OCCUPATIONAL EXPOSURE TO ORGANOPHOSPHORUS PESTICIDES: EXPLORATORY CASE STUDIES OF FACTORS INFLUENCING GLOVE PERFORMANCE AND SKIN PENETRATION

ISMANIZA ISMAIL
MBiotech, B.Sc. (Hons), Dip

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

Doctor of Philosophy

THE UNIVERSITY OF ADELAIDE
SCHOOL OF PUBLIC HEALTH, FACULTY OF HEALTH SCIENCES, THE UNIVERSITY OF ADELAIDE, SOUTH AUSTRALIA

2016
DECLARATION

I certify that this work contains no material which has been accepted for the award of any other degree or diploma in my name in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text. In addition, I certify that no part of this work will, in the future, be used in a submission in my name for any other degree or diploma in any university or other tertiary institution without the prior approval of the University of Adelaide and where applicable, any partner institution responsible for the joint award of this degree.

I give consent to this copy of my thesis when deposited in the University Library, being made available for loan and photocopying, subject to the provisions of the Copyright Act 1968.

The author acknowledges that copyright of published works contained within this thesis resides with the copyright holder(s) of those works.

I also give permission for the digital version of my thesis to be made available on the web, via the University’s digital research repository, the Library Search and also through web search engines, unless permission has been granted by the University to restrict access for a period of time.

Ismaniza Ismail
ACKNOWLEDGEMENTS

As with any large project of this kind, there are many people who need to be thanked for their involvement and support. I have not named everyone who has been associated with the successful completion of my long PhD journey, for this would fill a book and really put a strain on my memory box, but for those of you I have missed on this page, I thank you now.

I am especially grateful for the generous financial support provided by the Ministry of Higher Education (MOHE) Malaysia and Universiti Teknologi MARA (UiTM) that sustained the costly side of this endeavour.

I would like to thank the supervisory panels; Prof. Dino Pisaniello and Dr. Sharyn Gaskin for providing me the inspiration, knowledge and confidence to conduct this project. I would never be able to run if you did not teach me how to walk. My gratitude also goes to Assoc. Prof. John Edwards of Flinders University, for encouraging me to always ‘look at the bright side’ though I always put a frown and confused face.

A number of people have helped me along the road in some small or large way, bringing me out of the dark, especially the OEH Lab team; Dr. Joe Crea, Mr. Ganyk Jankewicz, Dr. Michael Tkaczuk, Dr. Linda Heath, Dr. Richard Evans and Mr. Len Turcynowicz.

Special thanks to Tim Burfield (IPM) and the farmers of Virginia farms for the preliminary observation experience, and also the statisticians from DMAC; Suzanne Edwards and Kara Cashman.

No less important are the members of the Thesis Writing Group for providing me critical insights for continual improvement and those who helped me to get to where I wanted to go; Prof Mike Capra and Prof Ross Barnard (UQ) and Assoc. Prof. Wan Ramlee (UiTM). Thank you very much.

Not forgetting, my best friends; Jane, So Young, Anep, Sharon, Winie, Yahya, Maisarah, Izzah, Saz and Ferial for contributing wisdom, humour, intrigue and spice to everyday life. This is something I am thankful for and will always be.

Those who have immeasurable influence in my life- my family, in-laws, Uncle Mokhtar, Aunty Hamidah and Mary, they have always been there for me when I needed them (and even when I thought I did not) and for that I am extremely grateful.

Finally, I would like to thank my dear husband and my two boys; Adam and Adib for their incredible patience, love and support, without which the going would have been considerably tougher.

_It was a hard road, but the best road I took to be where I am!_
ABSTRACT

Problem statement

The widespread use of organophosphorus pesticides (OPs) in agriculture and urban pest control has seen significant morbidity and mortality. They are the most important cause of severe toxicity and death from acute poisoning worldwide, with more than 200,000 deaths each year in developing countries. OPs act to inhibit acetylcholinesterase in the nervous system, and may elicit acute and long term health effects. Some commercially available OPs are also classified as Chemicals of Security Concern by the Australian Government and are subject to special restrictions and surveillance.

Occupational exposure may occur in manufacture, storage, transport, usage, disposal and in emergency situations. For example, ambulance workers may be exposed when attending accidental or intentional OP poisonings. The profiles may vary from short term high exposures for emergency personnel, to long term low exposures for farming communities.

The World Health Organisation and a range of other agencies have determined that the continued use of these chemicals represents an important public and occupational health issue.

A major route of exposure is skin absorption, and the use of protective chemical gloves is recommended, especially for the concentrated product. Safety data sheets (SDS) are meant to provide the user with information on appropriate glove protection. However, the concentrates are typically formulated products with solvents and additives that may influence the glove permeation behaviour of the active ingredient. Testing by glove manufacturers is rarely with the formulated product, and pertains to room temperature experiments with new gloves. Cheap or disposable
gloves may not have been tested. In addition, the effects of elevated temperature, abrasion and ultraviolet (UV) exposure are poorly understood.

Although these environmental factors are likely to be present in Australian workplaces, there is scant evidence of their practical significance to inform risk assessment and interventions, including glove selection and use. Apart from gloves, skin uptake may be influenced by high ambient temperatures. The limited dermal toxicological research literature suggests that skin penetration rates may be dramatically increased, with concomitant increases in health risk. Compounding the problem is the situation where chemical is occluded between glove and skin.

Gap analysis

Based on a review of the occupational hygiene literature, knowledge gaps exist in three areas, relating to glove performance and skin penetration.

Firstly, suppliers and formulators of agricultural OPs recommend the use of long length polyvinyl chloride (PVC) gloves. Indeed, distributors stock cheap versions of such gloves, but as they are not considered disposable, repeated use is the norm. Scientific uncertainty exists as to the protection afforded under real world conditions of solar radiation, elevated temperature and abrasion.

Secondly, the chemical barrier performance of disposable gloves, as worn by ambulance workers in poisoning episodes, has not been investigated.

Thirdly, it is known that skin absorption of chemicals depends on physicochemical properties such as molecular size and water solubility. However, there appear to be no skin penetration studies which have compared a range of OPs of different properties. Moreover, the studies that have been
conducted have often used a finite dose model. An infinite dose model is more applicable for worst case occupational risk assessment.

Purpose statement

This research aims to provide a better understanding of dermal exposure to OPs (under simulated real world conditions) and to generate data useful for predictive dermal risk assessment models and optimising control measures. It is anticipated that the improved evidence base will assist in reducing morbidity and mortality from OPs. A case study approach will be used, addressing both routine users and those in an emergency setting.

Broad Research Questions

The following questions were developed from a literature review and context scoping from fieldwork observations.

1. How do the recommended PVC gloves (unused, exposed to UV and abrasion) perform against OPs in various exposure conditions?
2. What are the effects of different variable conditions on the barrier performance of different types of disposable gloves worn by ambulance workers and is the current practice suitable?
3. How is skin penetration affected by physicochemical properties of OPs, concentration of OPs and elevated temperature?

Methodology

Four OPs widely used in Australia (omethoate, mevinphos, dichlorvos and diazinon) with differing physicochemical properties (e.g. octanol-water partition coefficient) were investigated. Tests were conducted at the full strength formulation (relevant for transport and mixing activities) and application strength (relevant for spraying), and at two temperatures.

In the glove permeation studies, American Society for Testing and Materials (ASTM) permeation cells were used at two temperatures (room temperature and 45°C).

Two case study scenarios were used, namely agricultural workers and ambulance workers.

For Case Study 1 two brands of elbow length PVC gloves recommended for handling OPs were tested in new unused condition, and following UV exposure or abrasion. In Case Study 2 disposable nitrile and neoprene gloves used by South Australia Ambulance Service (SAAS) workers were tested individually and in combination (as per current practice).

In the skin studies, human abdominal skin samples (heat separated epidermis) were exposed to the OPs (infinite dose) in static Franz cells according to OECD protocols for up to 8 hours, at room temperature and 37°C. Analysis for the OPs in both glove and skin experiments was undertaken using HPLC and UV detection.
Main Findings

Glove performance tests – Case Study 1

Differences were noted in breakthrough time and cumulative penetration at the end of the 8-hour experiments for the two PVC glove brands. In general, PVC gloves performed well against the four tested OPs. Full strength formulations had shorter breakthrough times and greater cumulative permeations. Breakthrough of diazinon did not occur unless at full strength at 45°C (120 - 180 mins). In contrast, breakthrough of dichlorvos was observed for all exposure conditions (between 60 to 240 mins) except for application strength at room temperature. Mevinphos, which was only tested at room temperature, demonstrated breakthrough only for one brand of gloves. In the case of omethoate, breakthrough was noted from 60 minutes onwards.

Permeation was uniformly higher at 45°C compared to room temperature. Gloves exposed to UV light (approximately equivalent to a week or more of extreme sunlight) or abrasion (5% thickness reduction) exhibited a small reduction in performance.

Glove performance tests – Case Study 2

In glove performance tests on gloves used by SAAS workers, disposable nitrile and neoprene gloves demonstrated good protection against the four tested OPs in 4-hour experiments. In general, the thicker neoprene gloves demonstrated longer breakthrough times than nitrile gloves.

At higher test concentration (full strength) of the four tested OPs, cumulative penetration at 8 hrs increased, and this was observed at both test temperatures. Elevated temperature to 45°C shortened
the breakthrough time (between 5 and 20 minutes) and resulted in greater cumulative penetration of
the OPs.
Combining the disposable gloves (nitrile on neoprene) as practised by SAAS workers demonstrated
better protection with longer breakthrough times and lower cumulative penetration of the tested
OPs, compared to individual gloves.

*In vitro skin studies*

Dichlorvos was found to rapidly penetrate the skin. Elevated temperature and higher OP
congestion resulted in faster penetration rate and increased cumulative penetration of the tested
OPs.
Comparison of the skin penetration data with Acceptable Daily Intakes (ADIs) of the respective
OPs showed that ADIs may be exceeded rapidly, with the order being dichlorvos, diazinon,
omethoate and mevinphos.

*Novelty of the research*

This research uses an experimental design based on real life scenarios.
Unlike most glove permeation and skin penetration studies this research used formulated OP
products that are commercially available in the market. All experiments were conducted using an
infinite dose model to simulate the worst exposure scenario (prolonged skin contamination from
spills and splashes), and thus to establish the maximum penetration rates for risk assessment.
Similarly, tests were conducted at elevated temperatures and with gloves exposed to ultraviolet (UV) and abrasion to mimic the common exposure conditions in real world scenarios.

**Implications**

The research has highlighted the importance of elevated temperature, and the dermal risks associated with dichlorvos. The increased barrier protection afforded by double gloving (combining relatively polar and non-polar materials), for OPs of different physico-chemical properties, was shown. This is reassuring for ambulance workers who may encounter a range of OPs. Surprisingly, the influence of UV exposure and abrasion on the performance of the thick PVC gloves was found to be relatively minor, under the conditions examined.

Data on the influence of physicochemical properties, concentration, and temperature are useful for refining dermal risk assessment models, and for worker education. On a broader public health level, the findings will allow a more rational use of gloves as chemical protective clothing to protect the population at risk from the danger of agricultural chemicals.

**Conclusions and Recommendations**

The data indicate that the use of concentrated OPs in warmer conditions will greatly increase skin uptake. Dichlorvos demonstrated rapid and extensive skin penetration, but the data for all the tested OPs suggest that toxicologically important uptake can occur in relatively short time periods without
skin protection. Glove permeation also increases with concentration, temperature, UV and abrasion.
The practice of double gloving by ambulance workers is supported by the evidence.

Following these exploratory studies, it is recommended that more glove permeation tests be conducted under realistic exposure scenarios. The findings should be compiled in a database for advisory purposes and made publicly accessible. Warnings for potential ADI exceedance should be included on labels or SDSs of the OP products to alert the users of the risks when handling OPs. Where changes may be made in the OP formulation by manufacturers, the suitability of glove recommendations should be reviewed.
TABLE OF CONTENTS

DECLARATION......................................................................................... i
ACKNOWLEDGEMENTS........................................................................... ii
ABSTRACT................................................................................................. iii
TABLE OF CONTENTS.............................................................................. xi
LIST OF FIGURES................................................................................... xix
LIST OF TABLES ...................................................................................... xxii
LIST OF ABBREVIATIONS ......................................................................... xxiv
GLOSSARY.................................................................................................. xxv
PUBLICATIONS, PRESENTATIONS AND RECOGNITIONS .................... xxvii
THESIS OVERVIEW.................................................................................. xxx

CHAPTER 1 GENERAL INTRODUCTION.................................................. 1
1.1 Organophosphorus pesticides (OPs) – Hazards and Exposures ............ 2
1.2 Organophosphorus pesticides classified as Chemicals Security Concern .. 10
1.3 Public health significance of OP poisonings ....................................... 15
1.4 The need for research on dermal exposure assessment and control .......... 16

CHAPTER 2 LITERATURE REVIEW...................................................... 18
2.1 Objectives of the literature review ................................................... 20
2.2 Literature search strategy .................................................................. 20
2.3 Search outcomes .............................................................................. 22
2.3.1 Compilation of the key literature .................................................. 22
2.3.2 Narrative synthesis of the relevant literature………………………………….. 40
2.3.2.1 Overview……………………………………………………………………… 40
2.3.2.2 Azinphos methyl…………………………………………………………….. 42
2.3.2.3 Cadusafos…………………………………………………………………….. 43
2.3.2.4 Diazinon……………………………………………………………………….. 44
2.3.2.5 Dichlorvos…………………………………………………………………….. 46
2.3.2.6 Disulfoton…………………………………………………………………….. 48
2.3.2.7 Ethion………………………………………………………………………….. 49
2.3.2.8 Fenamiphos…………………………………………………………………… 50
2.3.2.9 Mevinphos…………………………………………………………………….. 51
2.3.2.10 Omethoate…………………………………………………………………… 52
2.3.2.11 Parathion methyl…………………………………………………………….. 53
2.3.2.12 Phorate………………………………………………………………………… 53
2.3.2.13 Terbufos……………………………………………………………………….. 55
2.4 Existing guidance on glove selection……………………………………………… 56
2.4.1 Specific guidance………………………………………………………………. 56
2.4.2 General guidance………………………………………………………………. 60
2.5 Uncertainties and gaps in knowledge in relation to dermal exposure to OPs…… 63
2.6 Proposed focus for the current research………………………………………….. 64
2.7 Aims and broad research questions……………………………………………….. 68
2.8 What is not addressed in this thesis……………………………………………….. 69
CHAPTER 3 MATERIALS AND METHODS

3.1 Glove performance studies for Case Study 1 (PVC gloves used by agricultural workers)

3.1.1 Types of PVC gloves tested for performance against OP permeation

3.1.1.1 Agricultural PVC gloves (unused)

3.1.1.2 PVC gloves exposed to ultraviolet (UV) radiation

3.1.1.3 PVC gloves with abrasion

3.1.2 Test compounds (organophosphorus pesticides) used in the study

3.1.3 Glove performance tests using ASTM cells

3.1.3.1 Temperature setting and duration for glove tests

3.1.3.2 Receptor fluids for glove performance tests

3.1.4 Descriptors of permeation

3.1.5 Quality assurance/ quality control

3.1.6 Analysis of OPs

3.1.7 Limits of detection/ limit of quantification of the OPs

3.1.8 Calibration graph

3.2 Glove performance studies for Case Study 2 (disposable gloves used by ambulance workers)

3.2.1 Types of disposable gloves tested for performance against OP permeation

3.2.1.1 Disposable nitrile gloves

3.2.1.2 Disposable neoprene gloves

3.2.1.3 Combination of disposable gloves (nitrile on neoprene)

3.2.2 Test compounds (organophosphorus pesticides) used in the study

3.2.3 Glove performance tests using ASTM cells
CHAPTER 4 CASE STUDY 1: PROTECTIVE PERFORMANCE OF POLYVINYL CHLORIDE (PVC) GLOVES USED BY AGRICULTURAL WORKERS AGAINST ORGANOPHOSPHORUS PESTICIDES

4.1 Agricultural workers as a study population

4.1.1 Opportunities for exposure from preliminary field observation on work practices and behaviours

4.1.2 Rationale and aims of study

4.2 Worst case approach

4.3 Results

4.3.1 OP permeation through unused (out of the box) PVC gloves

4.3.1.1 Excalibur gloves

4.3.1.2 ProChoice gloves

4.3.1.3 Comparative glove performance

4.3.2 OP permeation through UV-exposed PVC gloves

4.3.3 OP permeation through abraded PVC gloves

4.3.4 Statistical tests

4.4 Discussion

4.4.1 Effects of concentration and exposure temperature on permeation of OPs through unused PVC gloves

4.4.2 Effects of UV exposure on OP permeation through PVC gloves

4.4.3 Effects of abrasion on OP permeation through PVC gloves

4.4.4 Glove use from preliminary field observation

4.5 Conclusion
CHAPTER 5 CASE STUDY 2: PROTECTIVE PERFORMANCE OF DISPOSABLE GLOVES USED BY AMBULANCE WORKERS AGAINST ORGANOPHOSPHORUS PESTICIDES

5.1 Ambulance workers as a study population

5.1.1 Opportunities for exposure and current work practices

5.1.2 Rationale and aims of study

5.2 Worst case approach

5.3 Results

5.3.1 Effects of concentration on OP permeation through disposable nitrile gloves

5.3.2 Effects of concentration on OP permeation through disposable neoprene gloves

5.3.3 Effects of concentration on OP permeation through combination of nitrile and neoprene gloves (nitrile and neoprene)

5.3.4 Time series cumulative permeation data

5.3.5 Comparison of the protection provided by individual gloves and the gloves worn in combination

5.3.6 Statistical tests

5.4 Discussion

5.4.1 Effects of concentration and exposure temperature on permeation of OPs through disposable gloves

5.4.2 Protection afforded by individual gloves vs combination of gloves against OPs

5.5 Conclusion
CHAPTER 6 SKIN STUDIES OF OMETHOATE, MEVINPHOS, DICHLORVOS AND DIAZINON

6.1 Introduction
6.1.1 Dermal exposure of OPs at work
6.1.2 Rationale and aims of study
6.2 Worst case approach
6.3 Results
6.3.1 Skin integrity checks- pre- and post-chemical challenge
6.3.2 Skin penetration of omethoate, mevinphos, dichlorvos and diazinon
6.3.3 Effects of concentration of OPs on skin penetration
6.3.4 Effects of exposure temperature on skin penetration
6.3.5 Statistical tests
6.3.6 Comparison of skin penetration outcomes with Acceptable Daily Intakes (ADIs)
6.4 Discussion
6.4.1 Major findings
6.4.2 Strength and weakness of this study
6.5 Conclusion

CHAPTER 7 GENERAL DISCUSSION

7.1 Novelty of the research
7.2 Major findings of the studies
7.2.1 Real world protection provided by gloves
7.2.1.1 The influence of concentration of OPs and exposure temperature on glove permeation
<table>
<thead>
<tr>
<th>Section</th>
<th>Title</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>7.2.1.2</td>
<td>The influence of UV exposure and abrasion on glove permeation</td>
<td>217</td>
</tr>
<tr>
<td>7.2.2</td>
<td>Skin uptake at infinite dosing</td>
<td>218</td>
</tr>
<tr>
<td>7.2.2.1</td>
<td>Variability of skin penetration for different types of OPs</td>
<td>218</td>
</tr>
<tr>
<td>7.2.2.2</td>
<td>The influence of concentration of OPs and exposure temperature on skin penetration</td>
<td>219</td>
</tr>
<tr>
<td>7.2.2.3</td>
<td>Other factors that might affect glove permeation and skin penetration</td>
<td>221</td>
</tr>
<tr>
<td>7.2.2.4</td>
<td>Comparison of skin penetration of OPs with Acceptable Daily Intakes (ADIs)</td>
<td>223</td>
</tr>
<tr>
<td>7.2.2.5</td>
<td>The application of skin penetration outcomes for predictive dermal risk assessment models</td>
<td>225</td>
</tr>
<tr>
<td>7.3</td>
<td>Generalisability of results</td>
<td>227</td>
</tr>
<tr>
<td>7.4</td>
<td>Strength and limitations of the overall research</td>
<td>228</td>
</tr>
</tbody>
</table>

**CHAPTER 8 CONCLUSIONS AND RECOMMENDATIONS** 230

<table>
<thead>
<tr>
<th>Section</th>
<th>Title</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>8.1</td>
<td>General conclusions from the research</td>
<td>231</td>
</tr>
<tr>
<td>8.2</td>
<td>General recommendations</td>
<td>232</td>
</tr>
<tr>
<td>8.2.1</td>
<td>Recommendations for future researchers</td>
<td>232</td>
</tr>
<tr>
<td>8.2.2</td>
<td>Recommendations for glove users</td>
<td>233</td>
</tr>
<tr>
<td>8.2.3</td>
<td>Recommendations for manufacturers and suppliers of OPs and gloves</td>
<td>234</td>
</tr>
<tr>
<td>8.2.4</td>
<td>Recommendations for regulatory and enforcement bodies</td>
<td>235</td>
</tr>
</tbody>
</table>

**REFERENCES** 236

**APPENDICES** 265
**LIST OF FIGURES**

| Figure 1.1 | Conceptual model of occupational OP exposure | 4 |
| Figure 2.1 | Labels of the commercial formulated OPs (omethoate-Folimit®; mevinphos-Phosdrin®; diazinon-Barmac Diazinon Insecticide) tested in this research | 66 |
| Figure 3.1 | Elbow length PVC gloves tested in the study; Excalibur (left) and ProChoice (right) | 72 |
| Figure 3.2 | Swatches of PVC gloves exposed to UV radiation in the UV box | 74 |
| Figure 3.3 | Comparison between PVC gloves exposed to UV light and abrasion | 76 |
| Figure 3.4 | Formulated omethoate used in the study, available as Folimat® | 78 |
| Figure 3.5 | The formulated mevinphos, manufactured as Phosdrin® | 79 |
| Figure 3.6 | Diazinon formulated by Barmac used in the study | 81 |
| Figure 3.7 | Assembly of the ASTM permeation test cell with glove swatch in between channelled stopper and modified stirrer | 83 |
| Figure 3.8 | Disposable nitrile gloves (Sterling® Nitrile) used by the SAAS ambulance workers for attending chemical-related incidents | 92 |
| Figure 3.9 | Disposable neoprene gloves (Micro-touch® Affinity® Neoprene) used by the SAAS ambulance workers for attending chemical-related incidents | 93 |
| Figure 3.10 | Cross section of the assembly of ASTM permeation test cells for testing the performance of combination of disposable gloves used by ambulance workers | 94 |
| Figure 3.11 | The process of harvesting the donated human skin | 99 |
| Figure 3.12 | Skin barrier integrity was checked using Tinsley LCR Databridge 6401 by placing electrodes in the donor chamber and receptor chamber containing saline | 101 |
| Figure 3.13 | The static Franz diffusion cell was covered with a rough marble to avoid evaporation loss, without creating pressure differentials | 102 |
Figure 4.1: Exposure to OPs may occur during loading, mixing (left) and spraying (right) ................................................................. 112

Figure 4.2: The hose roller and vehicle used for crop harvesting are potentially contaminated with pesticides from deposition and transfer from contaminated surface .................................................. 114

Figure 4.3: Comparison of cumulative permeation (mg) of OPs over the 8-hour test at application strength through Excalibur gloves at room temperature (top) and 45°C (bottom). ................................................................. 120

Figure 4.4: Comparison of cumulative permeation (mg) of OPs over the 8-hour test at full strength through Excalibur gloves at room temperature (top) and 45°C (bottom). ................................................................. 121

Figure 4.5: Comparison of cumulative permeation (mg) of OPs over the 8-hour test at application strength through ProChoice gloves at room temperature (top) and 45°C (bottom). ................................................................. 124

Figure 4.6: Comparison of cumulative permeation (mg) of OPs over the 8-hour test at full strength through ProChoice gloves at room temperature (top) and 45°C (bottom). ................................................................. 125

Figure 4.7: Comparison of 8-hr cumulative permeation of OPs (mg) through Excalibur and ProChoice gloves (unused gloves versus UV-exposed gloves) in various experimental conditions ................................................................. 131

Figure 4.8: Comparison of cumulative permeation of OPs (mg) through Excalibur and ProChoice gloves (unused gloves versus abraded gloves) in various experimental conditions ................................................................. 136

Figure 5.1: Simulation of exposure potential when SAAS ambulance worker wearing double gloves attending to a pesticide poisoning case............ 151

Figure 5.2: Double gloving method (nitrile on top of neoprene gloves) practised by SAAS ambulance workers ................................................................. 152

Figure 5.3: Comparison of average cumulative permeation (mg) of full strength OPs through disposable nitrile gloves at room temperature (top) and 45°C (bottom) ................................................................. 163
Figure 5.4  Comparison of average cumulative permeation (mg) of full strength OPs through disposable neoprene gloves at room temperature (top) and 45°C (bottom)……………………………………………………………………………… 164
Figure 5.5  Comparison of average cumulative permeation (mg) of full strength OPs through combination of nitrile and neoprene gloves at room temperature (top) and 45°C (bottom)……………………………………… 165
Figure 5.6  Comparison of average cumulative permeation of OPs (mg) in various experimental conditions by glove types…………………………………………………………… 167
Figure 6.1  Cumulative penetration of application strength OPs (mg) through human skin at room temperature (top) and 37°C (bottom)………………… 190
Figure 6.2  Cumulative penetration of full strength OPs (mg) through human skin at room temperature (top) and 37°C (bottom)……………………………………… 191
Figure 6.3  Comparison of cumulative penetration of OPs (mg) at application strength and full strength through human skin at room temperature (top) and 37°C (bottom) ……………………………………………………………… 193
Figure 6.4  Intercellular and transcellular routes of chemical penetration through the ‘brick and mortar’ structure of the upper skin…………………………… 200
LIST OF TABLES

Table 1.1  Shortlisting of organophosphorus pesticide (OP) chemicals from the Chemicals of Security Concern for this research………………………11

Table 2.1  Logic grid used in the literature search strategy……………………………………20

Table 2.2  Key findings from the shortlisted articles…………………………………………22

Table 2.3  List of typical commercially available OPs and recommended hand protection (gloves) .................................................................56

Table 2.4  Physicochemical properties of four OPs of interest that may influence skin penetration and glove permeation……………………………………64

Table 3.1  Summary of the OP concentrates tested……………………………………76

Table 3.2  Summary of HPLC setting for the tested OPs……………………………..88

Table 4.1  Average cumulative permeation (mg) of OPs after 8 hrs, and breakthrough times (with asterisk) through unused Excalibur gloves at room temperature and 45°C………………………………………118

Table 4.2  Average cumulative permeation (mg) of OPs after 8 hrs and breakthrough times (with asterisk) through unused ProChoice gloves at room temperature and 45°C………………………………………122

Table 4.3  Comparison of 8-hr cumulative permeation (mg) of OPs and breakthrough times (with asterisk) through unused Excalibur and UV-exposed Excalibur gloves at room temperature and 45°C……………129

Table 4.4  Comparison of 8-hr cumulative permeation (mg) of OPs and breakthrough times (with asterisk) through unused ProChoice and UV-exposed ProChoice gloves at room temperature and 45°C……………………………………129

Table 4.5  Average 8-hr cumulative permeation (mg) of OPs with breakthrough times (with asterisk) through abraded Excalibur gloves at room temperature and 45°C………………………………………133

Table 4.6  Average 8-hr cumulative permeation (mg) of OPs with breakthrough times (with asterisk) through abraded ProChoice gloves at room temperature and 45°C………………………………………134
Table 5.1  Average 4-hour cumulative permeation (mg) of OPs and breakthrough times through disposable nitrile gloves at room temperature and 45°C...

Table 5.2  Average 4-hour cumulative permeation (mg) of OPs with breakthrough times (with asterisk) through disposable neoprene gloves at room temperature and 45°C………………………………………………………… 158

Table 5.3  Average 4-hour cumulative permeation (mg) of OPs with breakthrough times (with asterisk) through combination of nitrile and neoprene gloves at room temperature and 45°C…………………………………… 161

Table 6.1  Summary of the outcomes of the in vitro skin studies on four OPs of full strength and application strength at two exposure temperatures for 8-hour challenge………………………………………………………… 188

Table 6.2  Estimated time to exceed ADI for a 70-kg human with corresponding calculated cumulative penetration for hands only and hands and forearms for the four tested OPs in various exposure conditions (application strength, and full strength, at 23°C and 37°C) and the level of ADI exceedance………………………………………………………… 198
**LIST OF ABBREVIATIONS**

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>AChE</td>
<td>Acetylcholinesterase</td>
</tr>
<tr>
<td>ADI</td>
<td>Acceptable Daily Intake</td>
</tr>
<tr>
<td>ANOVA</td>
<td>Analysis of Variance</td>
</tr>
<tr>
<td>APVMA</td>
<td>Australian Pesticides and Veterinary Medicines Authority</td>
</tr>
<tr>
<td>ASTM</td>
<td>American Society for Testing and Materials</td>
</tr>
<tr>
<td>AS/NZS</td>
<td>Australian Standards/New Zealand Standards</td>
</tr>
<tr>
<td>ATSDR</td>
<td>Agency for Toxic Substances and Disease Registry</td>
</tr>
<tr>
<td>BT</td>
<td>Breakthrough time</td>
</tr>
<tr>
<td>CAS</td>
<td>Chemical Abstracts Service Registry Number</td>
</tr>
<tr>
<td>EC</td>
<td>Emulsifiable concentrate</td>
</tr>
<tr>
<td>HPLC-UV</td>
<td>High Performance Liquid Chromatography – Ultraviolet Detection</td>
</tr>
<tr>
<td>IUPAC</td>
<td>International Union of Pure and Applied Chemistry</td>
</tr>
<tr>
<td>LD₅₀</td>
<td>50% lethal dose</td>
</tr>
<tr>
<td>Log K&lt;sub&gt;ow&lt;/sub&gt;</td>
<td>Log Octanol-Water Partition Coefficient</td>
</tr>
<tr>
<td>NIOSH</td>
<td>National Institute Occupational Safety and Health</td>
</tr>
<tr>
<td>NOEL</td>
<td>No Observable Effect Level</td>
</tr>
<tr>
<td>OPIDN</td>
<td>Organophosphate-induced delayed polyneuropathy</td>
</tr>
<tr>
<td>OP</td>
<td>Organophosphorus Pesticide</td>
</tr>
<tr>
<td>PPE</td>
<td>Personal Protective Equipment</td>
</tr>
<tr>
<td>PVC</td>
<td>Polyvinyl chloride</td>
</tr>
<tr>
<td>SAAS</td>
<td>South Australia Ambulance Service</td>
</tr>
<tr>
<td>SD</td>
<td>Standard deviation</td>
</tr>
<tr>
<td>SDS</td>
<td>Safety Data Sheet</td>
</tr>
<tr>
<td>WHO</td>
<td>World Health Organisation</td>
</tr>
<tr>
<td>WP</td>
<td>Wettable powder</td>
</tr>
</tbody>
</table>
**GLOSSARY**

**Permeation**
A process that works by diffusion; the movement of molecules through a layer from a high concentration area to an area of less concentration. Permeation through gloves occurs by absorption, diffusion and desorption of the chemicals.

For a chemical to permeate a glove material, the molecules of the chemical will absorb into the surface of the material, diffuse in the material and desorb to the opposite side of the material. Then, the chemical will come into contact with the skin and penetrates the skin. For the purpose of this research, the term ‘permeation’ is used for glove performance studies (Chapter 4 and 5).

**Penetration**
Direct passage of molecules through porous materials, or imperfections such as pinholes, cracks, tears and seams at a non-molecular level. It is easier to understand by saying penetration is relative to pressure. Therefore, penetration will occur at a faster rate when a higher pressure is applied. An example of this is the pinhole water test, which is done by filling in a glove with water and squeezed to test for any signs of penetration. For the purpose of this research, the term ‘penetration’ is used for *in vitro* skin studies.
Flux

The rate of permeation or penetration (μg/cm²/min) of which the amount of chemicals (μg) crossing a defined area (cm²) in a set time (min). Flux is commonly used to define penetration and permeation characteristics.

Breakthrough time

For gloves, breakthrough time (BT) is the time where the flux reaches 1 μg/cm²/min as set in the AS/NZS 2161 standard. Although BTs do not provide information on how much pesticide permeated through the gloves, it may be used to judge the quality of gloves, along with flux and cumulative permeation. Although some chemicals may have the same cumulative permeation after a set of exposure time, they may have different BTs. This information is useful in the selection of the correct type of gloves for hand protection.

However, breakthrough for the skin does not rely on the flux of the tested substances. As soon as the substances were proven to penetrate the skin regardless of the rate of penetration, it is considered a breakthrough.
PEER-REVIEWED PUBLICATIONS (to date of submission)


CONFERENCE AND PRESENTATIONS (to date of submission)

Dermal absorption of organophosphorus insecticides: effects of concentration and temperature on skin penetration of omethoate

- Abstract and Oral presentation
- 33rd Australian Institute of Occupational Hygienists AIOH Conference and Exhibition, Perth, Western Australia, 5th to 9th December 2015

Do concentration and temperature affect skin penetration of omethoate?

- Abstract and Poster presentation

The effects of temperature, ultraviolet (UV) and abrasion on the performance of gloves used by agricultural workers handling Organophosphate Pesticides (OPs)

- Abstract and Poster presentation
- 31st International Congress on Occupational Health, Seoul, South Korea, 31st May to 6th June 2015

Dermal exposures to Organophosphorus Pesticides for ambulance workers - permeation through disposable gloves: Findings for omethoate.

- Abstract and Poster presentation
- 32nd Australian Institute of Occupational Hygienists AIOH Conference and Exhibition, Melbourne, Australia, 30th November to 3rd December 2014

xxviii
Permeation of organophosphate pesticides through disposable gloves:
Are they protective enough for ambulance workers?

- Abstract and Poster presentation
- 2014 Florey International Postgraduate Research Conference, Adelaide, South Australia, 25th September 2014

AWARDS

- High Commendation (runner-up) for Academic Excellence in Postgraduate Research at the 2015 Study Adelaide International Student Awards, Adelaide, South Australia.
- Merdeka Award 2015 from Australia-Malaysia Business Council (South Australia) Inc. for outstanding academic achievement, ability to act as an ambassador for South Australia and contribution to the furthering of the South Australia-Malaysia relationship
- 1st place Student Poster Award for the best poster at the 31st International Congress on Occupational Health, Seoul, South Korea
- School of Population Health Prize for the best poster at the 2014 Florey International Postgraduate Research Conference, Adelaide, South Australia
THESIS OVERVIEW

The purpose of this overview is to assist the reader in rapidly appraising the content and key messages of individual chapters of the thesis. Structurally, this thesis has a traditional chapter format with a combination of introduction, literature review, research questions, methodology, empirical research findings, discussion, conclusions and recommendations.

It utilises a case study approach, informing laboratory experimentation, to enable a better understanding of dermal exposure to organophosphorus pesticides (OPs) in two settings; namely routine agricultural use and emergency ambulance response.

This thesis is comprised of eight chapters, with content and messages tabulated as follows:

<table>
<thead>
<tr>
<th>Chapter</th>
<th>Chapter content</th>
<th>Key messages</th>
</tr>
</thead>
<tbody>
<tr>
<td>1- Introduction and context</td>
<td>This chapter provides an overview of the working populations potentially exposed to organophosphorus pesticides (OPs). The public health and national security significance of organophosphorus pesticides in various settings are briefly described.</td>
<td>Large numbers of workers may be exposed to organophosphorus pesticides in manufacturing, storage, transport, use and disposal as well as in emergency situations. Significant mortality is evident in developing countries. Twelve of the 96 chemicals of security concern are organophosphorus pesticides used in Australian agriculture. The need for a review of these 12 chemicals and the issues associated with dermal exposure, e.g. work in hot weather, are highlighted.</td>
</tr>
<tr>
<td>2- Literature review</td>
<td>The objectives for the review are described and the search strategy is presented. The search yield is summarised to identify the knowledge gaps pertaining to dermal exposure and glove protection. A rationale for the selection of OPs and gloves for the proposed research is given. The aims and broad research</td>
<td>The dermal exposure assessment literature on the 12 OPs of security concern is sparse. A number of research opportunities were evident. It was decided that the broad research questions would relate to skin protection and uptake for a range of formulated OPs under a range of under-explored conditions, e.g.</td>
</tr>
</tbody>
</table>
questions are derived from the literature review, particularly in relation to issues of formulated OPs, UV exposure and abrasion. Out-of-scope research aspects are defined. elevated temperature and glove abrasion. It was decided that four OPs of varying physicochemical properties would be examined in an exploratory manner.

<table>
<thead>
<tr>
<th>3- Materials and methods</th>
</tr>
</thead>
<tbody>
<tr>
<td>Materials used and methods in the glove performance tests and <em>in vitro</em> skin studies are explained in detail. Statistical analyses used for experimental data are also described.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>4- Case Study 1: Protective performance of Polyvinyl Chloride (PVC) gloves used by agricultural workers</th>
</tr>
</thead>
<tbody>
<tr>
<td>This case study explores the performance of PVC gloves used by agricultural workers against formulated OPs under variable conditions of temperature, concentration, UV exposure and abrasion.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>5- Case Study 2: Protective performance of disposable gloves used by ambulance workers</th>
</tr>
</thead>
<tbody>
<tr>
<td>In this case study, disposable gloves used by South Australia Ambulance Service (SAAS) workers were challenged with OPs. Gloves are tested individually and in combination (as currently in practice) under reasonable worst case conditions.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>6- Skin penetration studies of omethoate, mevinphos, dichlorvos and diazinon</th>
</tr>
</thead>
<tbody>
<tr>
<td>Skin penetration of four formulated was assessed using an <em>in vitro</em> approach, at different concentrations under two temperature conditions. Extrapolated, worst case penetration data were compared with acceptable daily intakes.</td>
</tr>
</tbody>
</table>
The specific content and orientation of each chapter is outlined as follows:

**Chapter 1 General Introduction**

It is argued that the extensive use of organophosphorus pesticides worldwide is a significant public health issue. Evidence is provided for large scale health impacts in developing countries. Given the availability, toxicity and possible misuse of OPs, the Australian government has classified some OPs as Chemicals of Security Concern. On the basis of ongoing impact and the national concern, the general topic of OPs was considered worthy of further research.

It is well known that skin absorption is a major route of exposure of OPs, and it is self-evident that a good understanding of both skin penetration and the effectiveness of skin protection is needed. However, this knowledge needs to be contextualised in real world conditions, where formulated products rather than pure active ingredients are used, and where work may be conducted in hot environments.
In order to provide a focus, the topic (and subsequent literature review) is limited to those OPs of security concern and the occupational hygiene issues associated with dermal exposure.

Chapter 2 Literature Review

This chapter rationalises the development of the research questions. It starts with the objectives of the review, followed by the search strategy and a tabulation of the yield for each of the 12 OPs of security concern. A cross-cutting narrative review of existing dermal exposure-related literature is undertaken, highlighting the limited number, and types, of published glove permeation and skin penetration studies. The gaps in knowledge are summarised, justifying the aims and broad research questions. A case study approach, based on two real world situations, is proposed for subsequent research. A set of complementary glove and skin experiments, using a selected sample of formulated OPs, would allow exploration of the impact of high temperatures and practical issues such as glove abrasion and sunlight exposure. Finally, the scope of the proposed research is delineated.

Chapter 3 Materials and Methods

The test chemicals, equipment, instruments and methods used are described in this chapter. The arrangements for glove permeation testing according to Australian/New Zealand Standard 2161.10.3:2005 are outlined, along with a description of the PVC, nitrile and neoprene gloves, and formulated OPs under investigation. Details of the human skin samples, and the procedures for skin penetration testing, according to an OECD protocol are presented. A UV exposure chamber is described, along with a technique for glove abrasion. HPLC equipment and conditions for chemical analysis are presented.
Finally, the statistical tests used in the research are outlined. Apart from the glove abrasion and UV-exposure techniques, these represent standardised methods.

Chapter 4 Case Study 1: Protective performance of PVC gloves used by agricultural workers against OPs

Unused gloves were subjected to realistic worst case conditions, up to 8-hour duration. The resultant glove permeation data indicate that PVC gloves provide good protection against the four tested formulated OPs. When challenged with concentrated OPs, 8-hour cumulative chemical permeation is increased beyond that observed for application strength OP, especially under elevated temperature conditions. There is some variation between two brands of ostensibly the same glove. Reduced protective performance is noted for PVC gloves subjected to UV radiation (approximately equivalent to one continuous week of extreme sunlight) and abrasion (5% reduction in overall glove thickness, and 10% of the PVC coating). Statistically significant differences were noted for effects of concentration and temperature. The effects of UV and abrasion on gloves were relatively minor. The findings are discussed in terms of complementary research previously conducted in Tasmania.

Chapter 5 Case Study 2: Protective performance of disposable gloves used by ambulance workers against OPs

This case study simulates worst case OP dermal exposures for ambulance workers handling accidental or intentional poisoning patients. The permeation resistance of disposable nitrile and neoprene gloves used by SA Ambulance Service workers is examined. Cumulative permeation increased significantly with concentration and temperature. However, the data show that the current practice of double gloving by South Australian ambulance workers offers good short term protection against the tested formulated OPs. Wearing a combination of gloves is better than
individual gloves, and this may be attributed to the thickness, and the relative polarities of OPs and the glove materials. Consideration should be given on frequent changing of gloves when exposed to concentrated OPs under elevated temperature conditions.

**Chapter 6 In vitro skin studies of omethoate, mevinphos, dichlorvos and diazinon**

This Chapter reports on *in vitro* human skin penetration studies conducted with four OPs under conditions of variable concentration (application vs full strength OP) and temperature (room temperature to 37°C). An infinite dose arrangement was used with exposures up to 8 hours. Statistically significant increases were noted with increased concentration and temperature, and some significant differences between the formulated pesticides were also observed. Comparisons of extrapolated cumulative penetration with Acceptable Daily Intakes indicate rapid exceedances when concentrated OPs are handled with unprotected hands.

**Chapter 7 General Discussion**

This chapter begins by emphasising the novelty and significance of the research. Unlike most glove permeation and skin penetration studies this research used formulated OP products that are commercially available in the market. All experiments were conducted using an infinite dose model to simulate the worst exposure scenario (prolonged skin contamination from spills and splashes), and thus to establish the maximum penetration rates for risk assessment. By combining glove and skin studies, a more complete picture of dermal exposure is available. The performance of the gloves was not only tested in new (unused) condition, but also after exposure to UV radiation and abrasion to reflect the common condition of gloves being used by agricultural workers. The effect of elevated temperature was shown to be important for both gloves and skin and is discussed in a real world context. Four OPs with differing physicochemical properties were
selected for comparison of glove permeation and skin penetration. The skin data can potentially be used for predictive dermal risk assessment models, although no simple trends between OPs were discerned.

The generalisability of the research outcomes is discussed, and the chapter is completed with a discussion of the strength and limitations of the overall research.

**Chapter 8 Conclusion and Recommendations**

The data indicate that the use of concentrated OPs in warmer conditions will greatly increase skin uptake. Toxicologically important uptake can occur in relatively short time periods without skin protection. Glove permeation also increases with concentration, temperature, UV and abrasion, and this needs to be better understood by users. The practice of double gloving by ambulance workers is supported by the evidence. Recommendations are made for future researchers, the glove users, manufacturers and suppliers of OPs and gloves as well as regulatory and enforcement bodies.

**Broad Research Questions**

The following questions were developed from the literature review and from preliminary field observations.

- How do the recommended PVC gloves (unused, exposed to UV and abrasion) perform against OPs in various exposure conditions?
- What are the effects of different variable conditions on the barrier performance of different types of disposable gloves worn by ambulance workers and is the current practice suitable?
How is skin penetration affected by physicochemical properties of OPs, concentration of OPs and elevated temperature?

Specific Research Questions

RQ1 Are the recommended PVC gloves suitable for protection against formulated OPs?
RQ2 What are the effects of UV radiation on gloves with respect to formulated OP permeation?
RQ3 How does abrasion of the PVC glove materials affect permeation of formulated OPs?
RQ4 What are the effects of variable experimental conditions (diluted and undiluted concentration, room temperature and hot conditions) on the protection provided by the disposable gloves used by ambulance workers?
RQ5 How do individual gloves perform in terms of protection afforded, as compared to when gloves worn in combination?
RQ6 To what extent are the workers protected when implementing double gloving (combination of nitrile and neoprene gloves) as currently in practice?
RQ7 How do skin penetration outcomes differ between formulated OPs?
RQ8 How does the concentration of OP formulations and an elevated temperature affect skin penetration?
RQ9 How does the amount of OPs penetrated through the skin compare to the respective Acceptable Daily Intake (ADI)?