., 2015</td>
<td>Apply acid steps of AAA treatment to remove shell from sample and identify by scanning electron microscope.</td>
</tr>
<tr>
<td>Plasticizers incorporated in the sample</td>
<td>Baumer and Dietemann, 2008; Hedges et al., 1992:911; Hyman and Rowe, 1997:64; Koller et al., 2001</td>
<td>Removal strategy is not known because identification so far is only by sample-destructive GC/MS. Therefore, avoid storage in plastic containers.</td>
</tr>
</tbody>
</table>
5.2.4 Residue type, sample size and yielded carbon mass

For the purposes of residue radiocarbon dating, it is important to understand the relationship between sample type, sample size and yielded carbon mass dated. Table 5.4 gives an overview of previous studies including research in which food residues from ceramics and organic material from rock art were AMS dated. These radiocarbon dates can be correlated with dated stratigraphy, typological chronological markers or rock art linked to linguistic correlates in order to validate those dates.

5.2.4.1. Sample size and yielded carbon mass

The sample size required in terms of yielded micrograms of carbon (µgC) is set by the AMS laboratory. For ANSTO, this is 5 µgC (e.g. Smith et al., 2007, 2010a, 2010b; Yang et al., 2013).

The starting sample weight is dependent on the age of the material and on its purity, for example, the extent to which the sample is intermixed with other non use-related materials including inorganic substances, sand, carbonates, humic acids and rootlets. Also, the carbon content of a material is relevant. For instance, charcoal contains generally between 50% and 60% carbon, while marine shell has 12% carbon and bone carbon content is dependent on diagenetic or biogeochemical factors and can vary between 1% and 30% (Taylor et al., 2014:117). Therefore, the actual weight of the original sample can range "between 4 to 10 times that of the final amount of graphitic carbon used for the measurement itself" (Taylor et al., 2014:117).
Table 5.4. Overview of AMS radiocarbon dated archaeological residues (*from lithics, rock art, and food residues from ceramics) and correlation of residue dates with residue quantity microgram of Carbon (µgC) (if available) and associated age control.

<table>
<thead>
<tr>
<th>Residue type*</th>
<th>Reference</th>
<th>Quantity dated</th>
<th>Notes on residue dates</th>
</tr>
</thead>
<tbody>
<tr>
<td>blood</td>
<td>Nelson et al., 1986; Loy, 1987, 1993</td>
<td>between 50 µgC and 1 mg carbon mass</td>
<td>Two residue samples dated from two lithics with correlating age obtained from a hearth and associated typology.</td>
</tr>
<tr>
<td>wood</td>
<td>Yates et al., 2014</td>
<td>10.5 µgC</td>
<td>By testing contaminant addition in the laboratory, a high correlation between residue and original material was obtained.</td>
</tr>
<tr>
<td>Wood</td>
<td>Yates et al., 2015</td>
<td>18.97 µgC from wood residue</td>
<td>Wood residue with an age of 7890±180 BP dated ~1000 years younger than the associated stratigraphic date.</td>
</tr>
<tr>
<td>Charcoal (organic residue from rock art paint)</td>
<td>McDonald et al., 2014</td>
<td>Samples containing ≤10 µgC were considered to have insufficient Carbon mass to be measured. Small µgC measured samples contained a minimum of 30 and &lt;10 µgC, while samples ≥50 µgC had between 50 and 240 µgC.</td>
<td>The plasma oxidation method together with AMS radiocarbon measurement was used as a direct technique for dating both charcoal and inorganic-pigmented pictographs. Age measurements were in agreement with the age frame of the rock art style.</td>
</tr>
<tr>
<td>Adhesive material</td>
<td>Yates et al., 2015</td>
<td>33.30 µgC and 44.38 µgC</td>
<td>The samples were dated ~2 ka BP, and the acid treated sample was dated slightly younger. Obtained dates were in agreement with the expected Late Holocene time frame indicated by associated typology.</td>
</tr>
<tr>
<td>Resin</td>
<td>Lampert et al., 2002</td>
<td>Not indicated</td>
<td>Two resin samples of one potsherd from Spirit Cave date too young compared with dates achieved in stratigraphy. Further, two resin samples from one potsherd dated from the Noen U-Loke site were dated in alignment with related charcoal dates.</td>
</tr>
<tr>
<td>Nut</td>
<td>Lentfer et al., 2013</td>
<td>Sample size not discussed</td>
<td>The authors report that the achieved radiocarbon age is in accordance with dates obtained from the find context.</td>
</tr>
<tr>
<td>Cooking residues from bones, shells, freshwater fish and mollusks, lipids, tree resin, plant and meat processing</td>
<td>Fischer and Heinemeier, 2003; Roper, 2013; Reber and Hart, 2008</td>
<td>Not indicated</td>
<td>Reber and Hart (2008) found that lipids make up the large majority of cooking residues. Among which visible residues comprise a wider range of compounds but less yield compared with residues which were absorbed in ceramic pot walls. Dates reflect long period of pottery use in the region.</td>
</tr>
<tr>
<td>Lipids from archaeological ceramic cooking pots</td>
<td>Stott et al., 2001</td>
<td>Material containing &gt;300 µgC of extractable lipid was selected for this study. Typically, samples of &gt;200 µg were needed to achieve precisions of ±50 years or better.</td>
<td>Absorbed material from a burial environment with not-use related contamination is suspected to be linked to dates that are 100–150 yrs too young.</td>
</tr>
<tr>
<td>Starch</td>
<td>Zarillo et al., 2008</td>
<td>Not indicated</td>
<td>Starch residues were separated from cooking residues inherent to pot sherd or grinding stones. Obtained radiocarbon dates were in agreement with expected ages.</td>
</tr>
<tr>
<td>Beeswax</td>
<td>Pierluigi et al., 2014</td>
<td>Not indicated</td>
<td>The substance was radiocarbon dated from rock art pigments.</td>
</tr>
</tbody>
</table>
5.2.4.2. Residue type

AMS dating of residues has been successful on nut, adhesives, wood, charcoal, blood, lipid, starch and beeswax extracted from lithics, ceramic and rock art (examples are presented in Table 4). Information relevant for AMS dating for these residue types is presented below.

Nut: As a yearly crop, nuts represent an excellent sample for AMS dating. They appear to be a resistant material, allowing harsher pre-treatment including 30% HCl solution (Lentfer et al., 2013).

Adhesives: Adhesive residues, e.g. resins, also consist of a “tough” material and are resistant to decay. Various adhesive types were found on artefacts and are often interpreted as remnants of hafting mastic or sealing agent. Dating adhesive residues can be difficult due to their heterogeneity, e.g. they can have up to 80% of their weight made up from filler material (Dickson, 1981:164) and this can result in age distortions.

Wood and charcoal: Wooden residues were found preserved under various environmental conditions. In an ideal situation, wooden residue will be identified as use-related residue, and then removal and treatment can follow. Alkali Acid Alkali (AAA) treatment prevents humic acid influence on AMS dates, but more research must be undertaken to determine whether fibers (wooden, plant, or rootlets fibers – assessed as contaminants) should be removed from the sample as a routine pre-treatment step for AMS samples. A similar concern was raised by McDonald et al. (2014) for charcoal residues from rock art. Thorough sample description with microscopic residue analyses is therefore a crucial step prior to treatment and dating. For example, if the microscopic analysis shows that the cleaning treatment has eliminated post-depositional contaminants, removal of remaining
fiber from the sample may not be indicated since it might be part of the residue and could increase the sample mass and thus improve the chance of dating the residue.

Blood: We know of only one example where blood residues extracted from lithics were AMS dated. Between 50 µgC and 1 mg carbon mass was required to achieve dates which correlated with dates obtained from a hearth (Nelson et al., 1986; Loy, 1987, 1993). While in general significantly smaller blood residues are found on lithic tools, their survival, identification and removal were also demonstrated on ceramics (Matheson et al., 2009). Biochemical methods in conjunction with microscopic analyses may increase detection of residue amounts present on an artefact. Possible carbon introduction by these methods therefore needs to be evaluated prior to application on artefact surfaces. Water was found to be most successful in removing blood from stone tools (Matheson and Veal, 2014:236). The use of Milli-Q™ water would also be indicated in relation to blood residue dating.

Lipids: More than a decade ago, 200 µgC were required to radiocarbon date specific compounds from the lipid material surviving in archaeological cooking pots (Stott et al., 2003). We are not aware of research that has examined the amount of lipids inherent on stone artefacts. However, recent research has demonstrated that the vast majority of a stone tool sample contained lipids which had been trapped in the rock’s pores and cracks (Mazzia and Flegenheimer, 2015). This is significant as it illustrates the potential of dating lipids from stone tools.

On the other hand, there are indications that lipids are more susceptible to contamination than other residues. For example, contamination from the burial environment was linked to lipid dates 100–150 years younger than the anticipated age (Stott et al., 2001). This notion was supported by the discovery of plasticizers (Hedges et al., 1992:911) and synthetic
Camphor introduced via cosmetics or sunscreen (Buonasera, 2007), both of which were found in archaeological lipid residues.

Starch: Starch residues from cooking remains have been isolated from residues adhering to pot sherds and grinding stones using heavy-density liquid separation after oxidation treatment. The results provided the earliest direct dates for maize in Early Formative Ecuadorian sites (Zarillo, 2008).

Beeswax: This substance was added to hafting fixatives (Regert et al., 2003) and was radiocarbon dated from rock art pigments (e.g. Pierluigi et al., 2014).

From this list we conclude that wood and nut residues are most suitable for dating because of their homogeneity and good preservation. Adhesive residues are equally suitable, however, chemical identification of their ingredients would help to validate the resultant dates. The possibility of dating molecules such as lipids from resin by GC-MS could be further developed. To do this, particular carbon transfer from chemicals used in the sample needs to be examined. The applicability to radiocarbon dating the remaining residues from lithic artefacts requires detailed testing, e.g. to ensure sufficient matter is present. This present study also illustrates the need for more transparency in the sample sizes used for residue dating.

The feasibility of radiocarbon dating wooden and adhesive residues from archaeological stone tools is described in the following case study. The examined stone artefacts provided reference ages. The ages of previously dated reference samples, ranging from 9000 to 12,000 years BP led us to expect reasonable carbon content in the residue samples.
5.2.4.3. Case study 2

As the first case study (Yates et al., 2014) indicated, ultra-small sample processing and AMS dating in the laboratory are feasible on wooden residues when contaminants are contained. In the second case study, this technique was applied to artefacts from real archeological contexts. Dated stone tools from three different sites (Yates et al., 2015) were correlated with our residue AMS measurements. Among them were five artefacts retrieved from a bog site which showed excellent residue preservation. Microscopic observations substantiated previous interpretations (Pawlik, 2011a) of wooden and adhesive residue present on the bog site tools. Residues from two further stone tools, found elsewhere, were also interpreted as putative adhesive material. Typical diagnostic criteria for adhesive residues such as droplet appearance and mud crack structure were also observed. In addition, inherent contaminants from handling, such as fabric fibre, skin scales and graphite from sketching the artefact, were identified on all artefacts. Diluted Decon immersions (followed by Milli-Q™ water rinses) were trialed as a cleaning technique to remove these contaminants after first testing the solution experimentally.

Further results showed that, in some cases, causes for radiocarbon age offsets may not be microscopically detectable. A previous GC/MS analysis on more abundant deposits from bone points had shown that birch bark tar was used on the bog site. All four examined samples showed plasticizers (DBP) from bag storage incorporated into the birch bark tar (Figure 5.1) (Baumer and Dietemann, 2008). This could only be detected by GC/MS analyses. It is significant that earlier research described plasticizers absorbed in samples as responsible for age overestimations (Hedges et al., 1992:911; Hyman and Rowe, 1997:64; Koller et al., 2001).
These results suggest, therefore, that plasticizers could cause age overestimations obtained from two lithic adhesive residues in this study. Furthermore, the AMS date offsets could have been caused by the intermix with or use of bitumen. A SEM-EDX analysis alone was not able to identify bitumen. However, the technique revealed the presence of micro shell, likely derived from the flint stone, in the residue matrix. Fossil shell is a known component of flint stone and could have also caused the age overestimations. The elemental composition of residues on a stone tool from another site points to Manganese deposits rather than the anticipated adhesive residue.

These examples illustrate the importance of additional chemical identification during microscopic interpretation and that this is particularly desirable for adhesive residues.

**Figure 5.1.** Gas-Chromatogram from birch bark pitch of Friesack, find 1977:7/P4. Betulin, lupeol and lupenone, biomolecular markers of birch bark. Additionally, plasticizers in the form of Phtalatestern, Dibutylphthalat (DBP) (Courtesy of Baumer and Dietemann 2008, Doerner Institute, Munich).
Two adhesive residues from one stone tool, which contained 33.30 μgC and 44.38 μgC, could be dated within an expected Late Holocene time frame. Age deviations of wooden residues (7890 ±180 BP instead of dating between ~9000 and ~9250 years BP) were found on material containing 18.97 μgC (Table 5.4). The deviation from the stratigraphical age measurement may still represent the authentic age if we assume that taphonomic processes could have caused upward tool movement. However, post-depositional root growth through the cavity of the stone tool or modern contaminant introduction cannot be excluded. Nonetheless, the results allow the inference that dating these small samples is possible. This is especially significant considering all these artefacts were considerably contaminated.

The lithic residues we examined had been contaminated following their retrieval but before preparation for AMS. This is probably true to some extent for the majority of lithics with residues that have been found to date.

These considerations highlight the extra care required when radiocarbon dating residues. In the following section, we look at the different stages and situations during the treatment of archaeological artefacts where carbon transfer into samples and consequential age offsets can occur.

5.3. Discussion

5.3.1. Implications for retrieving, handling and storing archaeological objects

From the above review and the two case studies presented, we suggest the following adjustments in the treatment of archaeological artefacts destined to be residue dated.
As demonstrated, some site types and environmental conditions preserve residues on lithic artefacts. If a site shows indications of good residue preservation (e.g. waterlogged or desiccated), archaeological field techniques, storage and further handling can be adjusted to limit contaminant transfer. We recommend that AMS residue dating should preferably be undertaken only when it is linked to a research question that justifies the removal and subsequent destruction of the residues that occurs during that process. For instance, a research project involving use-wear and residue analyses, e.g. resolving questions of tool function or the identification of plants, could be combined with residue dating. Furthermore, for an overall picture of residue preservation in an assemblage, it would be valuable to document the ratio of residue and use-wear-analyzed artefacts and those that were not examined.

A residue analysis would show whether tools contain residue amounts sufficient for dating. If ample residues are present, some should be left on the tool for future research and potential replication of dates, whilst the remainder could be removed for dating.

At this stage wood, nut and adhesives (resin, tar or gum) may be most promising for residue dating because, as discussed above, they are often better preserved. Additional chemical identification would be beneficial, especially for adhesives, to avoid age biases caused by mixed-in ingredients. Other residue types can be trialed for dating, provided enough material is present.

Analyses involving residue and use-wear documentation is time consuming and consideration needs to be given to the available time and funding resources. Further sample processing, including extraction and treatment for residue dating, requires planning.
and time commitment. These analytical steps should preferably be conducted by researchers with experience in the field.

Once these parameters are clarified, adjustments in the treatment process of archaeological artefacts that are beneficial for residue dating can be summarized as in our preliminary sampling protocol suggestions (Table 5.5).

5.3.1.1. Fieldwork and storage:

When an artefact is found in the sediment, it is desirable that it is touched and retrieved by wearing starch-free gloves or by placing it with adhering soil via a metal excavation instrument into a container. Ideally, sampling the sediment surrounding the artefact will facilitate residue identification analysis. Storing artefacts in sealable, clean aluminum or glass containers (examples are shown in Table 5.5) avoid carbon transfer. Labels should not come in contact with the sample but be attached outside of the sample storage container. To avoid the development of fungus, granular silicate in an extra aluminum or glass container can be added to the artefact. If practicable and field conditions allow, sample freezing is a satisfactory interim option until it is possible to freeze-dry in a laboratory.

5.3.1.2. Cleaning and preparing

Careful brushing with clean nylon brushes to remove dirt from each individual artefact may be sufficient in some cases. Further cleaning can be carried out, depending on residue type and find conditions. We suggest Milli-Q™ rinses (Q-pod if available), and/or 10–20 s sonic cleaning, followed by oven drying in a glass container followed by storage in a
sealed aluminum or glass container until further examination. It is important to use glassware that has been thoroughly cleaned, acid washed and baked. An acetone wash may be practical to remove lipids that are present from handling.

5.3.1.3. Microscopic examination

To reduce the risk of carbon transfer, the artefact should be placed on an appropriate tray (glass, quartz or aluminum) when monitored under a microscope. Moving and turning around the artefact could be conducted by using clean tweezers or it can be held in place with clean needles. The instruments can be placed on a clean glass dish when not used. The artefact should be placed back in a sealed glass or aluminum container as soon as the examination is finalized.

5.3.1.4. Chemical identification

If accessible, Raman Spectroscopy and/or FTIR could be carried out after microscopic identification, e.g. for the identification of adhesive materials such as resins. Depending on the range of the measuring equipment, residue extraction needs to be carried out. Analyses can be carried out in situ, in reflection mode with a microscope attachment. In this case, it might be advisable to prior-test the intensity of the beam to avoid damaging the residue and to check potential contamination introduction when applying pressure during measurement. Sample measurements in transmission mode on a bench or salt disc require residue removal from the tool. If ample residues are present on an artefact, remaining parts not used for AMS dating can be examined with SEM-EDX to enhance residue interpretation.
Table 5.5. Sampling protocol suggestions to avoid contaminant introduction prior to AMS radiocarbon dating.

<table>
<thead>
<tr>
<th>Action</th>
<th>Carbon Transfer Contaminants - X</th>
<th>Carbon Transfer Avoidance Strategy - ✓</th>
</tr>
</thead>
<tbody>
<tr>
<td>Excavating</td>
<td>Plastic instruments</td>
<td>Use metal instruments</td>
</tr>
<tr>
<td>Retrieving artefact</td>
<td>Bare hands, leather gloved hands</td>
<td>Use metal instruments, e.g. trowel, tweezers, starch free gloves.</td>
</tr>
<tr>
<td>General Handling</td>
<td>Bare Hands</td>
<td>Touch artefact only with starch free gloves or metal tweezers.</td>
</tr>
<tr>
<td>Storing</td>
<td>Plastic or cardboard containers.</td>
<td>Use sealed glass or aluminum containers.</td>
</tr>
<tr>
<td>Fungus avoidance</td>
<td>Open plastic bags (potential contamination with ubiquitous starch).</td>
<td>Place loose silica gel or silica in a sachet in a separate container to artefact or use a dessicator.</td>
</tr>
<tr>
<td>Labeling container</td>
<td>Paper or plastic label to the artefacts' container</td>
<td>Place label outside the artefact’s container.</td>
</tr>
<tr>
<td>Labeling artefact</td>
<td>Labels with pencil and/or ink annotations.</td>
<td>Do not label (!) until examined, and residues have been removed and AMS dated.</td>
</tr>
<tr>
<td>Microscope examination</td>
<td>Artefacts placed on plastic surfaces, use of bare hands to move samples.</td>
<td>Place artefact on a clean glass surface or quartz slide and turn or move with tweezers and needles. Place instruments on clean glass dish when not used.</td>
</tr>
<tr>
<td>Sketch artefact</td>
<td>Artefacts sketched with pencil or pen (graphite or ink contamination).</td>
<td>Do not sketch until microscope examined, documented, residues removed and AMS dated.</td>
</tr>
</tbody>
</table>
5.3.1.5. Extraction/removal

The residue type determines the kind of removal. Detailed descriptions of extraction methods are given by Fullagar (2006b). To avoid extraneous carbon transfer into the sample, the use of fresh unused scalpel blades is required to scrape off residues. Pipette extraction needs to be carried out with Milli-Q™ water and stored in clean sealed vials for further processing. If sonication is chosen, it may be advisable to use glass or aluminum floating boats instead of their plastic counterparts.

The case studies presented above have illustrated the relevance of sample storage to age determination by demonstrating the effect of contamination introduction into samples, such as plasticizers. Geochemical studies have demonstrated the relative short time period in which plasticizers migrate into samples (Grosjean and Logan, 2007), even into the interior wall of stones (Brocks et al., 2008). While procedural blanks are crucial to assess contamination occurring during sample processing in laboratories, they are not able to detect contamination during fieldwork and storage. This highlights the necessity to minimise and quantify contaminants that occur during excavation, collection, storage and examination. It also points to the desirability of undertaking age determinations from multiple artefacts, from one archaeological context, when they are available and funding permits. Finally, consideration needs to be given as to whether dating the sample is feasible if the sample is small.
5.4. Conclusion

This review was carried out to facilitate the accuracy of future lithic residue radiocarbon dating through AMS. After reviewing the occurrence and impact of contaminants on radiocarbon dates, it has been possible to develop sampling strategies to lessen their effect.

Reasons for age measurement offsets have been highlighted in this revision. Several studies suggest sample contamination from fungal, microbial or bacterial activity when samples were stored in a warm climate or in plastic bags. It was also demonstrated that processes during fieldwork and storage contributed notably to sample contamination. Laboratories, often unknowingly, also contain starch contaminants. These various sources of contamination may be responsible for, or contribute to, radiocarbon age offsets in ultra-small samples. More precautions are therefore required to avoid introducing these materials into archaeological residue samples. Such contaminations can easily cause significant radiocarbon measurement offsets as it can be difficult to detect them and it is still uncertain as to whether they can be isolated and removed.

The interrelation and complexity of various variables associated with residue dating have become more apparent. While we have no control over post-depositional or burial factors such as plants (e.g. from root growth) or animals (e.g. insect remains) influencing samples, we can avoid or mitigate the remaining four aspects that contribute in sample contamination (Figure 5.2). This has prompted the development of a preliminary sampling protocol for artefacts considered promising for residue dating. Clearly, the scope of the particular research goal and the reality of fieldwork may, in the majority of cases, only allow a small percentage of artefacts to be sampled from beginning to end according to those suggestions. Also, the requirement for sufficient residues to be present in an artefact,
limits the method’s application. Our proposals are at the early development stage and will be refined and improved in the near future. Hopefully, this review will already limit common contamination and facilitate better artefact sampling and handling during field and laboratory work.

Figure 5.2. Stages of potential non-use related contaminants transfer on archaeological artefacts.

We would also like to propose further improvements to achieve better dates in the future. First, it would be desirable to routinely include in publications an accurate measurement of the carbon mass used for AMS dating. Second, it is suggested that more examination is essential into the development and occurrence of fungi, microbes and bacteria after artefact retrieval and during storage. Finally, we suggest further research needs to be conducted into: (i) the conditions favoring the development of a protective shield around residues, (ii) differential residue type preservation and their suitability for radiocarbon dating, and (iii) residue preservation and inherent contaminants from surface and buried sites artefacts. This would greatly improve the accuracy of residue dating.
5.5. Acknowledgments

We express our appreciation to Ursula Baumer and Dr Patrick Dietemann, Doerner Institute, Munich, Germany for generously generating and providing GC Chromatograms to be published in this paper and for sending their reports on GC/MS analyses of birch tar from Friesack. Many thanks to my Southern Cross University supervisors: A/Prof Anja Scheffers for encouragement to write this paper and Dr Renaud Joannes-Boyau for earlier comments on the manuscript. We owe thanks to Dr Virginia Finch, formerly Principle Research Scientist (CSIRO, Rockhampton), for comments on earlier drafts of this paper. Thanks also to Angela Rosenstein and Satyaa Susanne Lohmann for their comments. Thanks to three anonymous reviewers whose comments contributed substantially to the quality of this paper.
CHAPTER 6

Synthesis and Conclusion
6.1. Synthesis

This study aimed to provide new knowledge on AMS radiocarbon dating of plant residues extracted from archaeological stone tools. Radiocarbon dating of plant residues was first investigated experimentally followed by an attempt on archaeological samples.

Inspired by previous work on direct dating of blood residues (Loy, 1987; Nelson et al., 1986), and working from the notion that "In many respects the artefact itself becomes a site from which we extract a molecular 'artefact'" (Loy, 1993:47), an expanded model was developed for dating plant residues from stone tools. A new contribution to this research direction was achieved by following specific research steps.

Early on, the research question focused on laboratory contamination and whether it can be controlled to such a degree that small sized residue samples extracted from stone tools could be radiocarbon dated accurately. By using replicated, clean stone tools as well as wood and peat of a known age, specific contamination sources (e.g. from field, storage, curation) were excluded. Concurrently, it also allowed for the isolation of potential laboratory contamination. The results showed that laboratory contamination had no significant impact on the wooden residue measurements. For peat, deviation in ages seemed to be related to non-homogeneity of the material. These preliminary results suggest that careful laboratory procedures do not lead to significant contamination able to distort residue measurements. This knowledge was crucial to establish in order to date extremely small samples such as use-related plant remains from lithic tools. Any laboratory pre-treatment procedure that introduced considerable contamination would have had to be
discarded, obviously. Furthermore, this initial experimental work validated the feasibility to date wooden samples containing as little as 10.5 μgC. Results showed that contaminants could be confined, allowing the study to move on to archaeological samples.

Subsequent radiocarbon dating of wooden and adhesive residues from stone tools from archaeological contexts confirmed the applicability of the method. For the first time, AMS radiocarbon dates were obtained from a wooden residue attached to a Mesolithic core axe, with only 18.97 μgC. An offset from the reference age could be interpreted as upward movement of the artefact through the sediment layers (bioturbation, taphonomy), but one cannot exclude the possibility of contamination. Nonetheless, the two adhesive residues, both extracted from the same stone tool (33.30 μgC and 44.38 μgC), yielded overlapping dates matching the expected Late Holocene age of the artefact.

A careful assessment of residues’ age offsets led to a better understanding of contamination provenance and impact. The age difference in adhesive residues appeared to be linked to bitumen intermix, fossil micro shell (from the flint stone matrix) or the incorporation of plasticizers (most likely from plastic bag storage). The use of GC/MS and SEM-EDX analyses proved to be helpful in detecting plasticisers and fossil shell, respectively, as potential sources for sample age offsets. Notably, the in-built causes for age offsets in these samples could not have been prevented by laboratory sample pre-treatment. This highlights the importance of chemical residue identification methods, especially for adhesive residues, at the beginning of the analysis. An understanding that is further substantiated by the SEM-EDX analysis is the identification of manganese dendrites as the actual substance rather than putative adhesive residue. Overall, the applied methodological sequence was appropriate and led to successful outcomes. Future work would ideally further evaluate non-destructive techniques such as vibrational spectroscopic
methods, staining solutions and chemiluminescence and their suitability for non-contaminant transferring residue identification. This would aid in guiding the choice of residue treatment steps.

Previously unrecognized causes for archaeological sample age offsets have been identified as related to radiotracers used in medicine and industry. These substances are suspected to severely contaminate samples, as was witnessed on a sample with a modern carbon value of 338.9% and an age of 10,000 years into the future. This unreported contamination represents a new consideration for future sample handling.

For other microscopically identified contaminants (fabric fibre, skin scales and loose pencil graphite), removal with 2% diluted Decon 90 (followed by Milli-Q™ water rinses) was successful. Nonetheless, further unidentified causes for age underestimation are likely related to curation contamination or transfer of contamination (modern carbon source) into the sample. Further research is needed to isolate all potential contamination sources. Nevertheless, one of the main results of this research is the demonstrated ability to manage contaminants from archaeological residues as well as the feasibility of AMS dating of extremely small wooden and adhesive residues. One has to keep in mind that these results are remarkable considering that all examined archaeological stone artefacts were heavily contaminated. The experience and results obtained from this study show the importance and impact of contaminant transfer that can occur during sample storage, handling and curation.

Following on, key issues related to residue radiocarbon dating of very small sample sizes were investigated. Examining the interconnection between residue identification, preservation and contamination, as well as residue type, enhanced understanding around
dating capabilities. The overall results were synthesised into new sampling strategies for all stages of artefact treatment. Protocols were developed to provide easy-to-follow guidelines for artefact retrieval, handling, storage, analysis and extraction in order to minimise contaminant transfer. While these strategies will certainly be refined in the future, they currently support researchers in making appropriate choices to enhance residue dating accuracy.

6.2. General conclusions

This study has contributed new knowledge to the field of archaeological sciences by means of accurate AMS radiocarbon dating of micro-sized residues. The capability of the method was tested and demonstrated through both a purely experimental approach and an applied analysis. In particular, this study emphasises the good age agreements obtained between wooden residues and the reference age.

New insights gained into contamination have proven to be the key to understand age offsets and should be further investigated. Nevertheless, the results of this study have implications for archaeological residue treatment and artefact sampling when residue dating is the aim of a project. While insufficient prior explorations into contaminant prevention and cleaning methods have thus far hampered dating capabilities, this research provides new guidelines to improve accuracy and dating strategies.
6.3. Limitations

To some extent the small size of the experimental sample (n=10) and the archaeological sample (n=7) might weaken the global confidence in the techniques. One has to keep in mind that the main limitation of this problem was the cost and access to AMS facilities. Yet, the residues’ examination yielded innovative results and contributed clearly to new knowledge. New research directions were revealed, opening new chronological application for archaeological objects and this value is also enhanced by new sampling strategies as elaborated in this work.

As discussed initially (Chapter 1), it was challenging to obtain a) artefacts that contained considerable preserved residue amounts and b) artefacts that concurrently have a documented and confident reference age. The limited amounts of stone tools available as well as the lack of a sufficient, previously established method, justifies trial of the technique first on a smaller number of archaeological artefacts.

It also needs to be considered that there were no other examples in the archaeological record where as many stone tool residues were radiocarbon dated as in the present study. As a comparison, in an initial attempt to radiocarbon date residues from stone tools only two blood residue samples were dated (Loy, 1987, 1993; Nelson et al., 1986). Furthermore, this study is unique in presenting the results of radiocarbon dating wooden and adhesive residues in the microgram carbon scale while simultaneously addressing contamination as well as integrating additional identification methods.
6.4. Future work

AMS radiocarbon dating of archaeological residues requires that thorough residue and use wear analyses precede the process. If the latter method boasts exceptionally good residue preservation, the value of additional research into radiocarbon dating some of these residues could be signified.

In general, future research could follow two major paths:

1) to radiocarbon date residues from already retrieved stone artefacts; a prerequisite here would be to further examine and explore adequate cleaning methods to remove contamination.

2) to radiocarbon date residues from newly retrieved artefacts, which were sampled according to the suggested guidelines proposed in the present study.

Either way, it would be instructive to continue within a Holocene time frame. These artefacts would simultaneously be retrieved from site types where residue preservation is most likely to occur. The former suggestion promises greater carbon contents in the residues compared with Pleistocene samples; the latter may provide an above average amount of preserved residue. In general, site types favouring residue preservation include bog and wetland sites, caves, sites in proximity to river streams or lakes and sites from cold climates, e.g. with permafrost (for instance, some Canadian sites), as well as sites from arid conditions including open air sites and sites linked to clay rich soils.

Theoretically, all organic residue types have the potential to be radiocarbon dated as long as they contain a sufficient amount of carbon. Nonetheless, it is suggested to continue with
homogenous residue types, as heterogeneous residues open another level of complexity. Wooden residues would represent the ideal candidates as shown in this study.

6.4.1. Further recommendations

1. Link between stone raw material and residue preservation as well as particular requirements for radiocarbon dating pre-treatment.

2. Contaminant development and prevention, especially fungi development after artefact retrieval.

3. General preservation conditions, in particular regarding the conditions that develop a protective shield around residues. This pathway could include examining how patination on stone tools acts as a preservation condition to protect residues from decay and how these organic remnants can be removed.

4. Residue preservation and the comparison between inherent contaminants from surface stone tools and buried sites’ artefacts.

5. Development of residue-specific protocols for non-destructive and non-contaminant transferring characterisation methods (e.g. vibrational spectroscopic techniques or staining solutions).
References


REFERENCES


REFERENCES


REFERENCES


REFERENCES


REFERENCES


REFERENCES


REFERENCES


REFERENCES

and Spätboreals sowie des älteren Atlantikums. Veröffentlichungen zur brandenburgischen Landesarchäologie 43/44, 7–84.


REFERENCES


REFERENCES


REFERENCES


Jones, P.J., 2009. A microstratigraphic investigation into the longevity of archaeological residues, Sterkfontein, South Africa. In: Haslam, M., Robertson, G., Crowther, A.,


REFERENCES


REFERENCES


REFERENCES


REFERENCES


REFERENCES


REFERENCES


REFERENCES


REFERENCES


REFERENCES


REFERENCES


REFERENCES


APPENDIX 1

Author contribution statement
Contributor statement

I agree with being listed as a contributor of this thesis and to the conflicts of interest statement as summarised. I am a co-author of the manuscript entitled "AMS dating of ancient plant residues from experimental stone tools: a pilot study", which has been published in the Journal of Archaeological Science on 4 March 2013 (available online) and September 2014 (in print).

I am also a co-author of the manuscript entitled "Radiocarbon-dating adhesive and wooden residues from stone tools by Accelerator Mass Spectrometry (AMS): challenges and insights encountered in a case study", which has been published in the Journal of Archaeological Science on 12 March 2015 (available online) and will be available in print in September 2015.

I am also a co-author of the manuscript entitled "Residue radiocarbon AMS dating review and preliminary sampling protocol suggestions", DOI: 10.1016/j.jas.2015.06.011, which has been accepted for publication in the Journal of Archaeological Science in 19 June 2015.

I have had access to all the data in these studies and accept responsibility for their validity.

Dr Andrew M. Smith
Signature: Date: 24/06/15.
Contributor statement

I agree with being listed as a contributor of this thesis and to the conflicts of interest statement as summarised. I am a co-author of the manuscript entitled "AMS dating of ancient plant residues from experimental stone tools: a pilot study ", which has been published in the Journal of Archaeological Science on 4 March 2013 (available online ) and September 2014 (in print).

I am also a co-author of the manuscript entitled "Radiocarbon-dating adhesive and wooden residues from stone tools by Accelerator Mass Spectrometry (AMS): challenges and insights encountered in a case study ", which has been published in the Journal of Archaeological Science on 12 March 2015 (available online ) and will be available in print in September 2015.

I am also a co-author of the manuscript entitled "Residue radiocarbon AMS dating review and preliminary sampling protocol suggestions", DOI: 10.1016/j.jas.2015.06.011.", which has been accepted for publication in the Journal of Archaeological Science on 19 June 2015.

I have had access to all the data in these studies and accept responsibility for their validity.

Fiona Bertuch
Signature: 
Date: 24/6/15
Contributor statement

I agree with being listed as a contributor of this thesis and to the conflicts of interest statement as summarised. I am a co-author of the manuscript entitled "AMS dating of ancient plant residues from experimental stone tools: a pilot study", which has been published in the Journal of Archaeological Science in 4 March 2013 (available online) and September 2014 (in print).

I am also a co-author of the manuscript entitled "Radiocarbon-dating adhesive and wooden residues from stone tools by Accelerator Mass Spectrometry (AMS): challenges and insights encountered in a case study", which has been published in the Journal of Archaeological Science in 12 March 2015 (available online) and will be available in print in September 2015.

I have had access to all the data in these studies and accept responsibility for their validity.

Dr Jeffrey Parr

Signature: 

Date: 26/06/2015
Contributor statement

I agree with being listed as a contributor of this thesis and to the conflicts of interest statement as summarised. I am a co-author of the manuscript entitled "Radiocarbon-dating adhesive and wooden residues from stone tools by Accelerator Mass Spectrometry (AMS): challenges and insights encountered in a case study ", which has been published in the Journal of Archaeological Science on 12 March 2015 (available online) and will be available in print in September 2015.

I have had access to all the data in this study and accept responsibility for their validity.

Dr Birgit Gehlen  Signature:  Date: 23.06.15
Contributor statement

I agree with being listed as a contributor of this thesis and to the conflicts of interest statement as summarised. I am a co-author of the manuscript entitled "Radiocarbon-dating adhesive and wooden residues from stone tools by Accelerator Mass Spectrometry (AMS): challenges and insights encountered in a case study", which has been published in the Journal of Archaeological Science on 12 March 2015 (Vol 61, available online) and will be available in print in September 2015.

I have had access to all the data in this study and accept responsibility for their validity.

[Signature]

Dr. Bernard Grinsell

Signature: [Signature]

Date: June 27, 2005
Contributor statement

I agree with being listed as a contributor of this thesis and to the conflicts of interest statement as summarised. I am a co-author of the manuscript entitled "Radiocarbon-dating adhesive and wooden residues from stone tools by Accelerator Mass Spectrometry (AMS): challenges and insights encountered in a case study", which has been published in the Journal of Archaeological Science on 12 March 2015 (Vol 61, available online) and will be available in print in September 2015.

I have had access to all the data in this study and accept responsibility for their validity.

Dr Alfred Pawlik             Signature

Date: 23 June 2015
Contributor statement

I agree with being listed as a contributor of this thesis and to the conflicts of interest statement as summarised. I am a co-author of the manuscript entitled "Radiocarbon-dating adhesive and wooden residues from stone tools by Accelerator Mass Spectrometry (AMS): challenges and insights encountered in a case study", which has been published in the Journal of Archaeological Science on 12 March 2015 (available online) and will be available in print in September 2015.

I have had access to all the data in this study and accept responsibility for their validity.

Dr Martin Heinen

Signature: [Redacted] Date: 29.06.2015
APPENDIX 2
Appendix removed due to copyright restrictions
Citations Only

Conference Presentations

ABSTRACT: The study presents and evaluates a process to enhance residue AMS radiocarbon dating with a focus on contaminant confinement. Methods applied include 1) optical residue and use-wear analyses, 2) experimental designs addressing cleaning treatments to mitigate impact of contaminants, 3) preparation and extraction of residues from (mostly) previously dated stone artefacts, and 4) establishing the elemental characteristics of residues by using SEM/EDX as a final step to avoid sample contamination during analyses. The alkaline surfactant Decon 90, 2% diluted proved useful for the removal of skin scales and fabric fibre but effects on graphite contamination introduced by pencil lead were more limited. A number of artefact residues were dated within the anticipated age range, while other stone tools residues attained AMS dates which appear to be affected by modern contaminants or by fossil shell derived from flint stone, plasticizers or from a fixative substance older than the fabrication and use of the artefact. Chemical residue identification early on in the method sequence, using non-destructive and non-contaminating methods would guide the choice of residue treatment and improve reliability of age determination.
ABSTRACT: AMS radiocarbon dating of archaeological residues on a carbon microgram scale is a relatively new field in archaeological chronometric research. Successful applications of the method demonstrate its feasibility when working with such small sample sizes, but also reveal challenges and problems arising around contamination. In this paper we present a case study on testing the feasibility of radiocarbon-dating putative adhesive and wooden residues from archaeological stone tools which contain also contaminants. Methods applied include: (1) optical residue interpretation; and (2) experimental designs in which we address contamination by testing the affectivity of removal techniques. Furthermore, we examine how SEM-EDX analyses assists in residue interpretation, conducted as the final step to avoid sample contamination during analyses. Even though this methodological sequence was in general successful in dating some lithic residues within the anticipated age range, difficulties were encountered with other artefacts. We found that the alkaline surfactant Decon 90 is a useful solution for the removal of skin scales and fabric fibre but has limited effect on graphite contamination, introduced by pencil lead. While some artefact residues attained AMS dates which appear affected by modern contaminants, other residue radiocarbon dates were seemingly affected by fossil shell derived from flint stone, plasticizers, graphite or from a fixative substance older than fabrication and use of the artefact. One outcome from this study is that early non-destructive residue identification in the method sequence would guide the choice of residue treatment and improve the reliability of the age determination. Specific sampling protocols, would assist in enhancing residue AMS dating.
ABSTRACT: Radiocarbon dating of microgram residues is a relatively new field in archaeological science, so far limited by analytical protocols and instrumentation. While successful applications using Accelerator Mass Spectrometry (AMS) have demonstrated the potential of the technique on such small samples, the analysis revealed challenges and problems, especially with contamination. Frequently, the presence of contaminants on the residue sample induces radiocarbon ages offset. Therefore, both residue identifications and contaminant removal protocols are keystone to achieve accurate dating. Along with this consideration, several other important steps in the sampling protocol sequence can influence the results and need to be carefully assessed. A constant improvement of sampling strategies and extracting methods to avoid contaminants compromising AMS dates is essential. Here we propose strategies for artefact handling (e.g. field work and archive) favorable for accurate AMS dating of stone tools residue.
ABSTRACT: A previous study by Yates et al. (2013) demonstrated the feasibility of AMS dating residues from recently manufactured stone tools, on securely dated plant material by careful control of post-depositional contaminants including those possible during sample preparation. Since the results of that project demonstrated the applicability of AMS dating on residues, it seems plausible to now establish the possibility of applying the successful aspects of the methodology on artefacts from archaeological contexts. Therefore, the focus of the current study will be to conduct follow up research on European, Late Palaeolithic and Mesolithic stone tools, which originate from excavated and dated archaeological sites including a well-dated stratigraphy. We examine residues from stone tools of the German bog site Friesack 4, which comprises the most detailed stratigraphy from the Mesolithic in Europe known so far and from the Late Palaeolithic site in Wesseling (Germany) where the one phase occupation is revealed by four matching 14C dates. While the stone tools come with securely dated ages and ample residues, these artefacts also contain a range of post-depositional related contaminants. In this present study we examine ways to remove non-use-related residues from artefacts, without compromising the integrity of use-related residues. Additionally we focus on residue sample preparation for AMS dating.

Yates, AB, Smith, AM, Bertuch, F, Scheffers, A, parr, J & Joannes-Boyau, R 2013, 'New approaches to AMS dating residues from stone tools (poster)', paper presented to the AAA Annual Conference, 2-4 December, Coffs Harbour, NSW.

Published version available from: http://australianarchaeology.com/gallery/yates-etal-2013/
ABSTRACT: Residue analyses on stone artefacts have contributed to resolve functional questions. While identifying the function of stone tools through the analysis of their micro-residues is possible, to date it has been difficult to establish a chronology for stone artefacts from surface scatters and those without a clear stratigraphic sequence. This paper explores the possibility direct dating of residues on stone artefacts by accelerated mass spectrometry (AMS) radiocarbon dating. While direct dating of blood residue on stone artefacts has been published previously, the innovative approach here is 1) the use of mid and late Holocene dated plant material; wood, and peat processed with contemporary manufactured stone flakes under controlled laboratory conditions and 2) the use of a carbon mass less than 22 µg. Wood residues delivered dates corresponding with the original sample, while dated peat residues showed an expected wider variation range, due the materials lack of homogeneity, nevertheless within an expected perimeter. These results clearly demonstrate that direct dating of residues on stone artefacts using recent advances in AMS dating techniques is possible.

ABSTRACT: Stone artefacts found in the Tweed and Byron Shires in northeastern New South Wales were produced from a range of raw materials, reflecting the diversity of geologic formations in the study area. These include volcanic rocks such as basalt, obsidian, chert and chalcedony, which were formed in the cavities of the volcanic host rocks, and metamorphic rocks such as greywacke. The aims of this study are firstly to identify potential quarry or stone resource sites for artefact production and secondly to explore how the raw material resources were reflected in the archaeological record. This has resulted in the identification of 38 outcrops and stone resource sites, which are outlined within this poster. Archaeological investigated sites with indication of raw material proportions show a preference for chert for stone tool production.


Published version available from:
http://dx.doi.org/10.1016/j.jas.2013.02.016

Abstract:
Residue analyses on stone artefacts have contributed to resolving functional questions in stone tool research. Although identifying the function of tools through the analysis of their micro-residues is possible, the establishment of a sound numerical chronology for stone tools lacking a clear stratigraphic sequence, such as surface scatters, remains a challenge. While radiocarbon dating of blood residue on stone artefacts has been published previously (Loy 1987, 1990, 1993; Loy et al., 1990; Nelson et al.1986), this paper reports on an experiment designed to assess the possibility of directly dating residues on stone artefacts by accelerator mass spectrometry (AMS) based radiocarbon measurements. Innovative with this approach is (1) the use of mid and late Holocene pre-dated plant material (wood and peat), processed with contemporarily manufactured stone flakes under controlled laboratory conditions and (2) the use of very small carbon masses (less than 22 mg) for radiocarbon dating. The 14C results of the wood residues are in excellent agreement with the original sample, whereas the 14C results of the peat residues yield a wider age variation as expected due to the inhomogeneity of the material, but nevertheless, provided dates within an expected age range. Preliminary results demonstrate the feasibility of dating very small amounts of plant residue on lithics directly when contaminants are confined.
CHAPTER 4 - Published Manuscript
CHAPTER 4 - Published Manuscript


Published version available from:
http://dx.doi.org/10.1016/j.jas.2015.04.022
APPENDIX 5
Appendix removed due to copyright restrictions

Citation Only

CHAPTER 5 - Published Manuscript


Published version available from:
http://dx.doi.org/10.1016/j.jas.2015.06.011
APPENDIX 6

Appendix removed due to copyright restrictions

Citation Only

Side Publication

APPENDIX 7

Appendix removed due to copyright restrictions
Citation Only

Side Publication