NOTICE

The quality of this microform is heavily dependent upon the quality of the original thesis submitted for microfilming. Every effort has been made to ensure the highest quality of reproduction possible.

If pages are missing, contact the university which granted the degree.

Some pages may have indistinct print especially if the original pages were typed with a poor typewriter ribbon or if the university sent us an inferior photocopy.

Reproduction in full or in part of this microform is governed by the Canadian Copyright Act, R.S.C. 1970, c. C-30, and subsequent amendments.

AVIS

La qualité de cette microforme dépend grandement de la qualité de la thèse soumise au microfilmage. Nous avons tout fait pour assurer une qualité supérieure de reproduction.

S'il manque des pages, veuillez communiquer avec l'université qui a conféré le grade.

La qualité d'impression de certaines pages peut laisser à désirer, surtout si les pages originales ont été dactylographiées à l'aide d'un ruban usé ou si l'université nous a fait parvenir une photocopie de qualité inférieure.

La reproduction, même partielle, de cette microforme est soumise à la Loi canadienne sur le droit d'auteur, SRC 1970, c. C-30, et ses amendements subséquents.

THE UNIVERSITY OF ALBERTA

THE EFFECT OF DICHLORVOS RESIN STRIPS ON WOOL FIBERS

BY

SHARON HAMMICK

A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES AND RESEARCH

IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF

Master of Science

Department of Clothing and Textiles

EDMONTON, ALBERTA

SPRING, 1989

ISBN 0-315-52826-5

THE UNIVERSITY OF ALBERTA

RELEASE FORM

NAME OF AUTHOR: Sharon Hammick

TITLE OF THESIS. The Effect of Dichlorvos Resin Strips on Wool Fibers

DEGREE: Master of Science

YEAR THIS DEGREE GRANTED: Spring 1989

Permission is hereby granted to THE UNIVERSITY OF ALBERTA LIBRARY to reproduce single copies of this thesis and to lend or sell such copies for private, scholarly or scientific research purposes only.

The author reserves other publication rights, and neither the thesis nor extensive extracts from it may be printed or otherwise reproduced without the author's written permission.

(SIGNED) Sharon Hammick

PERMANENT ADDRESS:

5716 Lac Marie

Edmonton, AB

DATE: 26.9.88

THE UNIVERSITY OF ALBERTA

FACULTY OF GRADUATE STUDIES AND RESEARCH

The undersigned certify that they have read, and recommend to the Faculty of Graduate Studies and Research for acceptance, a thesis entitled The Effect of Dichlorvos Resin Strips on Wool Fibers submitted by Sharon Hammick in partial fulfilment of the requirements for the degree of Master of Science in Clothing and Textiles.

Nancy Kerr
.....
Dr. Nancy Kerr
Supervisor

.....
Dr. W. G. Evans

Elizabeth K. Richards
.....
Professor E. Richards

Date: 16.01.89.....

ABSTRACT

Dichlorvos (2,2-dichlorovinyl dimethyl phosphate), marketed under the trade names, Vapona® or S.W.A.T.®, is a time released insecticide commonly used for continuous fumigation in museum and natural history collections. The suitability of dichlorvos for space fumigation was investigated. Wool fibers were fumigated with dichlorvos resin strip (S.W.A.T.®) at 50°C for a maximum of 35 days to determine the effect of dichlorvos on the morphology, and physical-chemical properties of the fibers.

Wool fibers exposed to high concentrations of dichlorvos resin strip exhibited physical and chemical changes such as binding of phosphorus and chlorine, yellowing due to chlorine absorption, decrease in pH, and acid and oxidative damage. In tensile tests, elongation initially increased. Fumigation at 50°C accelerated the rate of chemical and physical degradation compared to ambient temperature fumigation. Similiar observations of degradation were found in historic textiles exposed to continuous fumigation in a museum collection.

The critical factor in accelerated fiber degradation due to exposure to dichlorvos appeared to be the removal of tightly bound lanolin wax with methanol. Scoured fibers given a methanol rinse

degraded more rapidly than fibers cleaned with detergent only, probably because of increased chlorine absorption.

ACKNOWLEDGEMENTS

My special gratitude and appreciation is foremost to Dr. W. G. Evans of the Department of Entomology, for his keen interest, research expertise, unswerving encouragement, technical guidance, and emotional support from the inception to the completion of my thesis. Special appreciation is extended to Georg Brook and Dave Hildebrandt from the Entomology SEM Laboratory for the hours devoted to overcoming technical difficulties and developing an efficient system for EDX analysis of keratin fibers. My thanks is extended to Dr. G. E. Ball, Department of Entomology, who heightened my respect and admiration of the diverse and fascinating *hexapods*.

Thank-you Elaine Bitner, Jane Good, and Dr. Kathy Rigakis, from the Department of Clothing and Textiles, for providing technical support, sharing equipment, and providing a protective coverall to augment my *Superstore* regalia. Thank-you Linda Turner, and Janice Smith for technical assistance.

I am indebted to Dr. John Buckland-Nicks, Department of Zoology, and Ron Koss for providing me with assistance with fluorescent microscopy, and to Barry Mandrake, Histology, for mounting medium and equipment for slide preparation. Thank-you Dr. Cass, Department of Botany, for demonstrating mounting techniques and providing me with the use of equipment.

My appreciation is extended to Sherri, Eric and Bill, from the Edmonton collection for providing a history of fumigation in

the Edmonton Artifact Center and for extended access to the collection. I wish to extend my appreciation to Mary-Lou Florian, Conservation Scientist, British Columbia Provincial Museum, for her assistance and expertise in integrated pest management. Thank-you, S. L.Williams, Carnegie Museum, for sharing your research findings, and Greg Young, Canadian Conservation Institute, Ottawa, for the examination and report on a leather artifact damaged by Vapona® during storage in the Edmonton collection.

Thank-you to the many people in the Department of Chemistry for supplying small quantities of chemicals and advice on the efficacy and parameters of analytical techniques. I wish to acknowledge the generous donation of fumigant from Ciba-Giegy, and technical information from Occupational Safety networks.

Without the generous and devoted assistance from the librarians, whose skills at researching obscure documents enabled an extended literature review, I would not have been able to integrate so much information from diverse sources.

It is imperative to acknowledge my daughter Kathleen whose sacrifice and contribution to my research enabled this research. My deep appreciation is shared with my daughter Ellen who shared the responsibility of guardian and caregiver for Kathleen during my absence.

Thank-you Frank for your computer assistance, programming, companionship and support. My graduate days were enriched with the

friendship and mutual support of Heather Prince, Edith Francois, and Michael Marendy.

Finally I wish to extend my graditude to my supervisor Dr. Nancy Kerr whose diligent and conscientious lectures were well appreciated. Thank-you Dr. Kerr for allowing me to work unobtrusively on my research and giving me the opportunity to explore. The hours we spent editing massive volumes of text and translating wool science terminology into common terminology will be well appreciated by others. I hope my exploratory research will provide you and others with an information source for further studies in biodegradation and pest management. Thank-you Elizabeth Richards for suggestions in editing this thesis.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

1. INTRODUCTION

1.1 Problem Statement

Museums and cultural depositories have amassed vast storehouses of textiles and furs for insect species to devour. Wool and furs are a desirable banquet for keratin feeding insects. Curators and conservators whose responsibility is to care for culturally significant objects are well aware of the rampant destruction caused by an infestation of keratin feeding insects. The conflict between insects and the preservation of cultural resources has resulted in indiscriminant use of chemicals often to the detriment of the artifact (Moncrieff, 1950; McPhee, 1971).

Dichlorvos resin strips such as Vapona® are one of the most popular chemical weapons used in museums and historic sites for insect control. Curators and conservators have found that Vapona® is a convenient long-term fumigant. Vapona® strips are used for continuous fumigation in textile and natural history collections to prevent infestations of keratin feeding species. The effects of the emitted chemicals on keratin objects such as wool fibers are unknown.

Many objects in ancient and historic textile collections are made of wool and other keratin fibers. Wool, alpaca, vicuna, angora cashmere, mohair, and furs are keratin fibers commonly found in ancient, historic, ethnographic, and natural history collections.

Dichlorvos is commonly used to protect objects composed of wool and fur from insect attack. Since the effects of dichlorvos on keratin fibers have not been thoroughly explored to preclude possible adverse consequences, an investigation of the physical and chemical changes in wool, the most common keratin fiber, is both timely and urgent.

It is the conservator's responsibility to assure that treatments are appropriate to the preservation and best interest of the object. The conservator must avoid materials that endanger the safety and integrity of the object. Since there is some evidence that dichlorvos space fumigation damages textiles the effects of dichlorvos on wool fibers are in need of investigation.

The effect of dichlorvos on textile fibers has not been answered by researchers. The focus of this research is to investigate the effects of dichlorvos space fumigation on wool fibers.

1.2 Objectives

The primary objective of this research is to investigate the damage to wool exposed to dichlorvos resin strips under controlled laboratory conditions. The objectives of the literature review are:

1. To review the literature to establish the extent of reported dichlorvos use in cultural depositories.
2. To characterize dichlorvos resin strips including by-products of decomposition.

3. To survey the literature to determine the known effects of dichlorvos on textiles.
4. To assess the toxicity of dichlorvos to Homo *sapiens*.
5. To survey wool science (keratin fiber) literature related to the molecular structure, fiber structure, fiber morphology, histochemistry, chemical reactivity and mechanical properties of wool (keratin) fibers.

The main objective of the laboratory research is:

To determine if dichlorvos space fumigation changes or damages wool fibers.

The specific laboratory objectives are identified as:

1. To determine changes in the tensile properties of fumigated wool yarn.
2. To determine color change in wool fibers after fumigation.
3. To determine the effect of fumigation on sensitive cystine residues.
4. To determine if the fumigant is sorbed and desorbed by wool fibers.
5. To identify changes in the cuticle of wool fibers fumigated with dichlorvos resin strips.
6. To determine changes in the pH of the aqueous extract of fibers after fumigation.
7. To determine the relationship between the pH of the aqueous

extract and degradation of wool fibers.

8. To determine the effect of dichlorvos fumigation on the absorption of acid and basic dyes.

1.3. Assumptions

1. The chemical and physical changes observed in wool fibers are representative of changes in keratin fibers fumigated with dichlorvos resin strips in museums and cultural institutions.

1.4. Limitations

1. Only one patented dichlorvos strip (S.W.A.T.®) was used in this study.
2. Greasy wool fibers, which were scoured in a mild detergent or scoured in a mild detergent and soaked in methanol, were used for all tests except tensile properties. Ancient and historic wool textiles and furs have a complex history; therefore, the effects of the fumigant may be different.
3. The use of heat and high concentrations of fumigant to accelerate fumigation may induce different physical and chemical effects than ambient temperature fumigation using concentrations recommended by manufacturers.
4. Most test methods used in this study were designed for estimating severe damage during industrial processing. Their use in this study is exploratory.

5. The wool science literature reports basic and applied research Characterization and identification of damage after fumigation were based on laboratory experiments and theories from industrial processes and basic research. The chemicals in basic and applied research and in the wool science literature are different from the assumed degradation products emitted from the dichlorvos resin strips.

6. The relative humidity in this study was not controlled. Moisture can have profound effects on the chemical reactivity and physical properties of wool fibers and on the reactivity of dichlorvos resin strips.

2. LITERATURE REVIEW

2.1 Museum Fumigation

2.1.1 History of fumigation

Long before Matthew (circa 40 AD) wrote "Do not store up riches for yourselves on earth where moth and rust destroy", keratin feeding insects had fully established themselves in an ecological recycling niche (Holy Bible, 1977). Throughout history fumigants and deterrents have been used to protect textiles (Moncrieff, 1950). In 1735 Father Jean-Baptiste Du Halde wrote in the *Description of the Empire of China and of Chinese Tartary* that Chinese workmen melted seal fat to drive away the the flies "because these insects settle on the woven silk and damage it very considerably" (Bussagli, 1980). Camphor and cedar oil have been used to deter insects (Plenderleith & Werner, 1971). Plenderleith advocated "p-dichlorobenzene in the textile bag to ensure against the risk of moth eggs being present" (Hueck, 1972).

In 1952, the Textile Museum in Washington, D.C. issued a paper on rug preservation and storage. Annual fumigation directions included sprinkling the face of a 4' by 6' rug with approximately 1/2 lb. of par-di-chloride-benzene (sic) flakes, and raising the temperature of the storage area to between 90°F and 100°F for three days. Textiles to be fumigated were sealed in chests with 1 pound of

flakes to 10 cubic feet (Myers, 195?). In 1973 thymol crystals in a saucer over a 40-watt light bulb in an air-tight compartment were being recommended for fumigating museum textile collections (Fall, 1973).

2.1.2 Fumigation trends

By 1972 "an astonishing number of insecticides" were available (Hueck, 1972; Grayson, 1978). Hueck (1972) writing in *Textile Conservation* suggests the use of the insecticides p-dichlorobenzene, ethylene oxide, and the "more modern DDVP (dichlorvos)" which are "active as a gas" (Hueck, 1972). Hueck writes, "DDVP may well prove to be useful. Its high activity makes hermetically closed containers unnecessary". Incorporated in resins it gives "protection for some months' duration" (Hueck, 1972). Commercially prepared DDVP impregnated paper inserted between individual pieces of fabric "with constant renewal" is suggested as a preventative measure (Hueck, 1972). Rice (1962) advocates ethylene oxide applied to textiles "packed in polyethylene bags" as an effective fumigant (Heuck, 1973). In 1969, Rice stated that ethylene oxide "a modern space fumigant" is "much less toxic to human beings than many other fumigants" and "is a very good fumigant because it leaves no bad after-effects" (Rice, 1969).

Vapona® for the control of museum pests came into vogue when Ryckmann (1969) reported the efficacy of Vapona® as a control for

dermestids. Ryckmannn reported that Vapona® resin strips at concentrations equal to 32 times the manufacturer's recommendations, "quickly curtailed the destruction of museum specimens". Although Vapona® resin strips "tend to sweat", a piece of aluminum and "seve al thickness of blotter paper" prevent staining (Ryckmann, 1969). Ryckmann reported that "human subjects exposed continuously to Vapona® resin strips at 10 times the recommended dose showed no cholinesterase depression" (Ryckmann, 1969).

Bogle, a conservator at the Merrimack Valley Textile Museum, recommended the use of p-dichlorobenzene at the rate of "one pound of moth crystals per 100 cubic feet" in an air-tight area, and the use of dichlorvos pest strips for the control of moths, dermestids, and flying insects (Bogle & Hueck, 1979). During the UNESCO regional conference, *The Conservation of Cultural Materials in Humid Climates, 1979*, in Australia, conservators proposed that museums in the Cook Islands, Malaysia, the Philippines, Thailand, New Zealand and other Pacific rim countries use Vapona strips to control pests (UNESCO, 1979). Dichlorvos is a recommended fumigant at the Indian Government Museum in Madras (Jeyaraj, 1983). At the 1980 International Conference, *Conservation and Restoration of Textiles* in Como, Vigo presented a paper suggesting microencapsulation of textiles in plastic bags, and the use of dichlorvos as a fumigant (Vigo, 1980). Fumigants enclosed in polyethylene bags with textiles are also recommended by Lawson in *Textiles, Primary Care and First Aid* (nd).

At the ICOM (1984) Copenhagen *Committee for Conservation 7th Triennial Meeting*, Armes reports that "dichlorvos slow-release units...have been used successfully in museums for a number of years" (Armes, 1984). In spite of the effectiveness and benefits many museums were reluctant to use dichlorvos because of "its corrosive properties towards metals and staining of specimens" (Armes, 1984).

Many museums continue to use and advocate DDVP as a convenient and effective fumigant. In the 1981 edition of *Conservation and Restoration for Small Museums*, the use of DDVP blocks in a sealed cabinet is suggested as an alternative to the preferred ethylene oxide fumigation chamber. The Western Australian Museum suggests that after initial fumigation, furs and silk should be wrapped in acid-free tissue, and ethnographic material placed in polyethene bags before placing the objects in containers with a DDVP vapour block in the lid (Pretty,1981). Ward (1976) advocates the use of fumigation boxes with dichlorovos impregnated strips in the lid. Although dichlorvos is recognized as "extremely dangerous and (has) caused serious illness and some fatalities...(it) is a pity because (the strips) are undoubtedly effective" (Ward, 1976). Ward proposes the use of DDVP strips in dioramas containing mounted mammals and birds, and for closets containing woolen clothing (Ward, 1976).

In Britain, Vapona® is used by the British National Museum and a number of provincial museums (Stanfield, 1985). Dichlorvos is "recommended in recent Museums Association Diploma practicals", in

spite of the observation that some fading of pigments of green colored moths occurs indicating need for further research (Stanfield, 1985). Hey (1980-81) writing in the British journal *The Paper Conservator* lists dichlorvos (Vapona®) as an insecticide recommended for fumigation in conservation and archives.

A comprehensive survey of deleterious insect species found in museums throughout the world and by Alam (1983) in *The Journal of Indian Museums* (Bombay) includes fumigants and insecticides for use in pest management. Alam, quoting Hueck (1972), reiterates the effectiveness of DDVP fumigation, and advocates interleaving textiles with commercially DDVP impregnated paper (Alam, 1983). Impregnated DDVP paper "should be inserted between individual pieces of fabric and renewed after six to nine months" (Alam, 1983, p152).

2.1.3 Hazards of dichlorvos fumigation

Some fumigators have circulated warnings about the possible adverse effects of fumigants. Westrate (1984), a pest consultant and philatelist, cautions that sprays, aerosols and toxicants can stain, discolor and adversely affect collections, and says that "An ounce of prevention is worth a pound of pesticide". Pest control publications circulated to museums indicate that care must be exercised when applying fumigants to avoid staining clothes, walls, or furniture (Edwards *et al.*, 1981).

In 1955 sheep growers began dipping sheep in dieldrin to

prevent an ectoparasite from damaging fleeces (Katz, 1981). Since dieldrin has an affinity for wool and forms a permanent molecular bond to the fibers, wool was inadvertently mothproofed. When dieldrin was banned, the fecundity of insects presented problems in collections (Katz, 1981). With more food, better shelter, higher humidity and warmth, and the withdrawal from the market of persistent chlorinated hydrocarbons, fabric pests increased (Katz, 1981).

The persistence of chlorinated hydrocarbon insecticides was becoming a problem (G 1984; Lave, 1987; Larose, 1955; Laughlin & Gold, 1987, ne (an organochlorine compound) and diazinon (an organophosph us compound) were used to control the sheep blowfly, *Lucilia cuprina* (McKenzie & Whitten, 1984). Levels of about 130 ppm have been detected in pharmaceutical lanolin (Warner, 1980). Removal and separation of diazinon, an undesirable component in wool grease, is a problem in industry (Warner, 1980). The removal of dichlorvos from wool fibers (See Section 6.11) appears to be a problem confronting museums (Glastrup, 1987; Kowalik, 1979; 1980).

During 1984-1985 the Danish National Museum conducted pesticide residue analyses on samples taken from objects and exhibition cases. By using gas chromatography, seven different insecticides were identified. Although "erroneous peaks (noise) on the chromatograms...around lindane, aldrin and dieldrin" hampered analysis, levels as high as 43% (p-diclorobenzene) for wool and duck down are reported (Glastrup, 1987). Duck down and woven wool, both

keratins, absorbed significant quantities of insecticides. A 1971 report on Danish textile pest control indicates that satisfactory protection against *Dermestidae*, especially *Attagenus piceus* (carpet beetle) was difficult to obtain with dichlorvos (Funder, 1972; Markkula, 1973).

In 1981 two major reports were published, *Safety and Health in the Paper Conservation Laboratory*, and *Pest Control in Museums*. The former publication is an extensive compendium outlining the basic concepts of toxicology, including the physiological effects of substances which gain entry to the human body through inhalation, ingestion and absorption through the skin (Harrington & Petherbridge, 1980-81). Harrington, a professor at the Institute of Occupational Health, Medical School in Birmingham, utilized his expertise to compile chemical and safety data, and first aid procedures for toxic chemicals used in conservation. Dichlorvos was the most toxic chemical. In the same report pesticide regulations, the use of protective clothing, equipment for handling chemicals and fumigants, and maintenance procedures are discussed (Harrington & Petherbridge, 1980-81). In America, an increasing awareness of the potential dangers of pesticides resulted in a coordinated status report on pest control in museums. Survey results indicate that Vapona® is used by 40 per cent of the museums surveyed.

Many museum attendants have not recognized that handling artifacts fumigated with DDVP may be hazardous. In 1977 the Carnegie

Museum published a guide for DDVP fumigation warning that the volatile insecticide evolves highly toxic vapors and that skin exposure should be avoided. Gunter *et al.* (1977) in *Pesticide Residues* reports that "sebum covers and permeates the ...stratum corneum...and has a high affinity for lipid-soluble substances and thus is ideal for the aquisition of most toxic pesticide residues". Since the average adult skin area is about 17,000 cm^2, dermal exposure poses a threat. Observations that residual organophosphates on crops may be transferred to exposed and covered parts of the body are of concern since an analogue may exist with museum staff who handle or are exposed to contaminated objects. Unusually high cholinesterase depression in agricultural workers is attributed to dermal exposure from an accumulation of residues in dew dampened foliage. Pesticide residues are transferred to the skin via damp clothing (Gunter *et al.*, 1977). (See section 2.6)

In 1987 the Center for Occupational Hazard published an article on museum pest control procedures including a survey of museum staff which "clearly indicates that museum staff had extremely limited knowledge of the hazards, precautions, and regulations" (Peltz & Rossol, 1987). Of the 27 museum staff who used fumigants, only 7 workers used any protective equipment and only 1 person used the proper type. None of the ethylene oxide users checked for residual fumigants or leaks (Peltz & Rossol, 1987). The report indicates that many chemicals can destroy or damage objects. The

damage is "evident only days, months, or years after treatment". Dichlorvos is listed as "acting as an acid" in humid climates, causing plastics to dissolve, and metals to corrode (Peltz & Rossol, 1987).

Most fumigants are listed as designated priority pollutants (Coughtrey *et al.*, 1987; Frear, 1976). Priority pollutants are extremely hazardous chemicals. Williams *et al.* (nd) report that dichlorvos is readily absorbed on most plastics, metals, furs, cotton, burlap, paper, fiber, wood products. It also corrodes some metals, causes some resins and glues to become tacky, dissolves polystyrene plastics, and will affect baked enamel paint surfaces and aluminum. Dichlorvos is known to be corrosive to iron, steel, brass, silver, tin, and lead (Spencer, 1981; [illegible]ry, 1985).

A recent status repo[illegible]inting out the ignorance of museum staff, asks if fumigation helps or harms the artifact (McGiffin, 1985). McGiffin (1985) reports that Vapona® corrodes metals and *bleeds* on other items. Shrinkage temperature tests on leather conducted at the Canadian Conservation Institute (CCI) indicate that Vapona® may cause degradation (Young, 1987). A task force from the Canadian Conservation Institute, Smithsonian Institute, Dow Chemical Company, and the Getty Conservation Institute has been set up to examine fumigants (GCI, 1986). Some conservators are asking "is fumigation possible" (Ballard & Baer, 1986) and a minority are searching for alternative methods (Florian, 1986). The problem is

fundamental. Too little knowledge of the nature and potential dangers of pesticides exists within the museum community and scientific community.

2.1.4 Effects of Dichlorvos on Textiles

Only a few studies have reported the effects of dichlorvos on textiles or textile fibers. A brief list of experimental reports is given in Table 2.2.1.

Table 2.1.1 Studies Investigating the Effects of Dichlorvos on Textiles or Fibers.

Textile	Observations	Source
Silk cocoons	yellowing	Nakamoto, 1984, Japan Sericulture
Burlap/cotton sacking	sorption	McGaughey, 1973 Rice mill
Cotton hospital garments	sorption	Cavagna, 1969 Italian Hospital
Polyester/nylon carpets yarns	dye changes	Reagan *et al.*, 1984 Laboratory tests
Wool and cotton test fabric	increased tensile strength, and slight dye change	Spivak *et al.*, 1981 Laboratory tests

Several earlier research projects indicate that residues may

accumulate on textiles. The frequently cited Cavagna report (1969), conducted in Italian hospital wards, includes an experiment examining the effect on red blood cell cholinesterase when DDVP impregnated garments were worn by babies. The garments were aerated before use and apparently desorbed some of the pesticide. The babies garments were initially reported to retain 0.26μg/gm DDVP on cotton and 0.40μg/gm DDVP on cotton flannel, after exposure to one Vapona Ministrip® in a 200 litre capacity cupboard (Cavagna, 1969). Another report from a rice mill indicates that residues on bagging material increased from 136.7 $\mu g/dm^2$ to 662.2$\mu g/dm^2$ DDVP on paper exposed for 1-13 treatments, and ranged from a low of 245.4$\mu g/dm^2$ to a high of 3753.4$\mu g/dm^2$ DDVP on burlap. The apparent anomaly on cotton, which ranged from 3.2$\mu g/dm^2$ with 1 exposure to 531.8$\mu g/dm^2$ with 13 exposures was a consequence of room size and concentration (McGaughey, 1973). Lellinger (1972) suggests that dichlorvos may crystallize on surfaces.

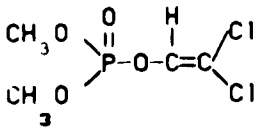
Preliminary studies to assess the effects of pesticides on textiles were initiated by Spivak *et al.* (1981). Wool and cotton samples were suspended in a museum case with Vapona® for a period of four weeks. Changes in the tensile strength and colour of the dyed fabrics were measured. The results indicate that a slight change in madder root dye occurred and that the tensile strength of wool samples was "inexplicably higher in some cases" (Spivak *et al.*, 1981). A Carpet Manufacturer Association reported that dichlorvos

can cause color change and fading in some dyestuffs (Reagan, 1982). Another report (1983) indicates that DDVP creates an unacceptable color change in some dyes. The committee investigating color change in dyes reported that the future of the problem lies with the pesticide industry. In 1984 Reagan, with a committee investigating the effect of insecticides on acid and disperse dyes on nylon and polyester carpets, found that dichlorvos caused some color changes. As a result of the findings the committee recommends that dye and carpet manufacturers select dyes less sensitive to insecticial chemicals (Reagan *et al.*, 1984).

2.2. DICHLORVOS

2.2.1 Introduction to dichlorvos

Although dichlorvos (O,O-dimethyl 2,2-divinyl phosphate, DDVP) was first synthesized in the late 1940's, active insecticidal investigation was not initiated until 1954 (Zavon & Kindel, 1966). Dichlorvos was first discovered as an insecticidal impurity of the organophosphate insecticide trichlorfon in 1955 (Eto, 1974). Above pH 6 trichlorfon is rapidly converted to dichlorvos (Eto, 1974). About 7 million pounds of dichlorvos manufactured each year by Amvac Chemicals, Los Angeles, and Chemagro Chemicals, Commerce City, CO., from trimethyl phosphite and chloral by the Perkow reaction (Eto, 1974, Santodonato, 1985).



O,O-dimethyl 2,2-dichlorovinyl phosphate

Synonyms and trade names include: DDVP (USA), DDVF (USSR), dichlorvos, *Vapona*®, *Nogos*®, *Nuvan*®, *Dedevap*®, *Diptera*®, *Astrobot*®, *Canogard*®, *Divipan*®, *Mafu*®, *Cekusan*®, *Herkal*®, *Marvex*®, (Gillett, 1972; Merck Index, 1983; Spencer, 1981; WHO, 1986). Dichlorvos is marketed in various formulations under the trade names *No-Pest Strip*® (Shell), *Vaporette*® and *Star-Bar*® (Thuron Industries), *S.W.A.T*® (Ciba-Geigy) and, as an antihelminthic in *Atgard*®, *Dichlorman*®, *Equigard*® and *Task*® (Gillett, 1972; ACGIH, 1987; Merck Index, 1983). Spray formulations include *Aminatrix*®, *OL*®, *Baygon Spray*®, *Precor Residual Fogger*®, *AE*®, *Safrotin Aerosol*® and *Safrotin Liquid*® (Worthing, nd). The analogues trichlorfon and naled (*Dibrom*®) are converted to dichlorvos *in vivo* and *in vitro* (Kaemmerer & Butenkötten, 1973).

Dichlorvos is a chlorinated organic phosphate pesticide that is characterized among common insecticides by its high insecticidal activity in the vapor phase. It is a nonflammable, amber liquid that is only slightly soluble in water (1%) but miscible in organic solvents. Although dichlorvos is stable to heat, it is hydrolysed by water (Worthing, nd; Spencer, 1981; Attfield & Webster, 1966). At

room temperature in aqueous saturated solutions dichlorvos hydrolyzes at the rate of about 3 per cent per day. Some investigators have found that residues may persist for long periods of time (Rowlands, 1975). The physical and chemical properties of dichlorvos are shown in Table 2.2.1.

Table 2.2.1 Physical and Chemical Properties of Dichlorvos

CAS registry number	62-73-7
Molecular weight	220.98
Specific gravity	1.415 at 25° C
Boiling point	140° C at 20 torr 120° at 14mm Hg
Vapor pressure	0.012 torr at 20°C
Refractive index n_D^{25}	1.4523
Water solubility (room temperature)	10 g/l

Dichlorvos is often incorporated into polyvinyl chloride resin strips which slowly release the active ingredient, dichlorvos, as a vapor (Gillett *et al.*, 1972; Brooks *et al.*, 1977). The tough flexible resin strip was originally developed by the World Health Organization, primarily for use in controlling malaria (Zavon & Kindel, 1966). Vapona® is a trademark for technical grade dichlorvos containing at least 93 per cent DDVP. Ciba Geigy markets the resin strip under the trade name S.W.A.T®. The constituents of Vapona® pest strips are found in Table 2.2.2.

In 1975, 80 per cent of the dichlorvos manufactured in the United States was formulated into resin strips (Santodonato *et al.*,1985). Since most resin strips are used for domestic control of *Diptera* species (*Mucsa domestica* [housefly] and *Cutex spp.* [mosquitoes]) future museum collections will possibly be exposed to dichlorvos residues (Rosenburg, 1984)

Table 2.2.2 Constituents of Vapona® Pest Strips

Active ingredient:	20%
DDVP	95 - 97%
Dipterex (trichlorfon or 0,0-dimethyl-2,2,2-trichloro-hydroxyethylphosphate)	1.5 - 3%
0,0-Dimethyl 2-chlorovinyl phosphate	0.4 - 0.7%
0,0-Dimethyl methylphosphonate	trace - 0.1%
0,0,0-Trimethyl phosphate	0.3 - 0.5%
Chloral (trichloroacetaldehyde)	0.1 - 0.5%
Plasticizer	30 %
Polyvinyl chloride	50 %

Components of the resin strips includes Dipterex® or trichlorfon, a dichlorvos precursor and a widely used insecticide that is considered equivalent to DDVP for toxicological evaluation. Chloral, an eye irritant, is a volatile hypnotic with a toxic oral dose of 40mg/kg or fatal dose of 10 grams for man (Gillett, 1972). Infrared analysis by Cardarelli (1980) indicated that the plasticizer

in the Shell strip formulation is dioctylphthalate (Williams *et al.*, nd). Although plasticizers are usually stable, transesterification may occur to form phthalic acid (Williams *et al.*, nd). Oxidative degradation of the polyvinyl chloride carrier, induced by γ-radiation at 25^{0}C produces the principal oxidation products hydroperoxide, alcohol and carbonyl compounds, carboxylic acid, and hydrogen chloride (Decker, 1976). A chemist at Ciba Geigy, Dr. J. Purdy, suggests that the PVC carrier may be detrimental to museum collections if the strip is not removed immediately after fumigation (Dr. Purdy, 1987). Vinyl chloride is a toxic chemical that is being investigated and controlled because of its adverse effects (Morcos, 1986).

DDVP resin strips are a unique insecticidal formulation with sustained fumigant activity that assures constant biotic exposure by inhalation, ingestion from particulates and objects, and continuous dermal exposure. Dichlorvos is readily absorbed on most surfaces (Gillett *et al*, 1972, Ashman *et al.*, 1974). The air concentration of DDVP varies with temperature and relative humidity. High humidity reduces the effective lifespan of the strip due to hydrolysis (Gillett, 1972). High temperatures may "increase the rate of hydrolysis in the air, increase the rate of diffusion throughout the air space, decrease the amount of sorption" and alter ventilation and air exchange rates (Gillett, *et al.*, 1972). A mathematical model of air concentration based on mass balance equations may be found in

Residue Reviews, 44 (Gillett *et al.*, 1972). The toxicity of dichlorvos is shown in Table 2.2.3.

Food chain concentration exposures as low as 0.01 ppb are toxic due to bioconcentration (CHRIS, 1984). Residues persist in aquatic systems for 62 days in water (CHRIS, 1984).

Table 2.2.3 Toxicity of Dichlorvos

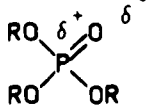
Class	6 or IB (highly hazardous)[a]
TWA[g] (former)	0.1 ppm[b] or 1 mg/m^3
TWA (revised, 1987)	0.33 mg/m^3 or 0.033 ppm
MPC[h] (USSR, 1968)	0.2 mg/m^3
STEL[i]	deleted by WHO (1986)[c]
ADI[j]	0-0.004 mg/kg[d]
LD_{50} (Oral, rats)	56-108 mg/kg[e]
LD_{50} (bluegill)	0.7 ppm[f]

[a] World Health Organization 1986 standard for liquid pesticides; CHRIS lists as Class 6 (most toxic and hazardous classification)
[b] (AGIH) *American Governmental Industrial Hygienists Documentation of Threshold Limit Values*.
USSR source Gillett, 1972.
[c] No safe limit determined
[d] WHO, 1986, and CCOHAS, 1987
[e] Gossel *et al.*, 1984 (for rats)
[f] Spencer, 1981, LD_{50} chickens is 14.8 mg/kg
[g] Time Weighted Average
[h] Minimum Physiological Concentration
[i] Short Term Exposure Limit
[j] Average Daily Intake

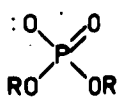
2.2.2 Chemistry

DDVP, an ester of phosphoric acid has a tetrahedral structure with a stable phosphoryl, P=O bond, which partially includes $p\pi$-$d\pi$ bonding with a σ-bond. The phosphorus-oxygen linkage by $p\pi$-$d\pi$ overlap contributes to the high affinity of phosphorus to the oxygen atom and formation of a stable phosphoryl bond (Eto, 1974).

Most biocidal phosphorus compounds are neutral esters. The electron deficient phosphorus atom is reactive as an electrophile because of the polarity of the phosphoryl group (Eto, 1974). An inductive effect from the strongly electronegative substituents contributes to the reactivity of phosphorus with nucleophiles (Desmarchelier *et al.*, 1976). Dissociation in partial esters diminishes this property (Eto, 1974).



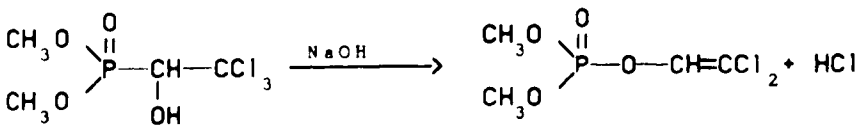
polarity of phosphoryl group



partial ester

Dichlorvos is prepared by the Perkow reaction where a trialkyl phosphate reacts with an α-halogenated carbonyl to give a vinyl phosphate. The proposed mechanisms include an initial nucleophilic attack by the phosphate on the electrophilic carbonyl carbon with rearrangement analagous to the base catalzyed attack which transforms trichlorfon to dichlorvos (see below) or a

nucleophilic attack on the carbonyl oxygen by phosphate (Kraemmerer & Buntenkötter, 1973; Eto, 1974).



trichlorfon dichlorvos

In neutral or slightly alkaline solutions one mole of HCl and one mole of dichlorvos are formed from the action of a base on trichlorfon (Kaemmerer & Butenkötter, 1973). Intramolecular phosphorylation initiated by the loss of chlorine ions is suggested to be the mechanism that converts trichlorfon to dichlorvos (Eto, 1974). Alternate names for trichlorfon are: *Tugen*®, *Neguvon*®, *Dipterex*®, *Bayer L 13/59*®, *Dylox*®, *Wotexit*®, chlorophos, chlorofos, and trichlorphon (Kaemmerer & Butenkötter, 1973; Vettorazzi, 1976; Mendoza, 1972).

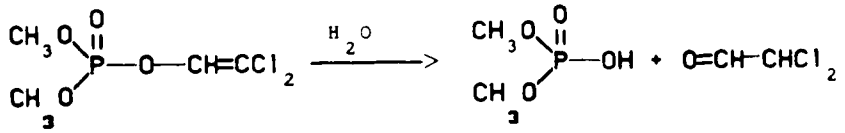
The suceptibility of dichlorvos, an esterified phosphoric acid, to alkaline hydrolysis contributes to the detoxication of the pesticide. In museums the environment is probably not alkaline.

Hydrolysis, which is primarily catalyzed by the -OH ion under alkaline conditions, is estimated to increase tenfold with each

increase in pH unit (Eto, 1974). Dichlorvos is apparently stable in some products under specific conditions (See 2.2.5).

In alkaline hydrolysis the hydroxide ion is suggested to hydrolyze the phosphate ester linkage at the phosphoryl P-center rather than at carbon (Eto, 1974). Water selectively attacks the phosphorus atom which results in fissi at the most suceptible acidic oxygen group in the molecule. The stereospecific nucleophilic displacement takes places with inversion of the configuration at phosphorus which is analogous to a S_N2 reaction.

In acid solutions hydrolysis is very slow. Isotope studies with water in acidic and neutral conditions indicate that C-O-bond fission occurs (Eto, 1974). Eto (1974) suggests that "the hydroxide ion attacks phosphorus to cause hydrolysis, and alcohol attacks methyl groups to cause dealkylation". Desmarchelier *et al.* (1976) state that both acids and bases cleave the enol ester linkage to yield the corresponding carbonyl compounds. The hydrolysis yields imethyl phosphoric acid and 1,1-dichloroacetalde de.



Dichlo os

Dimethyl phosphoric acid

1,1-dichloro-acetaldehyde

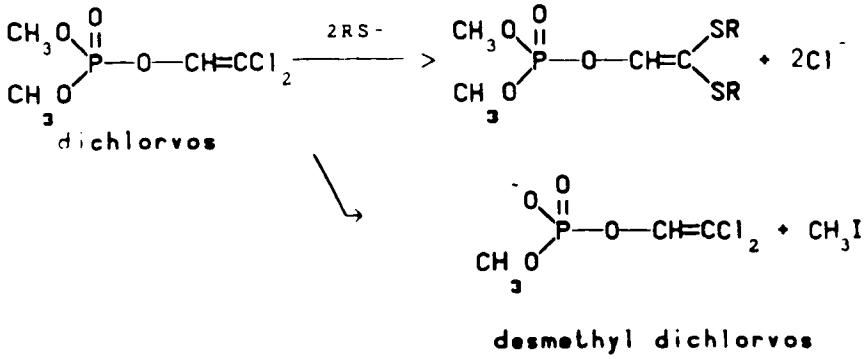
Reactions of dichlorvos in alkali are more rapid than acidic reactions. The half-life of dichlorvos at pH 11 at 28^0C is 2 hours (Eto, 1974), whereas, Attfield and Webster (1969) report that 2 ppm in aqueous solution at 38^0C will have a hydrolysis half-life of 60 hours at pH 1.1 (Attfield & Webster, 1969).

Metal ions such as Cu^{2+}, Pb^{2+} and Zn^{2+} also catalyze hydrolysis of dichlorvos The cupric ion, which probably increases the polarization of the P=O bond and facilitates the approach of the hydroxide ion, is the most reactive metallic catalyst (Eto, 1974). Some basic oxy-anions with lone pairs of electrons such as hypochlorite and peroxides catalyze hydrolysis by nucleophilic attack on phosphorus yielding unstable intermediates which are immediately hydrolyzed (Eto, 1974). Dichlorvos is known to be corrosive to iron and steel (Spencer, 1981).

Biological activity with dichlorvos such as esterase inhibition and toxicity are attributed to phosphorylating reactions. Prerequisites for phosphorylating activity are an increased positive charge on the phosphorus atom accompanied by decreased $p\pi$-$d\pi$ contribution to the P-O bond. Lone pairs of electrons on oxygen are necessary for the π bond contribution (Eto, 1974).

Alkylating properties of organophosphorus pesticides are significant reactions in chemical and biochemical degradation (Rowlands, 1975; Matsumura, 1986). Since dichlorvos has methyl ester groups and dichlorovinyl alkylating groups it participates in many

reactions. The methyl ester group reacts with the iodide ion to form desmethyl dichlorvos and CH_3I. A variety of sulfur compounds such as sulfides, thiocyanates, thiourea and thiols participate in dealkylation (Eto, 1974). The chlorine ions of the dichlorovinyl group are replaced by thio groups.



2.2.3 **Biochemistry**

One of the surest ways of upsetting the body mechanisms of insects is to chemically poison the insect's nervous system. The primary target of dichlorvos is the nervous system. Although there are inherent differences in the insect and mammaliam nervous systems, acetylcholine is identified as an important neurotransmitter in both the hexapod's and primate's central nervous system. The somatic system reacts to incoming (afferent) and outgoing (efferent) sensory stimuli by sending through the efferent nerve system impulses which terminate at a neuromuscular junction where the message is passed on

to voluntary muscles (Matsumura, 1985). The passage of the impulse is controlled by the cholinergic system which releases the chemical mediator, acetylcholine, to conduct the impulse across the gap or synapse (Matsumura, 1985). The normal function of acetylcholinesterase is to terminate neurotransmission (WHO, 1986).

Cholinesterases, enzymes that preferentially hydrolyze choline esters, are divided into two groups. The first, acetylcholinesterase or erythrocyte cholinesterase (AChE), occurs in the erythrocytes, nerve tissue, ganglia, motor end-plates, and electrical organs, and the second, cholinesterase or serum cholinesterase (BuChE), is found in serum pancreas, heart, and liver tissues. BuChE does not participate in nerve function (Eto, 1974).

Most knowledge about the activity of organophosphates has been obtained by investigating the reactions of esterases or proteases. The substrate specificity of AChE for esters containing a cationic group suggests that the presence of an anionic site will facilitate attack on the ester site. Hydrolases sensitive to organophosphate pesticides require a "dibasic amino acid...preceding the phosphorylizable serine" (Eto, 1974 pp 131). Eto (1974) suggests that the amino acid sequences Glu-Ser-Ala or Asp-Ser-Gly are an important part of catalytic sites. Histidine and serine are postulated to play an important role at esteric sites. Apparently the phenolic hydroxyl group of tyrosine contributes to the site specific activity. Dichlorvos mimics acetylcholine by occupying the

esteric site. A more in-depth discussion of binding and catalytic mechanisms for AChE is presented in Eto (1974).

Detoxication in some tissues proceeds at a slower rate than product literature discloses. Tracey *et al.* (1960) found that the stomach and brain of rats treated orally with dichlorvos were highly toxic to *Diptera*. Blood, muscle, fatty tissue, and the liver were not toxic to flies (Kaemmerer & Buntenkötten, 1973). Th implies that detoxication in tissues such as the human brain may be slower than venous acetylcholinesterase blood samples reveal.

Recently, free, bound and conjugated metabolites have been recognized as pesticide residues (Dorgough, 1976; Kaufman, 1976; Frear, 1976). Bound metabolites are conjugates which cannot be removed from the substrate by extraction (Dorough, 1976; Paulson, 1976). Bound and conjugated metabolites are seldom reported in the pesticide residue literature. Since conservation ethics state that treatments resulting in irreversible chemical change are unacceptable, bound residues are an important consideration in pest management.

2.2.4 Fate in the environment

Although dichlorvos is generally conceived to be rapidly hydrolyzed in organisms and in the environment many anomalies exist in the literature. Since various formulations continue to be used for commodities and in museum collections a brief summary of

incidences of persistence is recounted (Rowlands, 1975; Lippmann, 1987).

DDVP is regularly applied to grain (Hattori, *et al.*, 1976). The affinity and translocation of dichlorvos within grain are dependent on the lipoproteins, viability, moisture content and size of the grain or sample. Extensive binding and redistribution of pesticides occurs in grain. If the dosed grains are deliberately made non-viable (irradiated) the persistence of dichlorvos is increased (Rowlands, 1975). Both water soluble and insoluble proteins in wheat have been demonstrated to bind dichlorvos. Since dichlorvos is stable to heat residues may persist through the milling if bound to proteins (Rowlands, 1975).

Although most studies have concluded that dichlorvos is rapidly hydrolyzed in the environment others indicate that loss of the pesticide is slow. Wheat samples with "moisture contents of 9.3, 11.1, 12.9, and 13.7 per cent (wet basis) treated with 50 ppm of dichlorvos and stored at -15^{0}C, had levels of 49, 43, 34, and 34 ppm" after 11 months of storage (Kawar *et al.*, 1973). Although textiles exposed to Vapona® in the Edmonton collection were removed from the collection in the fall of 1986 the odor of dichlorvos still persists (See 4.1).

Often more than one insecticide is used in museums. Rowland reporting the research of Elms *et al.* (1972) indicates that joint residues of malathion *per se* and dichlorvos *per se* should be

considered as the total of the joint residues and treated as the most toxic or as dichlorvos (Rowlands, 1975). Dichlorvos is postulated to prevent the breakdown of more stable residues such as tetrachlorvinphos (Rowlands, 1975).

Removal of pesticide residues is of concern. One researcher reports that decontamination of dichlorvos treated surfaces may be achieved by washing with ethanolamine and propyl cellosolve using a 10:1 dilution (Rowlands, 1975).

2.2.5 Efficacy on target species

The toxic action of organophosphates is based on the inhibition of acetylcholinesterase enzymes at the synaptic gap of nerves. DDVP deactivates the enzyme and causes the nerve to keep firing. This results in involuntary muscle spasms, convulsions and death to the organism. However, it is believed that the neuromuscular junctions are not cholinergic in insects and that other transmitters are present (Eto, 1974).

Dichlorvos is an effective insecticide for the adult stages of most, but not all, common household and museum insects. Commercial usage recommends that one strip of Vapona per 1000^3 ft should be sufficient to eradicate target organisms.

Laboratory tests conducted at the Carnegie Museum using *Dermestes m. ulatus* (hide beetles) indicate that larvae may be more resistant to DDVP than adults (Williams, *et al.*, 1977). According to

the 1977 Carnegie Museum report, dermestid beetles were "still alive two weeks following treatment" (Williams *et al.*, 1977). Lellinger refers to a study by Stern *et al.*, (1968) that indicates that the black carpet beetle larvae, *Attagenus megatoma*, may survive a week's fumigation (Lellinger, 1972). The same study reported only a 19% mortality for *Attagenus megatoma* (black carpet beetle) in 7 days. (Lellinger, 1972). Apparently some species are more resistant than others and require dosages higher than the manufacturer's recommendations. Continuing research at the Carnegie Museum demonstrates that hatching success and cumulative effects on the adult dermestid occur in controlled experiments. Adult dermestids fumigated in 3.8 liter test jars for 3 minute intervals died after four or five exposures that ranged from 24 to 41 hours apart (Williams *et al.*, nd). Attfield and Webster (1969) found substantial differences in mortality among insect species. They also reported that dichlorvos has a cumulative effect on insects (Attfield & Webster, 1969).

Several studies have found variability in suceptibility. Although one week of fumigation resulted in 93%-100% mortality to *Attagenus megatoma* larvae, Lellinger (1972) reports that dichlorvos is "largely ineffective in killing insect eggs". Ashman *et al.* (1974) found that a number of carob pests survived treatment at 10 times the manufacturer's recommendation (Ashman *et al.*, 1974). A study of dichlorvos vapor for insect control in a rice mill indicated

that the per cent reduction of progeny after exposure to dichlorvos vapours ranged from 83% for the red flour beetle in open containers, to 30% for the lesser grain borer adult in closed containers. Although almond moth larvae was more susceptible to dichlorvos than pyrethrins, only 27% mortality after 7 days of exposure was achieved (McGaughey, (1973).

Insect toxicology is beyond the scope of this report. It is sufficient to recognize that there is an urgent need to examine the efficacy of biocides on target species within the context of museum pest management (Florian, 1987; Story, 1985).

Dichlorvos is also toxic to many economically and ecologically important non-target species. Bees and other pollinating insects are very sensitive to dichlorvos, naled and trichlorfon (Markkula, 1973). The toxicity to benthic algal communities has severe environmental consequences (Butler, 1977; CHRIS, 1984). The cost to the environment and non-target species is enormous (Lave & Upton, 1987; Haque, 1980; Carson; 1962; Stoker & Seager, 1976).

2.2.6 **Health Hazards to** *Homo sapiens*

An assessment of potential biological damage is one of the most important considerations in the analysis of residues. Automated analysis of residues against serum and erythrocyte cholinesterase with analytical accuracy has made it possible to monitor

cholinesterase depression in high risk occupations. However, no studies have been conducted on museum personnel exposed to continuous or intermittent dichlorvos fumigation. A survey of the literature indicates that concentrations beyond the TWA (Time Weighted Average; 0.3 mg/m^3) are common in museums (Lellinger, 1972; Ryckman, 1969). Since indications of insidious long term effects are recognized in current reports, a summary of the current status of dichlorvos is imperative (WHO, 1986; OH&S, 1986; Williams & Wilk. .984). The most commonly reported accidental organophosphate pesticide poisonings are with malathion, dichlorvos, and parathion (Matsumura, 1986). The high risk groups are manufacturers and spray operators, children, and researchers. Occupational poisonings are a serious problem (Gunther *et al.*, 1977). According to a 1957 report, 14,188 cases of accidental organophosphate poisoning were reported in California alone (Matsumura, 1986).

The toxicity of pentavalent phosphorus esters such as dichlorvos is attributed to their phosphorylating and alkylating properties. Phosphorylation of acetylcholinesterase (AChE), one of the hydrolytic enzymes for acetylcholine, inhibits normal nerve function at the synapse or neuromuscular junction. Acetylcholinesterase hydrolyzes acetylcholine to acetic acid and choline re-establishing the original state of the postsynaptic gap. Phosphorylation of AChE by dichlorvos prevents the enzyme from functioning. Loss of AChE activity leads to a range of effects that

are a result of excessive nervous stimulation (WHO, 1986).

Acute toxicity is attributed to acetylcholinesterase suppression with subsequent accumulation of toxic levels of the neurotransmitter acetylcholine. Acetylcholine operates at the cholinergic sysnapses which include motor, sensory, ganglionic and postganglionic sympathetic and parasympathetic nerve terminals, and the adrenal medulla. Acetylcholinesterase rapidly hydrolyzes the initial acetylcholine molecule to acetic acid and choline before a second impulse arrives. Inhibition of acetylcholinesterase disrupts the nerve junction which may result in peripheral paralysis and respiratory depression (WHO, 1986). The basic mechanism involves inhibition of AChE, accumulation of ACh, and over stimulation of central cholinergic neurons and the sympathetic and parasympathetic system (WHO, 1986).

Acute intoxication by organophosphate insecticides includes nicotinic, muscarinic, and central nervous system (CNS) manifestations (WHO, 1986). Organophosphates acting on the postganglionic nerve elements and excessive stimulation of the autonomic nerve effector cells produce muscarinic effects. These include excessive salivation, nausea, anorexia, diarrhea, bradycardia, a fall in blood pressure and cyanosis (Matsumura, 1986). Actions on the somatic nerve elements produce nicotinic effects which include twitching or spasms, flaccidity, a rise in blood pressure and paralysis (Matsumura, 1986). The symptoms of central

nervous toxicity may be headache, tension, fever, restlessness, mental confusion, ataxia, convulsions and coma (Matsumura, 1986). General symptoms occur at a critical level which is usually about 75 per cent depression below the no-exposure limit. Intoxication symptoms are described as cumulative. They include recurring headache, weakness, nausea blurred vision, tightness in the chest, hyperactivity, vocalization, nervousness and muscle twitching, and can culminate in coma and death (Lelligner, 1972, ACGIH, 1986-87).

Dichlorvos resin strip's unique formulation exposes organisms, and objects, to sustained fumigation. The speed of effect of dichlorvos is due to rapid absorption through the respiratory epithelium, stratum corneum, and the intestine. Repeated exposures to dichlorvos are cumulative causing progressive depression of cholinesterase.

The monograph *Occupational Health and Safety, Alberta, 1986*, states that "it takes about 30 days for a 25% decrement in plasma cholinesterase to return to baseline levels" and 120 days to fully recover from red blood cell cholinesterase depression (OH&S, 1986). Repeated sub-clinical doses may result in overt symptoms at exposures well below the levels expected to produce an effect in an unexposed individual (OH&S, 1986). Pesticide workers handling dichlorvos are reported to have significant reduction in "blood cholinesterase as well as leukocytosis, neutrophilia, and a decrease in lymphocytes and monocytes (ACGIH, 1986-87 p 192).

Mo cupational toxicity is ascribed to dermal exposure including contact with contaminated surfaces (Gosselin, 1984). Toxic signs and symptoms are an indirect result of enzyme inactivaton. At low concentration organophosphate pesticides act as excitatory substances. High concentration fatalities are usually due to respiratory failure or to central nervous system paralysis. Although cholinesterase inhibition appears to be reversible for several hours after acute exposure, repeated exposures or prolonged contact appears to be cumulative (Gosselin, 1984).

Dermal exposure to dichlorvos is an immense problem because its lipophilicity, volatility, extremely high toxicity, and rapid ed of toxicity. The time of onset of death after lethal doses of dichlorvos is 2-5 minutes (Purshottam & Srivastava, 1984).

The World Health Organization states that dermal contact is "the most important route of exposure for organophosphates (W.. , 1986, p 36). In studies of several powder formulations "potential dermal exposure markedly exceeded respiratory exposure" (WHO, 1986, p 36). The lipophilicity and volatility of dichlorvos increases dermal exposure (WHO, 1986). Uptake through the skin can be very efficient for more "lipophilic agents, and, since they avoid the first-pass metabolic disposal in the liver" the dermal route is direct (WHO, 1986 p 39). Some organophosphates are at *least* as toxic by the *dermal* route as by the oral route (WHO, 1986). Durham *et al.*, (1972) found that dermal versus respiratory exposure was 1000:1.

Repeated exposure to threshold limit values (TLV) may lower tolerances. The margin of safety is reduced when temperatures rise over 32°C or exposure exceeds 40 hours (Lellinger, 1972). Lellinger postulated that museum cases saturated with vapors at 40-45% RH would exceed the TLV for at least 70 days from ambient temperature were 22°C (Lellinger, 1972). Lellinger suggested that the easiest way to reduce exposure "is to hold one's breath" while opening cases (Lellinger, 1972, p 94).

Recent reports (WHO 1986) indicate that dichlorvos and trichlorfon can cause delayed neurotoxicity. "It is known that numerous people treated with only atropine for poisoning by trichlorfon have survived and then developed neuropathy" (WHO, 1986, p 61). The first clinical symptoms are often tingling and burning sensations in the limb extremities followed by progressive weakness and ataxia which can progress from flaccid paralysis to spastic paralysis (WHO, 1986). Neuropathy is initiated with phosphorylation (inhibition) of neuropathy target esterase (NTE) followed by aging (WHO, 1986). This is followed by axonal degeneration which results in permanent nerve damage and paralysis (WHO, 1986, Matsumura, 1985).

It has frequently been suggested that dichlorvos has carcinogenic potential because of mutagenic effects *in vitro* (WHO, 1986). Extensive microbial assays show dichlorvos to be a positive mutagen inducing mutation in a number of microorganisms (Santodonato, *et al.*, 1985). Chromosomal alterations and point mutations were

produced with *Aspergillus nidulans* and *Saccharomyces cerevisiae* (Santodonaco, *et al*, 1985). Base pair substitution and methylation at DNA guanine residues are reported. Although *in vivo* mammalian assays are inconclusive, *in vitro* human lymphocytes and human fibroblast cultures exhibit chromosomal aberrations and chromatid exchanges (Santodonato, *et al.*, 1985). One case of acute lymphoblastic leukemia in a child exposed to dichlorvos at home is reported (Santodonato, *et al.*, 1985). Numerous cases of skin rashes in animals and children have been reported (Gillett, 1972)

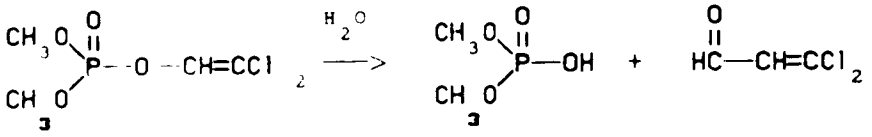
Teratogenic effects with moderate doses of trichlorfon in sows is reported (WHO, 1986). Teratogenic effects by embryonic somatic mutation or genetic mutation in eggs and sperm are often not identified due to abortion, death, or because morphological manifestations are subtle rather than overt (Matsumura, 1985).

Long term epidemiologic studies are absent in the literature. Subtle effects are not detected with current monitoring methods (except serum and RBC AChE). Nonspecific response may be related to abnormal levels of serum amino transferases such as SGOT and SGPT, serum bilirubin, plasma sodium and potassium, serum carotene and serum protein hemoglobin (Matsumura, 1985).

Many questions about the effect of dichlorvos remain unanswered. If safety is defined as the "incapacity to do harm then the issue is unresolved" (Gillett, 1972).

2.2.7 By-Products of Dichlorvos Degradation that Degrade Wool

Dichlorvos hydrolyzes readily, especially in basic environments. The principal products of dichlorvos hydrolysis in water at pH < 7 are:



Dichlorvos — Dimethyl phosphoric acid — 3,3-dichloro-2-propenal

The degradation of dichlorvos resin strips is postulated to produce a variety of chlorine derivatives. The hydrolysis of DDVP, DMVP and Dipterex and subsequent oxidation produces mono-, di- and tri- chloroperacetic and acetic acids, and the degradation of the polyvinyl chloride carrier produces small amounts of HCl (Williams, n.d.). All species listed are capable of causing topographical, chemical and physical changes in keratins. For example:

(a)	trichloroacetic acid	protein precipitant
(b)	dichloroacetic acid	topical keratinolytic
(c)	monochloroacetic acid	skin irritant
(d)	hydrochloric acid	severe skin burns, and dermatitis

(Williams, n.d.; Merck, 1983).

The phosphorus species present in the dichlorvos resin strips

include dimethyl phosphoric acid (50%), O,O-dimethyl methyphosphonate (<0.1%) and O,O,O-trimethyl phosphate (0.3-0.8%) (Williams, n.d; Gillet *et al.*, 1972). Dimethyl phosphoric acid hydrolysis produces peroxide species. Peroxides are highly reactive oxidants that not only produce skin (keratin) burns but also initiate chemical reactivity through free radical chain reactions. Phosphoric acid is a skin irritant as well as an acid catalyst (Merk Index, 1983). Many other phosphorus compounds attack keratinized tissue. Phosphines are used to solubilize fibrous keratins for amino acid analysis (Crewther, 1975). Reduction of disulfide to thiol groups is the major reaction of phosphines (Crewther, 1975).

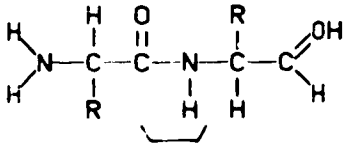
2.3. PROTEIN CHEMISTRY

2.3.1. Introduction

The complexity and structural heterogeneity of mammalian keratin and wool fibers led Sikorski (1975 p 103) to refer to an imminent unravelling of the three-dimensional structure "as prevailing scientific lore; where simplified models and facile interpretation appear to be in direct conflict with the ...facts". Fraser *et al.* (1972) recognizing the complexity of protein chemistry say direct unambiguous interpretations are rarely possible because all techniques are subject to artifacts and error and even the most

erudite researchers lack expertise in all facets of research. The tendency is to place reliance on unfamiliar results and techniques where one is unaware of the difficulties of interpretation. Despite these difficulties, an overview of the fine structure of wool fibers, according to current knowledge and theory, is presented in the literature review.

The fine structure of wool is a complex combination of long chains of molecules which aggregrate into supra-molecular structures forming the fiber (Alexander *et al.*, 1963; Fraser *et al.*, 1975; Hearle, 1963a; 1963b). Protein is the main constituent of wool. Proteins are composed of chains of amino acids linked together by peptide bonds or amide linkages. The peptide bond is formed between the carboxylic acid group of one amino acid and the amino group of the next amino acid by the elimination of a molecule of H_2O. There are 20 common amino acids in polypeptides, each with a different side chain attached to the α-carbon (Table 2.3.1). Chemical and mechanical properties of protein fibers are determined by the nature, arrangement and proportions of the amino acids in the polypeptide.

amino (N-terminus)  carboxyl (C-terminus)

peptide bond

2.3.2 Protein synthesis

Protein synthesis involves copying the coding regions of DNA into ribonucleic acid (RNA) by a process known as DNA transcription. The transcribed RNA undergoes major chemical changes before leaving the nucleus to serve as messenger RNA (mRNA) molecules which direct protein synthesis in the cytoplasm (Alberts *et al.*, 1983). The information contained on a small segment of DNA can direct synthesis of specific proteins in large quantities because each mRNA can be translated into thousands of protein molecules.

Amino acid sequencing and conformational studies of proteins has led to a greater understanding of keratin fibers (Fraser *et al.*, 1980; Happey, 1975; 1978). Although deciphering of the fine structure of wool is not complete, researchers are more able to predict and understand the physical and chemical changes that occur when the fibers are exposed to contaminants (Robson, 1975; Slater, 1975).

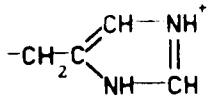
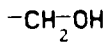
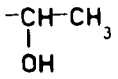
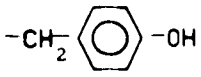
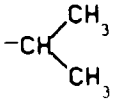
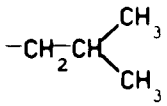
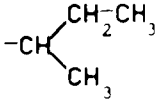
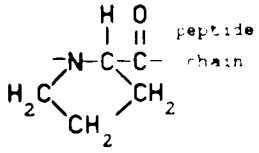
2.3.3 Primary Structure

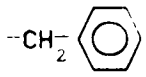
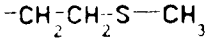
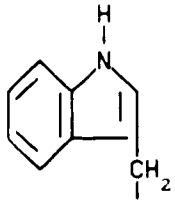
The protein molecule's unique characteristics are attributed to the amino acid sequence in the polypeptide (Maclaren & Milligan, 1981). The side chains of 20 amino acids residues found in wool fibers are presented in Table 2.3.1. A detailed knowledge of amino acid sequence, and reactivity is contributing to an understanding of the behaviour of proteinaceous fibres (Leeder, 1986; Alberts *et al.*,

1983; Sakamato, 1985a; Lindley, 1977; Fraser & McRae, 1973).

Table 2.3.1 Amino Acids Residues in Wool and Their Properties.

Residue	Side Chain	Properties
Cystine (paired cysteine)	$-CH_2-S-S-CH_2-$	diamino acid forms inter/intra chain disulfide bonds
Cysteine (Cys, or C)	$-CH_2-SH$	reacts as thiol interchange with (Cys)
Glutamic acid (Glu, or E)	$-CH_2-CH_2-C(=O)-OH$	reacts as a carboxyl pK≈4.3
Aspartic acid (Asp, or D)	$-CH_2-C(=O)-OH$	reacts as a carboxyl pK≈3.7
Glycine (Gly or G)	$-H$	uncharged, glycine/tyrosine
Glutamine (Gln, or Q)	$-CH_2-CH_2-C(=O)-NH_2$	reacts as amide strong H bond
Lysine (Lys, or K)	$-CH_2-CH_2-CH_2-CH_2-NH_2$	reacts as primary amine. pK≈10
Arginine (Arg, or R)	$-CH_2-CH_2-CH_2-CH_2-NH-C(=NH)-NH_2$	Reaction of guanidyl forms salt links E and D, pK≈13

Histidine (His, or H)		Nitrogens positively charged below pH 6
Serine (Ser, or S)		Does not behave as normal alcohol Sulfates with conc H_2SO_4
Threonine (Thr, or Y)		- OH group relatively unreactive, difficult to sulfate
Tyrosine (Tyr,or Y)		reacts like phenol high. . r proteins form crosslinks
Alanine (Ala, or A)		nonpolar side chain crosslinking with aspartic, glutamic,
Valine (Val, or V)		nonpolar side chain
Leucine		nonpolar
Isoleucine		nonpolar
Proline (Pro, or P)		absent from α-helix imino acid

Phenylalanine (Phe, or F)		hydrophobic interactions
Methionine (Met, or M)		sulfur relatively unreative
Tryptophan (Trp, or W)		reacts like indole

The primary structure of the polypeptide chain, or linear sequence of amino acids provides a framework for the characteristic reactions of specific proteins (Crewther, 1975; Asquith & Leon, 1977; Howitt, 1963; Light, 1974). Methods of separating amino acids from the polypeptide chain have developed rapidly in recent years (Lindley, 1977; Crewther, 1975; Maclaren *et al.*, 1960; Gillespie & Frenkel, 1973; Kulkarni & Bradbury, 1974; 1975). A few of the tools and methods available to study protein chemistry are: enzymes that attack specific bonds on the chain (Fletcher & Buchanan, 1977; Peters & Bradbury, 1972), ultraviolet absorption (Bendit, 1960), optical methods (Fatou, 1978), electron microscopy (Bendit, 1975; Carr *et al.*, 1986a, 1986b; Dobb & Murray, 1975; Dobb & Rogers, 1969; Dobb & Sikorski, 1971; Danilatos & Brooks, 1985), physical methods such as gel filtration (Marshall *et al.*, 1983-84), starch-gel

electrophoresis (Fraser, *et al.*, 1972; Stephanie & Zahn, 1985), thin-layer electrophoresis (Fletcher & Buchanan, 1977), monoclonal antibodies (Hewish *et al.*, 1983-84; 1984; Hewish & French, 1986), ultracentrifugation, hydrolysate analysis with automatic amino acid analyzers (Bradbury *et al.*, 1970) and gas chromatography (Fletcher & Buchaman, 1977)

Functional groups on the side chains of the amino acids determine the chemical activity of the residues (Maclaren & Milligan, 1981). The common amino acids are grouped according to their side chains and behaviour of functional groups (See Table 23.1). The more reactive amino acid residues contain the carboxyl, amine, and hydroxyl, and cystine groups. Acid-base characteristics of the amino acid residues influence the fiber's reactivity with hydrogen and other ions in aqueous environments (Makinson, 1967, Mathieson *et al.*, 1964; Mathieson & Whewell, 1964). Cysteine forms disulfide bonds which play an important part in the conformation and reactivity of keratin protein fibers (Lindley, 1959; Ebert & Zander, 1985; Maclaren & Milligan, 1981; Crewther, 1975; Sakamoto *et al.*, 1985).

2.3.4 Secondary structure

The polypeptide chain of a protein fiber can interact with itself to form the secondary structure (Crewther, 1975). Resonance may contribute to the unusually short carbon-nitrogen bond of the amide linkage (Dickerson & Geis, 1969). Consequently rotation about

the carbon-nitrogen bond is severely hindered (Solomon, 1984). However, rotations of groups attached to the carbonyl carbon and amide nitrogen are relatively free and allow the molecules to form different conformations.

Secondary structures are formed by folding sections of the polypeptide chain (Lotz & Black, 1979). Distribution of the polar and non-polar side chains is an important factor governing the folding of the polypeptide chain (Aubry *et al.*, 1984). Hydrophobic side chains are usually pushed towards the interior of the molecule to avoid contact with water, whereas, the polar side chains tend to be in areas where they can react with other polar groups (Fraser & MacRae, 1973). Non-polar groups are found on the surface for specific reasons (Dickerson & Geis, 1969). Hydrogen bonds, between adjacent amino and carbonyl groups, stabilize the folds and are crucially important for many of the interactions on the surface of protein fibers (Huggins, 1975). Aspartic acid and glutamic acid, with free carboxylic groups, and, lysine, arginine, and histidine residues, with free basic groups can, combine to form salt linkages that contribute to the secondary structure (Howitt, 1963, 1962; Fletcher & Buchanan, 1977; Alexander *et al.*, 1963; Ziegler *et al.*, 1967).

Cystine is assumed to form covalent interchain cross-linking between two adjacent chains, and intrachain links (Maclaren & Milligan, 1981). Cysteine residues link together to form disulfide

bonds. Inter and intrachain disulfide links are believed to stabilize the three dimensional tertiary structure of wool (Fraser *et. al.*; 1972, Alberts *et al.*, 1983). H bonds and salt linkages also stabilize the fiber (Howitt, 1963, Arai, 1985, Meichelbeck & Spei, 1975; Ebert *et al.*, 1977; Ebert & Zahler, [illegible]

The major portion of wool consists of a cortex composed of different types of proteins. Fraser and MacRae (1973) state that "little is known about the disposition or conformation of matrix proteins" within the cortex. Evidence suggests that matrix proteins have a globular structure and microfibrils, a fibrillar structure (Fraser & MacRae, 1973, Menefee, 1977; Crewther, 1975; Dobb & Rogers, 1967). Continuing research is focused on the nature of crosslinks, and the arrangement of proteins in the matrix and microfibrils (Fraser & MacRae, 1973; Kulkarni & Bradbury, 1974; Dobb & Murray, 1975). Maclaren and Milligan (1981) suggest that "many of the original disulfide crosslinks in wool must be interchenic...it has been suggested that all of the polypeptide chains in wool may be crosslinked". Identification of the proteins bridged by crosslinks and the relative proportions of intra-molecular and inter-molecular bonds will assist in defining the tertiary structure of wool (Orwin *et al.*, 1985; Ziegler, 1977).

Asquith (1985) suggests that amino acids are divided into α-helix forming and α-helix-breaking residues which cause the peptide chain to form other conformations (Meichelbeck & Spei, 1975).

Although the codes required for folding protein chains into characteristic conformations are contained in the amino acid sequence, they have not been deciphered (Huggins, 1975). Most of the knowledge about folded conformations was determined by analysis of x-ray diffraction patterns (Astbury, 1933, Fraser & MacRae, 1973; Fraser *et al.*, 1975; Kulkarni & Bradbury, 19[illegible] Happey, 1975). X-ray analysis is a major experimental technique used in unraveling the secondary structure of protein fibers (Fraser & MacRae, 1973, Blakey, 1979).

2.3.5 Conformation determination

X-ray diffraction has been used to determine the three-dimensional arrangement of atoms within the crystalline areas, the relative amount of crystallinit and orientation of the internal structure of fibers (Fraser & MacRae, 1973; Happey, 1975; Happey, 1978; Jaksie & Jovanovic, 1985; Johnson & Walton, 1975). X-ray diffraction is also used to study structural changes in wool treated with chemicals or wool that is mechanically deformed (Farnsworth, 1964, Sikorski, 1975, Hearle, 1963). Despite the limitation of x-ray diffraction much knowledge about the structure of protein fibers has been gained (Farnsworth, 1964, Fletcher & Buchanan, 19[illegible]).

Astbury, and co workers at Leeds, studied the high angle patterns of wool and deduced that the prominent 5.1 Å meridional reflection was associated with the length of the fold and the 9.8 Å

equatorial reflection with the lateral separation of the folds, and that polypeptide chains in wool are foreshortened by some form of regular folding (Farnsworth, 1964, Astbury, 1933). By 1961, Pauling, Corey, and Bronson were able to describe the α-helix configuration as the basic structure of keratins including wool (Farnsworth, 1964). Pauling and Corey suggested that the helices formed coiled coils twisted together into a rope-like structure (Speakman, 1985).

Two folding patterns in three dimensional protein structures were correctly predicted in 1951 from the x-ray diffraction patterns of wool and silk. These are known as the β-pleated sheet of the protein fibroin, found in silk, and the α-helix, the unextended helix which occurs in keratin protein (Fraser et al., 1972). X-ray diagrams of keratin fibers usually show folded chain patterns that are designated as α-keratin (Astbury, 1973). If the fibers are straightened, the α-helix pattern is transformed into β-keratin pattern similiar to the x-ray diagram of silk. Fraser cautions that interpretation of x-ray diffraction diagrams is difficult because extracted proteins may yield α-, β-, or amorphous patterns depending on the keratin's chemical and mechanical history (Fraser *et al.*, 1972).

2.3.6 Tertiary structure

The tertiary structure arises from further foldings of a protein into three dimensional shapes. Folds superimposed on

α-helixes in wool contribute to the unique characteristics and performance of the wool fiber (Mclaren & Milligan, 1981).

The stabilizing of tertiary structures involves a variety of forces including the disulfide bond, hydrogen bonds, salt bridges, hydrophobic interactions and tyrosine crosslinks (Arai & Nagaoka, 1985; Ebert & Zander, 1985). The tertiary structure of fibrous proteins includes the arrangement of the α-helical strands. The strands of α-keratin associate to form the proposed super-helix (Spei, 1985; Kulkarni & Bradbury, 1975). Tertiary and quaternary structures of fibrous protein fibers are being investigated by conformational studies (Spei, 1975; Fraser *et al.*, 1975; Johnson & Walton, 1975; Bonart, 1975).

The exact nature of folds in α-keratin polypeptide chains remains elusive (Huggins, 1975; Jaksic & Jovanovic, 1985). SEM micrographs established the fibrillar nature of chemically disrupted α-keratin in wool keratin and defined the microfibril (α-filament) as an important element (Fraser *et al.*, 1960; Dobb, 1969; Fraser *et al.*, 1975). Although electron microscopic studies of cross-sections of wool have failed to reveal the organization or form of α-keratin microfibrils in the matrix, refractions from other hair keratins favor two strand coiled-coils (Fraser & MacRae, 1973). More recent studies using molecular weight sequences of the helix rich fragments support the hypothesis of a two or four subunit structure (Woods, *et al.*, 1979-80). Until the ultra-fine structure is determined the

secondary , ertiary structures of keratins cannot be definitively defined.

2.3.7 Summary of Structure of Proteins

Bio-polymers consist of long chains of covalently linked molecules with atoms that interact within and between chains. Although there is restricted rotation at the amide linkage carbon-nitrogen bonds, other single bonds rotate. Much research has been conducted to determine the amino acid sequences and structure of keratin fibers (Crewther, 1975).

Damage and distortion of fibers at the macroscopic level produces changes at the molecular level. The performance and survival of ancient and historic objects composed of keratin are dependent on the integrity of molecular chains within the fiber. Before the physical and chemical properties can be examined the morphology, chemical and physical characteristics of wool need to be discussed.

2.4 Keratin fibers

2.4.1 Morphology

Keratin, derived from the Greek word Κερασ, is a generic term for a group of highly specialized proteins that form the resilient structures such as hair, horn, feathers and skin of higher

vertebrates. Keratin is distinguished from other fibrous proteins by an unusually high proportion of cystine residues and birefringence[1] (Swift, 1977). Soft keratins occur in skin (stratum corneum) and eponychium of nails. Soft keratins contain fewer cystine residues than hard keratins. Hard keratins, which contain more than 3% sulfur, are found in hair, nails, quills, beaks and horn (Fraser, 1972, 1973, Bradbury, 1973, Swift, 1977).

Wool is a hard fibrous keratin protein produced by specialized epithelial cells in *Capra* (goats) and *Ovis* (sheep) (Kondo *et al.*, 1971). Keratin fibers in other mammalian species are generally referred to as hair (Harper, 1931, Wildman, 1953, Harris, 1954; Kidd, 1977). The wool fiber is a complex structure with normal cellular characteristics, albeit, modified through keratinization. Although the underlying process of keratinization (hardening of the fiber) is obscure, thiol groups are oxidized to form the diamino acid cystine (Truther, 1973, Fraser & MacRae, 1973). The extreme resistance of keratin to degradation by chemicals and enzymes is related to keratin's function as an environmental barrier. The resistance is attributed to the way "various components tend to be complementary in their inertness towards chemicals" (Bradbury, 1973) and the stability of the disulfide three dimensional network (Koenig,

[1]Fraser and MacRae report that wool cuticle and matrix differ in birefringence. Wool is less birefringent than some of the other keratins.

1977). Protein in medullated fibers contains γ-glutamyl formed by a reaction between lysine and glutamic or aspartic acid residues. These cross-links make the medulla resistant to keratinolytic solvents (Bradbury, 1977).

The morphology of wool is extremely complex. Wool may be described as a composite material consisting of cuticular cells, cortical cells, and in some cases a medulla, and minor histological components that form the cell membrane complex (Earland *et al.*, 1963, Hearle, 1963; Speakman, 1985). There are different types of components within the cuticle, cortex and medulla (Truter, 1973; Speakman, 1985). Wool fibers come into existence as an assembly of living cells originating from a single epithelial follicle. Follicles in keratin fibers are organized into multiple concentric cylinders that differentiate into specialized cellular structures. Each cell contains cytoplasmic and nuclear proteins bounded by a cell membrance. As the fiber differentiates and keratization occurs, changes in the morphology and chemistry take place (Marshall *et al.*, 1985). The principal cellular differentiation in fine wool is flattened, external cuticle cells, and long polyhedral cortical cells (Sikorski, 1963a, 1963b; Nishimi, 1971). A review of the general cellular organization will assist in the understanding of degradation of wool fibers (Earland, 1963; Fraser *et al.*, 1972, Swift, 1977).

2.4.2 Cuticle

The scaly outer layer formed by overlapping flattened cells has a laminar structure consisting of an outer resistant keratinous exocuticle, and an inner non-keratinous endocuticle that is formed from cellular debris (Fraser, 1972). The hard shell-like scales overlap leaving approximately three-fourths of the cell length exposed (Swift, 1977, Speakman, 1985). The projecting distal edge extends toward the fiber tip. Cuticle cells in wool fibers tend to be irregular in thickness, fluted on the exposed surface and have frequent thicker ridges at the exposed distal margins (Swift, 1977, Makinson, 1979).

Differentiation of matrix cells into cuticle occurs in the upper bulb region of the follicle (Fraser, 1972). Granules deposited on the periphery grow beneath the outermost cell membranes forming a granular layer that compresses the nucleus and other cytoplasmic constituents into an amorphous layer in the inner side of the cell (Bradbury & Ley, 1972). The granular layer forms large blocks that convert into the A and exocuticle layers (Swift, 1977). As the single layer of cells is pushed upward the cells elongate, flatten, tilt, and slide over each other emerging as an overlapping layer of cells. As keratinization proceeds deposits of amorphous material form in the intercellular space (Bradbury, 1973)..

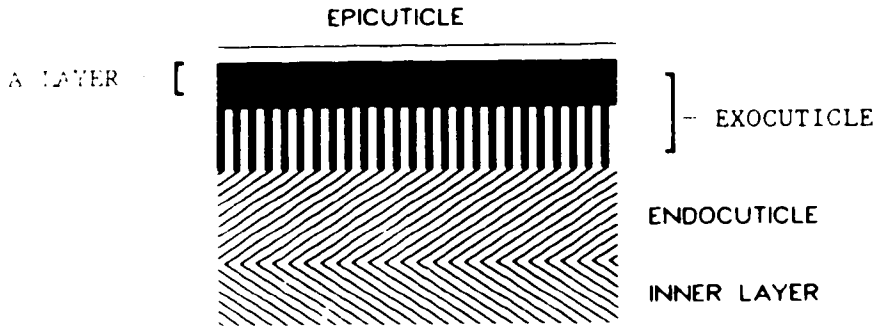
Junctions between adjacent cuticle cells, and the cuticle and cortex are usually planar (Swift, 1977). Different theories have

been proposed to explain cuticle cell adhesion. Swift (1977) describes ball-and socket interconnections, suggested to arise from partial phagocytosis between cells. According to Earland *et al.* (1963) the scales are linked together by rivet-like tono-fibrils (Alexander *et al.*, 1963). Leeder (1981) suggests that the cell surface glycoprotein called fibronectin, binds to cell surfaces and promotes cell adhesion.

Considerable controversy over the presence of a waxy or fatty layer on the surface of wool cuticle exists in the literature (Leeder, 1986; Bradury, 1973). The hypothesis that a hydrophobic layer exists on the surface of wool fibers is supported by the isolation of free fatty acids that may be derived from fatty acids "covalently bonded to serine residues by ester linkages" (Evans *et al.*, 1985; Nogues *et al.*, 1988).

Each cuticle cell is composed of lamellar subcomponents (Bradbury & Le 72; Nakamura *et al.*, 1975a). The principal laminae are the cystine rich exocuticle and cystine poor endocuticle. Selected staining and reduction with osmium or silver salts reveal additional laminae (Swift, 1977). The *A* layer which is found just under the cuticular cell membrane and inner layer, is extensively stained (Speakman, 1985; Bradbury, 1975).

Figure 2.4.1 Diagram of Cuticle Subdivision



Cuticular cells are covered with a hydrophobic thin membrane called the epicuticle (Sakamoto, 1985). The epicuticle is the proteinaceous layer which forms Allwörden sacs or bubbles in chlorine water (King & Bradbury, 1968; Bradbury & Leeder, 1972; Makinson, 1979). Earlier workers thought that the epicuticle formed a continuous sheath on the wool fiber (Trotman, 1984; Bradbury, 1975). Observations of Allwörden bubbles forming on the outer surface of individual cuticle cells conclusively shows that the epicuticle surrounds individual cells (Leeder, 1986; Ley *et al.*, 1985).

Allwörden sacs are formed, in the presence of chlorine water, by the generation of osmotically active degradation products from large amounts of oxidizable cystine (Leeder, 1986, Allen, *et al.*, 1985, Baumann & Setiawan, 1985). The *A* layer of the exocuticle, which is immediately beneath the epicuticle, is the only histological region in wool with sufficient cystine residues to produce the phenomenon (Makinson, 1979). Recent studies by Allen *et al.*, (1985)

suggest that the Allwörden membrane contains lipid, is of similar composition to low sulfur wool protein, and contains a filamentary protein (Allen *et al.*, 1985, Zahn 1985; Coderch *et al.*, 1985).

The δ-layer, and β-layers have been the subject of recent investigations (Nakamura *et al.*, 1985, Allen *et al.*, 1985; Nogues *et al.*, 1988). A hydrophilic δ-layer occurs between cuticle cells, and cuticular and cortical cells. The δ-layer, sandwiched between the hydrophobic β-layers, provides a canal for water penetration. The endocuticle is connected to the δ-layer by connecting canals through the β-layer (Leeder, 1986; Sakamoto, 1985).

By treating wool fibers with low concentrations of metal salts, catalytic metal can be dispersed in only the δ-layer. Subsequent treatment with oxidizing agents causes rapid oxidation inside of the δ-layer and endocuticle The process has been used by Hojo to release the exocuticle from wool (Hojo, 1985).

2.4.3 Cell Membrane Complex

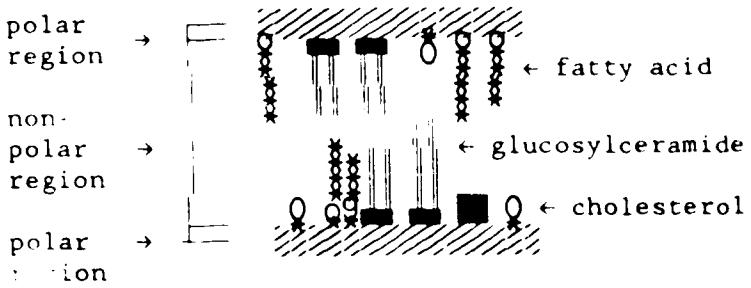
The cell membrane complex (CMC) forms a network structure which underlies the cuticle cells and completely surrounds the cortical cells holding the cellular components of the fiber together (Bradbury & Leeder, 1971; Bradbury, 1973; Fraser & Rogers, 1955). Recent research implies that the cell membrance complex is the preferable plane of fission (Sikorski, 1975). Its role in degradation extremely important (Orwin & Thomson, 1975). The CMC

is formed from plasma membranes. Plasma membrane modifications during biosynthesis result in deposition of a composite laminar layer formed from two plasma membranes separated from each other by means of intercellular cement (Bradbury, 1973). The intercellular cement has very low levels of cystine, and high levels of glycine, tyrosine and phenylalanine. The intercellular cement together with the endocuticle, intermacrofibrillar material and cell nuclei are termed *nonkeratinous* (Leeder, 1986; Bradbury & Leeder, 1971).

The original plasma membrances are modified to become the inert lightly stained β-layers separated by the central proteinaceous δ-layer (Leeder, 1986). Bradbury (1973) describes the β-layers as "two protein layers interleaved with a lipid bilayer". This has led to the conclusion that the *tram-line* apparent in stained sections may be caused by hydrophobic ends of the phospholipid bilayers from the plasma membranes formed in the follicle (Leeder, 1986; Herrling & Zahn, 1985). Glucosylceramide was recently isolated from wool and is suggested to be present in the β-layer of the CMC (Nogues *et al.*, 1988). The proposed structure of bilayers is illustrated in Figure 2.4.2.

The resistant membranes persist as a fibrous 'ghost' residue of the cortical and cuticle cells (Leeder, 1986). Since the resistant membrane contains the same proportion of cystine crosslinks as the whole wool fiber, Leeder (1986) suggests that steric sheltering of some of the disulfide links may prevent reduction, or

Figure 2.4.2 Illustration of Possible Lipid Bilayer



an unrecognized bond may exist. High levels of lysine in the resistant membrane may contribute to isopeptide cross-links such as ε-(γ-glutamyl lysine) that could contribute to the inertness of the membrances (Leeder, 1986). The only difference between cortical and cuticle resistant membranes is the increased amount of citrulline and ornithine in the cuticle cell resistant membranes (Leeder, 1986).

Several researchers have demonstrated that diffusion occurs between cuticle cells, at points of cuticle overlap, and that preferential diffusion occurs via the CMC and other nonkeratinous regions (Leeder *et al.*, 1985). Apparently high molecular weight compounds can diffuse into the wool along low crosslinked regions (Leeder, 1986).

2.4.4 Cortex

The complexity of the histo-morphology within the cortex exceeds that of the cuticle and cell membrance complex (Speakman,

1985). The cortex constitutes about 86.5 per cent of the fine wool fiber (Bradbury, 1977). The generalized cortical structure includes ortho and paracortex, which contain macrofibrils embedded in a matrix, cell membrances, and a medulla in some fibers (Marshall *et al.*, 1985b, Orwin *et al.*, 1984). The composite α-helix structures of the microfibrils embedded in a matrix act like a two-phase mechanical model (de Jong & Curiskis, 1985).

In cross-sections cortical cell boundaries are more clearly discerned in the paracortex than in the orthocortex. Cortical cells liberated by formic acid, hydrochloric acid, or enzymes are suggested to be spindle shaped with "fingerlike processes at their ends, which interdigitate with adjacent cells" (Bradbury, 1973). Sikorski (1963) describes cortical cells as "space filling polyhedra with unlimited sharing of faces and corners" and suggests abandoning the "spindle-shaped" concept. Cortical cells are reported to be linked by projections. Sikorski (1963) reports the occurrence of horns or arms at various points along the cell axis that provide transverse interlocking between neighbouring cells. Cortical cells adjacent to the cuticle appear flattened (Bradbury, 1973), and the sides of cortical cells adjacent to the medulla produce "fingerlike trabeculae" that separate and secure individual medullary cells in position (Bradbury, 1973).

Cortical cells are formed in the keratinocytes of the follicle (Orwin et al., 1985). The follicle basal cells actively

engage in protein synthesis to form the matrix. Matrix cells retain their products of synthesis as cellular remnants. Melanin is incorporated into the developing cortex before cortical differentiation occurs in the upper regions of the follicle. Melanocytes, which synthesize the pigments of the fiber shaft, are interspersed among the matrix follicle cells. As the matrix cells undergo differentiation and progressive cell division the pigment is carried up the hair shaft within the matrix cells (Swift, 1977).

Cortical differentiation occurs in the upper region of the follicle (Chapman, 1975). Lateral compression causes axial elongation of the cortical cells into a spindle form. Fraser observed that cortical cells become densely packed with fine filaments or microfibrils (Fraser, 1972). The fibrils align at the periphery parallel to the long axis of the cell (Kulkarni & Bradbury, 1972). At the same time electron dense material forms between the fibrils so that discrete bundles occur (Swift, 1977). The growth of the fibril aggregates fills the cell compressing other components, ribosomes, mitochondria, and cytoplasmic membranes, into an amorphous residue between the fibril bundles. The cell nuclei also undergo distortions finally becoming spindle shaped stellates in the central portion of the cell. Amorphous deposits that resist staining are laid down in the irregular intercellular spaces (Leeder, 1986).

In 1953 Horio and Kondo's observations of differential dyeing properties in crimped rayon led to their discovery of the

bilateral structure of wool. The bilateral structures are called the orthocortex and paracotex. The ortho- and paracortex structure in fine wool, which first appears at one third the distance from the root to skin surface, is responsible for crimp (Sakamoto, 1985; Chapman, 1975; Hunter & Gee, 1980). The ortho and para cortices wind around each other helically in phase with the crimp so that the orthocortex is on the outside and the paracortex is on the inside of the crimp curvature (Bradbury, 1973; Denton, 1982; Sparrow 1984-86). About 30-50 per cent of the cortex in fine wool is comprised of paracortex. The proportion of paracortex increases with fiber diameter until the bilateral asymmetry of fine wool is replaced by cylindrical asymmetry in coarse wools (Bradbury, 1973). In general, the bilaterally asymmetric fibers are crimped or curled and the cylindrically asymmetric fibers are straight. In Lincoln wool the paracortex may be found in the central core surrounded by a poorly defined orthocortex. Onions (1962) suggests that the ortho and para cells are mixed throughout the the cortex in Lincoln wool and mohair giving a mottled appearance. Variations occur from species to species (Bradbury, 1973, Higgerson *et al.*, 1985; Kaplin & Whitely, 1985).

Separation of the denser paracortical cells from the orthocortex may be attained by reduction and S-carboxymethylation using density differences (Ito, *et al.*, 1985). Bilateral staining may result from differences in the non-keratinous network, the

intercellular cement of the CMC, and amino acid composition (Miyamoto, et al., 1985). The CMC, not only provides binding sites for chemical reaction, but also, channels for infusion of chemicals (Leeder *et al.*, 1985b).

A core of meta-cortex has also been reported in fine crossbred wools (Onions, 1962). The metacortex is suggested to be more basophilic, to contain less sulphur than the para and orthocortex, and to be related to the medulla (Onions, 1962). According to Swift (1977) the mesocortex (meta-cortex) contains both the ortho and paracortex macrofibrillar arrangements. Kaplin & Whitley (1985) describe the mesocortex as being fused in appearance, with an intermediate staining reaction, a regular arrangement of microfibrils, and an intermediate fibril/matrix ratio (Kaplin & Whitely, 1985). The non-keratinous material is mainly nuclear remnants.

Electron microscopic examination of wool cortex cells reveals that the cells may be broken into thinner components (Dobb & Sikorski, 1971; Kulkarni *et al.*, 1971; Swift, 1971; 1975). These are described as sheets of parallel fibrils or macrofibrils which contain microfibrils with α-helices (Onions, 1962). Macrofibrils represent aggregrates of microfibrils. The arrangement of macrofibrils in the para and ortho cortex differs. In the orthocortex the macrofibrils are well delineated because of the abundance of nonkeratinous intermacrofibrillar material (matrix) that separates the bundles. The

arrangement of microfibrils within the macrofibrils usually occurs in whorls (Chester, 1962). The rodlike microfibrils are tightly embedded in the structureless (Swift, 1977) or ordered (Spei, 1975) intermicrofibrilliar matrix (Fraser *et al.*, 1975; Chapman, 1975; Kulkarni & Bradbury, 1975; Swift, 1977). In the paracortex the tightly packed macrofibrils are difficult to delineate because so little intermacrofibrillar material exists (Swift, 1977). Paracortex non- keratinous materials are located in the nuclear remnants, the microfibril matrix is more regular and has larger amounts of intermicrofibrillar matrix relative to the microfibrils than in the orthocortex (Bradbury, 1973).

Liquids can more readily flow through the elaborate network of intermacrofibrillar material of the orthocortex than the more regularly packed paracortical microfibril matrix cells. The non-keratinous material in the orthocortex swells easily and is more readily extracted with acids than the matrix of the paracortex (Bradbury, 1973).

The chemical reactivity of the ortho and para cortices is different (Miyamato *et al.*, 1985). The orthocortex is more reactive than the paracortex, has fewer cystine bonds, is more accessible to chemicals, and swells less due to a shorter interchain distance (Bradbury, 1973; Swift, 1977 p. 124). The orthocortex has slightly more charged groups when the pH deviates from the isoelectric point (pH 4.9). Transport of reactants through the intermacrofibrillar

network of the orthocortex is faster than transport in the paracortex. The orthocortex dissolves with acids following alkaline treatments or peracetic acid oxidation (Bradbury, 1973). The only treatment that affects the paracortex more than the orthocortex "is oxidation with peracetic acid which produces more [illegible] groups in the cystine rich paracortex" (Bradbury, 1973).

The amino acid analysis of separated orthocortical and paracortical cells show differences in composition. Solubilization of wool by reduction, followed by alkylation, gives rise to a complex mixture of proteins (See 2.5.3) that can be separated into low sulfur (SCMK-A), rich sulfur (SCMK-B), and a glycine and tyrosine rich fraction (Fraser, 1973, Bauter, 1985, Gillespie & Frenkel, 1973, Kulkarni & Bradbury, 1974). Microfibrils are low sulfur proteins and the matrix is high sulfur protein (Speakman, 1985). A mixture of high sulfur and hydrophobic high glycine-tyrosine proteins are found in the matrix between microfibrils (Speakman, 1985).

The effects of swelling and mechanical properties of wool imply that the majority of disulfide bonds are intramolecular, and there may be a regular arrangement of intramolecular disulfide cross-links in the fiber (Speakman, 1985). Ultra-high sulfur proteins are increased with sulfur-enriched diet (Marshall *et al.*, 1985*a*; Maclaren & Milligan, 1981). An increase in sulfur does not change longitudinal mechanical properties nor rigidity, however, less rapid decay of torque and fewer crimps appear in fibers (Speakman,

1985, Sakamoto, 1985).

Speakman suggests that the heterogeneity of amino acid sequences may prevent crystalline or regular molecular packing. Small differences within each family of proteins may come from different types of cells within the fiber. The heterogeneity of the amino acid sequence may contribute to wool's resistance to bacterial and fungal attack (Speakman, 1985).

2.4.5 Medulla

The centrally located, open textured, vaculated medulla may appear as an axial stream of cells, (Speakman, 1985) or a spongy central core (Koenig, 1977). In different species, and within species, the patterns of medullae may be continuous or interrupted (Bradbury, 1985; Wildman, 1953). The cell cavities may be formed during protein synthesis and follicular development. Subsequent desiccation of the fiber during keratinization results in open, light, stiff intracellular medullae structures with air spaces that contribute to the biological function of hair and wool (Bradbury, 1985).

Protein analyses indicate large amounts of citrulline and glutamic acid are found in the medulla (Speakman, 1985). The chemical stability to alkali and other keratinolytic agents is attributed to $\epsilon(\gamma$-glutamyl) lysine cross-linkages (Speakman, 1985; Fraser *et al.*, 1972; Bradbury, 1985).

2.4.6 Summary

Characteristics of wool, a biological composite, are derived from its unique chemical composition. Entry of chemicals is generally through the hydrophilic non-keratinous matter where cationic and anionic amino acid residues are present in large quantities (Hojo, 1985). Chemicals are dispersed throughout the fiber via the cell membrance complex (Miyamoto, *et al.*, 1985; Weigmann *et al.*, 1985). Dyes and other water soluble reagents enter the fiber through junctions of cuticle cells and diffuse through the lightly cross-linked non-keratinous endocuticle, intercellular cement and intermacrofibrillar regions. As diffusion continues reagents enter the sulfur rich exocuticle and macrofibrils (Leeder, 1985; Feldtman & Leeder, 1984; 1985). Therefore, any treatment such as hydrolysis or oxidation significantly promotes deterioration of the mechanical properties because of the transport of degradation agents through the CMC (Fuchenbush & Bauman, 1985; Ebert & Zander, 1985).

2.5 CHEMICAL REACTIVITY

2.5.1 Histochemistry

It is generally recognized from studies of protein derivatives extracted from keratin fibers that the wool fiber is

among the most heterogeneous materials to be subjected to physical and chemical studies (Asquith & Leon, 1977). In fact, the heterogenous mixture of proteins is only exceeded by immunoglobulins (Fraser *et al.*, 1973; Bradbury, 1973).

The chemical composition of the various morphological components of wool has been investigated by three main approaches (Swift, 1977).

1. Dissolution of the fiber and subsequent attempts to assign fractions to their morphological origin.
2. Microscopic techniques to establish which morphological components dissolve or are separated with given treatments
3. Selective staining with microscopic examination.

2.5.2 Selective Staining

Metal-staining procedures using osmium tetroxide (Kaplin & Marzinkowski, 1983), silver nitrate, ammoniacal silver salts, and potassium permanganate reveal the detailed internal structure of thin sectioned keratin fibers (Hojo, 1985). Many metal stains bind to the cystine rich exocuticle, A layer, and intermicrofibrillar matrix. Three cystine and cysteine specific methods are the organomercurial halide, silver-methenamine and S-carboxymethylation uranyl reactions. The distribution of cystine is highest in the exocuticle and A layer. There is little or no cystine in the endocuticle, the CMC, the amorphous medullary protein, the nuclear remnants, and

intermacrofibrillar matrix. Swift (1985) states that cystine is present in the "macrofibrils but is distributed in the intermicrofibrillar matrix and not in the microfibrils". Others question the assumption (Meichelbeck & Spei, 1975). The distribution of cystine defines important fiber characteristics.

Staining with phosphotungstic acid (PTA), and complex of silver with N-acetyl homocysteine thiolactone (AHCTL) are complementary methods of staining (Swift, 1985). PTA binds to the free cationic amino groups of the protein with a type of O-P bond both PTA and AHCTL stain fiber components with high concentrations of free amino and basic groups. The endocuticle, the intercellular membrane cements of the cuticle and cortex (δ-band), nuclear remnants, and the intermacrofibrillar matrix are deeply stained (Swift, 1985). The cystine rich exocuticle, A-layer and macrofibrils are unstained.

Uranyl acetate stain acts as a basic dye and binds to carboxyl groups. Very high dye uptakes are reported in S-carboxymethlyated keratin fiber sections (Swift, 1985). High concentrations are found in the δ-band, endocuticle, nuclear remnants, and intermacrofibrillar matrix.

There is a considerable interest in the high glycine-tyrosine proteins (Gillespie & Frenkel, 1975; Hewish & French, 1986). Swift devised a method of converting tyrosyl residues to a diazonium chloride, coupling to α-naphthol and binding platinum to the modified

tyrosine site (Swift, 1985). Preliminary investigations indicate that the thin layers on either side of the cuticle cell complex are sites of tyrosine containing proteins.

2.5.3 Extraction and Fractioning

By using selective reagents researchers are able to separate wool into a large number of different proteins. To avoid cleavage of the peptide bonds and amide side chains reagents which selectively cleave the disulfide crosslinks are used. Soluble proteins were first extracted by Goddard and Michaelis (1934-36) using reduction with thioglycollate, 2 mercaptoethanol or tributyl phosphine and carboxymethylation with iodoacetate (Crewther *et al.*, 1965; Maclaren & Milligan, 1981).

Extracted soluble proteins can be separated into three fractions; the low-sulfur, S-carboxymethyl kerateine A (SCMKA), the high-sulfur, S-carboxymethyl kerateine B (SCMKB), and the high glycine-tyrosine (Gly/tyr) matrix. Goddard & Michaelis' nomenclature of *kerateines* for the reduced form of S-carboxymethyl keratin has been adopted. Sulfonic acid derivatives of proteins described by Alexander & Earland (1930), are known as α-keratose (low-sulfur, equivalent to SCMKA), β-keratose (non-keratinous endocuticle, resistant membranes, and nuclear remnants), and γ-keratose (high-sulfur, equivalent to SCMKB). Crewther (1965) provides an extensive literature review describing the preparation

and separation of fractions for amino acid sequencing and analysis The tentative morphological distribution of the soluble fractions within the wool fiber is given in Table 2.5.3.

The generalized assignment of soluble proteins to morphological components is not absolute. The matrix is suggested to be composed of high sulfur, high glycine/tyrosine proteins and perhaps some low sulfur proteins (Bradbury, 1973). The isolation of completely homogeneous low-sulfur fractions has proved to be an

Table 2.5.3 Location of Proteins in Wool Fibers

Soluble proteins	Morphological component
Low-sulfur proteins	protofibrils
High-sulfur proteins	matrix, A layer
High gly/tyr protein	exocuticle and matrix
Insoluble residue	nuclear remnants, endocuticle resistant membranes

intractable problem, however, the protofibrils are known to be low-sulfur proteins. Separate filaments, which are presumed to be single protofibrils or coiled-coil (two or three stranded) ropes, have been observed in fragment preparations (Swift, 1977). The protofibrils have been frayed into smaller units which are thought to

to fibrous-protein chains. Bradbury (1973) comments "one of the interesting facets of structural protein chemistry (is) that although x-ray studies of keratins contributed greatly to the development of structures such as the α-helix for proteins, ...it is the family of proteins about which least is known".

Although the isolation of homogeneous low sulfur proteins is slow, advances in the characterization of the α-helix portion of the low-sulfur fractions is progressing (Crewther & Dowling 1971). The initial Type I and Type II partial sequencing, by Crewther in 1977, shows no evidence of short-period repeating sequences and "cast grave doubts on theories of keratin structure which rely on postulated periodicities" (Lindley, 1977). There is a probability that every seventh residue has a hydrophobic side chain which could infer the possibility of a coiled-coil keratin structure (Lindley, 1977). New interpretation of x-ray diffraction patterns utilizing low-angle near-meridional as well as meridional spacings indicates that there are a nonintegral number of structural units per turn in the coiled helix and suggests that the low-angle patterns are "largely determined by projections from the surface of the microfibril" (Lindley, 1985). Further indirect evidence suggests that β-crystallites are derived from short lengths of the α-helix. Coiled coils with anti-parallel chains are favored models (Lindley, 1977). The low-sulfur proteins of wool have a 50 per cent helical conformation in aqueous solutions. All other protein groups in wool

have no helical char[illegible] (Maclaren & Milligan 1981).

2.5.4 Microscopic Monitoring Techniques

Chemical reactivity can be monitored by a variety of stains and microscopic techniques in addition to the staining techniques with metals and reagents discussed in the previous section. SEM and the light microscope are used to characterize damage to the morphological components of wool fibers by acids, oxidants and alkali Scanning electron microscopes interfaced with computing devices enable elemental x-ray analyses of introduced elements (Hayat, 1974; Sikorski *et al.*, 1971, Zhao *et al.*, 1986; Zeronian *et al.*, 1986).

2.5.5 Acid Hydrolyis

roteins are amphoteric, and capable of combining with acids and bases. Since free amino acids exist in the ionized or zwitterion form, the acidic and basic amino acid residues in protein fibers are usually ionized in neutral solution (Von Bergen, 1963). Uptake of acid converts carboxylate anions to carboxylic acid groups and alkali uptake results in a conversion of the amino and guanidino groups to an un-ionized state (Maclaren and Milligan, 1981). The acid binding capacity of wool is 810 to 860 μmol g[illegible] which represents the sum of glutamic acid, aspartic acid, and the C-terminal residues (Maclaren & Milligan, 1981). Since wool is a complex mixture of heterogenous proteins different morphological components react with

acids di[illegible]ly (Von Bergen, 1963). Although wool is able to combine with acids and appears to suffer little apparent alteration up to its binding capacity, mineral and organic acids can cause severe damage (Larose, 1955; Nossar & Chaikin, 1975). Carboxylic acids form hydrogen bonds with functional and peptide groups and cause swelling (D'Arcy & Watt, 1966). Wool will absorb five times more formic acid than water (Maclaren & Milligan, 1981, Watt, 1964, Cook & Fleishchfresser, 1984).

During partial hydrolysis four main reactions occur (Maclaren & Milligan, 1981).

1. cleavage of amide side-chains
2. cleavage of peptide bonds
3. N—>O acyl shift serine and threonine
4. disulfide interchange

The cleavage of peptide bonds is not random. The more sensitive bonds are the first to be released. Cystine peptides are rapidly rearranged in concentrated acid. The interchange appears to involve the rearrangement of components along the peptide chain.

Acid hydrolysis with 6M HCl is used to cleave peptide bonds for sequencing (Crewther, 1965; Fraser & MaRae, 1972). Tryptophan is completely destroyed and asparagine and glutamine are converted to aspartic and glutamic acid. Serine and threonine are found in significant amounts in partial hydrolysates. The peptide bonds of

serine and threonine residues are made olytically more labile by N ->O acyl migration which forms an ester bond. The bonds formed by aspartic acid are suceptible to weak acids, hence, aspartic acid is the first amino acid to be released during hydrolysis. The peptide bonds adjacent to oxidized cystine residues (cysteic acid) are very sensitive t acid hydrolysis (Asquith & Leon, 1977).

The release of cuticle cells in formic and trichloracetic acid is probably due to the dissolution of proteins in the membranes surrounding the cuticle cells rather than peptide hydrolysis (Maclaren & Milligan, 1981). More vigorous treatments result in the hydrolysis of all the proteins in the cell membranes which results in separation of cortical cells as well as cuticle cells.

2.5.6 Chemical Reactions of Wool

The chemical reactions of wool are extremely complex. An overview of the principal chemical reactions is given in Table 2.5.6

2.5.7 Mechanical Properties

Many models have been developed to explain the mechanical properties of keratin fibers (Feughelman, 1978; Morton & Hearle, Hearle, 1963). By using x-ray diffraction, optical, and electri measurements applied to fiber in various states and environments researchers are unravelling the relationship between molecular organization and structural properties which control the mechanical

Table 2.5.6 Principal [illegible] Reactions [illegible] Common Reagents

Reaction	Reagent	[illegible] Residue
Oxidation	peracids peracetic	[illegible]e, methionine, [illegible]ophan
	peroxides	
	[illegible]lorine	[illegible]rosine, tryptophan, [illegible]eptide bonds
Acids	mineral acids	amides and peptides aspartic and glutamic acids, threonine, serine, trypthophan
	carboxylic acids	(swelling reagents)
Esterification	alcohol with acid catalyst	aspartic and glutamic acid residue amide groups
	methyl halides	tyrosine, serine
Reduction	thiols phosphines	cystine, cysteines, tyrosine, other residues
Alkylation	alkyl halides epoxides ethylenic derivatives	[illegible]ysteine, lysine, serine, histidine, methionine, threonine, tryrosine, aspartic and glutamic
Acylation	acid chlorides and anhydrides active esters amides	hydroxyl groups, serine threonine, tryrosine, and arginine, lysine, cysteine (S-, O-, and N-sites)

(Maclaren & Milligan, 1981; Speakman, 1985)

properties of the fiber (Menefee, 1971; Weigmann & Dansizer, 1971; Sikorski, 1975; Feugelman, 1978). By identifying the assemblies and molecular components responsible for the mechanical variation which occurs with temperature, moisture, and physical and chemical changes, the causative factors for failure can be elucidated (Bendit, 1960; 1975; Sketchy, 1960; Speakman & Hirst, 1933; Lipson, 1975; Ross *et al.*, 1986; Rottenberry, 1979).

Many proposed theories to explain the mechanical properties of wool fibers are based on ideas of the fine structure of keratin fibers (Morton & Hearle, 1975; Feughelman, 1971). These include the micellar theory, proposed by Nägeli over 100 years ago (Hearle, 1963), the fringed micelle theory, fibrils as crystallites, fibrils as alternating crystalline and non-crystalline regions (Hearle, 1963), and the brick and mortar theory (Leeder, 1986). Conformational analyses based on models such as α - β transformations developed from equatorial and meridional x-ray diffraction patterns (Dobb & Rogers, 1969; Dobb & Murray, 1975), give some insight into mechanical and chemical behaviour (Elliott, 1952). The voluminous literature hypothesizing mechanical response is contradictory (albeit useful and often founded on theories that ignore the complexity of the keratin fiber. To attribute the changes to any particular event or theory would be presumptuous (Morton & Hearle, 1975).

Speakman (1977) demonstrated that the longitudinal properties of wool vary with temperature, relative humidity, and time. All

fibrous α-keratins have similiar stress-strain curves with three distinct regions of mechanical behaviour; a pre yield region (often referred to as the Hookean region), the yield, and the post-yield region (Feughelman, 1980; 1971). Fibers extended for a limited time (an hour) in the yield region recover original mechanical properties in water within an hour at 52°C, or overnight at room temperature (Feughelman, 1980).

In the pre-yield regions the opposition to extension is proposed to be hydrogen bonds between helical turns (Feughelman, 1980) which are re-established in water. At temperatures above 55°C, Speakman (1985) concurred that degradation in wet wool fibers in the pre-yield region was via "the mechanism of sulphydryl disulphide interchange" or disulphide bond breakdown (Feughelman, 1980). Mechanically irreversible breakdown of disulfide bonds occurs in extensions beyond the yield regions. The $\alpha \longleftrightarrow \beta$ transformation equilibrium occurring in the yield regions breaks down at high temperatures and when the disulfide cross-links stabilizing the α-phase are ruptured (Feughleman, 1980).

Longitudinal mechanical properties seem to depend upon the α-helical material of the microfibrils and the water sensitive matrix. The side chains of the helices are water sensitive and interact with the matrix (Feughleman, 1980). Changes in extension are suggested to be governed by changes in hydrogen bonding, ionic bonding, electro-attractive and repelling forces, and covalent

bonding with introduced molecules (Speakman & Hirst, 1933; Speakman, 1985; Shaw, 1985; Szuctz, 1971; Morton & Hearle, 1975; Feughleman & Reis, 1967; Algie, 1979; Kopke, 1979; Robinson, 1981-83). Factors changing the extension of wool fibers will be discussed in Sec 6.4.

2.5.8 Additional Sources of Information

The *Textile Research Journal*, and *Journal of the Textile Institute* contain many wool oriented papers. The quinquennial international wool textile conferences are a major source of information about wool structure, chemical reactivity, and new developments in dye and processing technology.

The *Proceedings of the* (1st to 7th) *International Wool Textile Research Conference*, Australia, 1955; Harrogate U.K.,1960; Paris, 1965; San Franciso, 1970; Aachen, 1975; Pretoria, 1980; and Tokyo, 1985, are available as published proceedings. Excellent wool science reviews are published in *Advances in Protein Chemistry, Vol. 20*, and *Vol.27*. The most concise reference book on wool chemistry and reactivity is Maclaren and Milligan's (1981) book *Wool Science, The Chemical Reactivity of the Wool Fibre*. Relevant information from these sources is included in the discussion of the results in Chapter 6.

3. CASE STUDY

3.1. Dichlorvos Fumigation of a Textile Collection

Ten years ago the caretakers of a large collection of artifacts representing the history of the city of Edmonton were in a dilemma. Repeated infestations of insects were ruining the collection. It was not unusual to find the contents of a storage box devoured by insects such as clothes moths (*Tinea pellionella* or *Tineola bisselliella)* and dermestid beetles. Informants described the infested collection containers as "the whole box was a grey moving mass". Repeated spraying with a variety of insecticides only suppressed the infestations for a short period of time

In desperation the collection was transferred to a different building, a building infested with *Lepisma saccharina* (silverfish). Administrators consulted a National Museum organization for assistance in pest management. The consultant's advice was to encapsulate the textile collection with Vapona®. The space fumigant offered a solution to the sporadic and devastating outbreaks by providing continuous fumigation. All of the historic textile collection was transferred to a subterranean dark vault isolated from the major collection. The isolated textiles were enclosed in polyethylene bags with a piece of Vapona® ≈ 2.5 cm^2. Despite employee complaints of headaches and nausea the collection was systematically bagged and placed in the vault (circa 1980).

The author's first experience with Vapona® was during a

teaching assistant assignment in the autumn of 1986. The assignment was to assist conservation students with the design and implementation of a new textile storage area in the Edmonton collection center. A sweet musty ester-like odor emanating from the storage vault signalled an alarm response. It was immediately apparent that the environment in the vault was not suitable for textile and fur storage. Closer scrutiny of the encapsulated artifacts confirmed suspicions. The accession cards enclosed with the artifacts were being dissolved and discolored by an acid. Acid catalyzed hydrolysis is all too familiar to paper conservators and archivists.

A high concentration of Vapona® within the polyethylene bags created a microclimate deleterious to objects. By-products of the break-down of Vapona® increased the relative humidity (RH) in the polyethylene bags. Some of the textiles were moist and others exhibited signs of moisture. Brown oxidized cellulose water lines were present on both cellulosic textiles and paper labels. Some physical loss of paper and leather objects occurred in areas in contact with the Vapona® strips. Close contact with Vapona® caused tendering of fibers, loss of plastizers in plastics, dissolution of leather, color change in dyes, and embrittlement. The humid stagnant environment was conducive to fungal proliferation. Stains, caused by mildew colonies, scarred many artifacts. A musty odur permeated many of the furs. Follicle damage on some furs resulted in major

hair loss. Some of the damaged textiles were deaccessioned and destroyed. The condition of encapsulated articles was not recorded when the artifacts were removed from the polyethlyene bags. Although the author rescued a few items for research and, with another student, photographed the storage facility and some of the damaged objects, basic individual documentary information was not recorded.

Scanning electron micrographs (SEM), fluorescent microscopy, and energy dispersive X-ray analysis (EDXA) was performed on a white and pink wool knitted baby's bonnet. The EDX graph (Figure 3.1.1) indicates high phosphorus and chlorine levels. (Chlorine and phosphorus were recorded in EDX analysis of laboratory fumigated wool fibers; See Sec 6.10.) Although it is unlikely, phosphorus may be an artifact from phosphate detergents. Chlorine and phosphorus peaks are probably indicative of dichlorvos absorption and retention. The wool fibers were removed from Vapona® in the autumn of 1986. EDX analysis was performed in the spring of 1988.

SEM micrographs from fibers in close proximity to the Vapona® strip indicate severe fiber damage (Plate 3.1.1). A viscous-like material encircles a slightly eroded and deformed fiber (Plate 3.1.1). A denuded fiber fiber is observed in a corner of the micrograph (Plate 3.1.1). The fiber in Plate 3.1.2 has lost scale definition (Plate 3.1.2). A damaged fiber is observed in Plate 3.1.3. The cuticular surface appears to have lost all definition. The

fluorescent photomicrograph (Plate 3.1.4) indicates that localized acid damage contributed to degradation. It is apparent that an investigation of the effect of dichlorvos resin strips on keratin fibers is imperative.

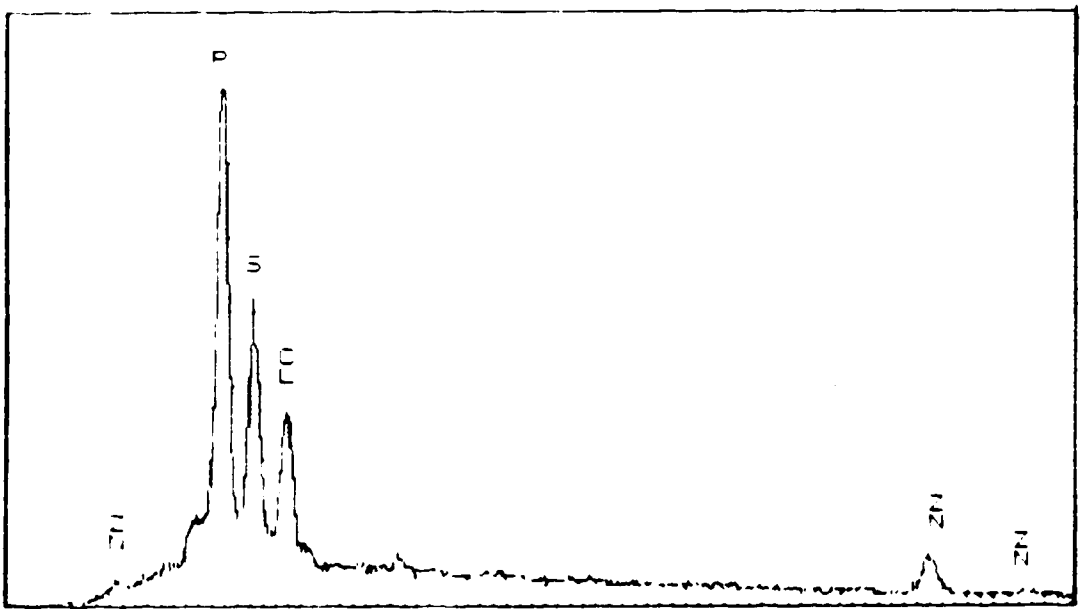


Figure 3.1.1 EDXA graph of fibers from wool bonnet fumigated with Vapona® at ambient temperature for 1-5 years.

Plate 3.1.1 Viscous-like material surrounds wool fiber fumigated with Vapona® at ambient temperature for 1-5 years.

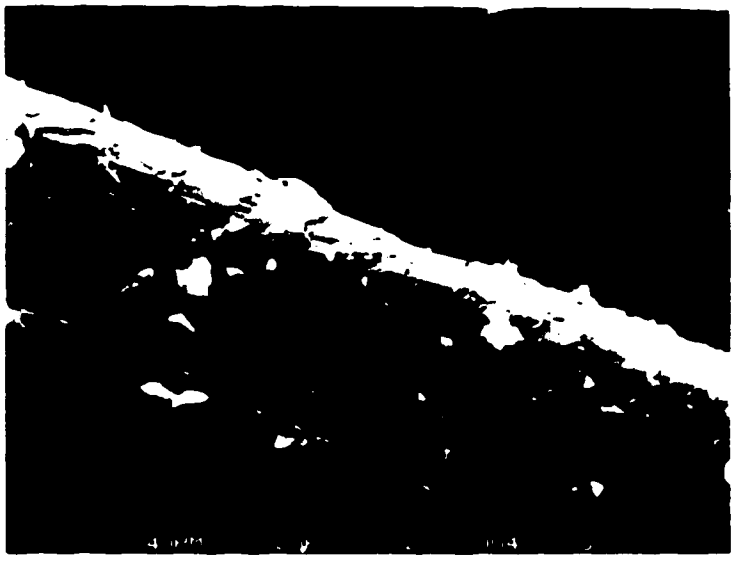


Plate 3.1.2 Scale loss in wool fiber fumigated with Vapona® at ambient temperature for 1-5 years.

Plate 3.1.3 Fracture in embrittled severely damaged fiber from wool artifact fumigated *in situ* with Vapona®.

Plate 3.1.4 Fluorescent photomicrograph of wool fiber from bonnet. The orange-red spots indicate acid damage.

4. MATERIALS AND METHODS

4.1 Design

Ideally the effects of chemicals used in fumigation would be assessed by observing changes in historic textiles *in situ*. However, the rate of reactions and morphological changes that occur with naturally aged fibers exposed to continuous fumigation at 20°C (+2) is often too slow to detect damage within the temporal restraints imposed upon research projects. Since even carefully controlled laboratory experiments produce conditions different from *in situ* conditions it was necessary to find a compromise that would allow a reasonable facsimile of the fumigation processes reported in museum literature.

The temperature for accelerated fumigation was higher than the recommended environmental standard of 21°C for museum objects (Thomson, 1981). Although the saturation concentration of dichlorvos chosen for accelerated fumigation was higher than that recommended by the manufacturer and by the regulatory agencies (WHO, 1986; OHAS, 1985) it was not greater than some of the concentrations reported in the literature (Ryckmann, 1969), and in the case study. A compromise temperature of 50°C was chosen because it was the reported baseline temperature for thermal degradation of wool (Maclaren & Milligan, 1981). In a pilot study, 50°C did not cause the pesticide strip to bleed as reported in the literature and by informants working at the case study collection center. However, some thermal yellowing

occurred at 50°C, therefore, most tests were performed with heated and unheated controls.

The case study (Sec 4.1) provided examples of keratin fibers exposed to high concentrations of Vapona® for 1-5 years. The damage to the wool and fur fibers was compared to that sustained by wool during laboratory experiments. Fibers exposed to long-term *in situ* fumigation were used to assess the feasibility of the methods selected for this study.

4.2 Fiber

The fibers examined were from greasy Corriedale and Merino fleeces with a mean diameter of 28μm and 21μm respectively. The diameter and condition of the fibers was estimated using a modified random sample of 20 fields. The fibers were washed 6 times in 0.5% Shurgain anionic detergent at 25 ±3°C, thoroughly rinsed with at least 10 exchanges of distilled water with a pH 6.5 and air dried. Although both wools retained a greasy hand, the Merino wool felt oilier than the Corriedale and was slightly whiter. Merino fibers were used for the 35 day exposures. Merino 2/22 commercial yarn was used for tensile strength testing. The fiber and yarn were conditioned for at least 6 hours at 65% RH and 21°C before fumigation (King & Wood, 1965).

A second set of fibers was soaked for 20 minutes in methanol and rinsed in distilled water to remove methanol soluble

contaminants. Methanol/Shurgain scoured fibers and commercially scoured handspun Merino wool yarn were used for auxiliary testing.

4.3 Sampling Procedure

The samples were randomly selected from throughout the fleece. No fibers were chosen that appeared to be stained or damaged. Tippy ends were trimmed from the fleece.

4.4 Fumigant

The fumigant was a dichlorvos resin strip manufactured by Ciba-Geigy and distributed under the trade name S.W.A.T.® Dichlorvos is dispersed in plasticized polyvinyl chloride.

4.5 Fumigation Procedure

One-half a fumigation strip (50 g) was placed in the lower chamber of each 2.5 L glass desiccator. Placing the DDVP strip in the lower portion eliminated dripping or bleeding, and direct contact with the fibers. The conditioned fibers (10 g) and yarn (1 g) were weighed and placed in the upper chamber. Fiber bundles were separated and loosely fluffed to allow maximum fumigant circulation. Three replications were made for each time period.

The desiccators were sealed with Teflon tape and placed in a dark oven at 50°C for 7, 21, and 35 days. Control samples sealed in desiccators without dichlorvos were heated for the same length of time.

4.6 Sorption of Fumigant

Tests to evaluate the extent of fumigant absorption were conducted at ambient temperature and at 50°C. Ambient temperature samples were withdrawn at 22.5 [illegible] min (0, 22.5, 45, 90...). The last samples were withdrawn after 1175 hours of exposure to the fumigant. Heated sorption samples were withdrawn at 6, 12, 18, 24 h and 2, 3, 5, 7, 14, and 21 days. Shurgain scoured fibers were used for ambient temperature sorption tests. Shurgain, methanol/Shurgain and commercially scoured fibers were used for heated sorption tests. The strips of S.W.A.T.® were weighed and placed in the lower chamber of 2.5 L Pyrex® desiccators. Jars were sealed with Telflon tape and placed in the fume hood, or in the oven. Samples were withdrawn at selected intervals and placed in glass screw top vials until sorption and desorption EDX analysis was performed. The vials were sealed with Teflon tape between the vial aperture and lid, and around the lid. Samples for sorption were placed in glass jars and frozen until analysis. Air desorption samples were mounted on standard aluminum SEM stubs and placed in a fume hood to desorb for one week. A 20 minute wash in methanol or distilled water was used for aqueous desorption.

5. Test Methods

5.1 Colorimetry

Color changes in the wool fibers were determined with a Hunterlab Tristimulus Colorimeter Model D 25M-9 recording spectrophotometer (Hunterlab Model D25 Optical Sensor) using AATCC test method 153-1985. The instrument was interfaced with a computing device which converted reflectance data to CIE tristimulus values. Colour difference, ΔE, was measured in CIELAB units using the CIE L* a* b* scale and color difference formula:

$$\Delta E_{CIELAB} = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2}$$

Where:

$$L^* = 25(100Y/Y_0)^{1/3} - 16$$

$$a^* = 500\ [(X/X_0)^{1/3} - (Y/Y_0)^{1/3}]$$

$$b^* = 200\ [(Y/Y^0)^{1/3} - (Z/Z_0)^{1/3}]$$

The parameters X, Y and Z are tristimulus values and X_0, Y_0 and Z_0 are tristimulus values for a perfect diffuser for the illuminant used (Hunter, n.d.). The parameters L^*, a^*, and b^* are defined as:

L^* represents reflectance (white$^+$/black$^-$)

a^* (red$^+$/green$^-$), and

b^* (yellow$^+$/blue$^-$) areas of the spectrum.

The fibers were formed into a pad and compressed between a white tile and clear glass plate. Each sample was measured 5 times, with rotation and repacking of fibers between measurements (Hunter, n.d.).

5.2 Breaking load and yarn elongation

The breaking load and elongation of Merino yarn were determined in accordance with ASTM Method D 2256-80, option A, single strand method after conditioning at 65% RH and 21^{0}C. Samples of yarn were randomly selected from along the skein length. A total of 45 samples from the controls and each of the 7, 21, and 35 day fumigation exposures (135 yarns per heating period) were tested on the Instron Model 4202. The rate of elongation was 250 mm/min and gage length 25 cm. Yarn clamps were used. No jaw breaks occurred.

Although the test method specifies a breaking time of 20 $\pm$3 sec for single yarns the great variation within yarn sets prompted the decision to use a standard elongation rate of 250 mm/min (Tweedie *et al.*, 1959; Taylor, 1985).

5.3 pH of Aqueous Extracts

The pH of aqueous extracts of two samples from each of the three replications from the 7, 21, and 35 day fumigant periods was measured using a Pope Model 1500 pH/ion meter in automatic temperature mode, or a Fisher Accumet Model 230 pH/ion meter. The

instrument was calibrated using potassium biphthalate buffer (pH 4) and potassium phosphate buffer (pH 7).

Specimens were prepared as directed in ASTM Test Method D2165-78. The wool fibers were randomly selected Extraction was at room temperature with 0.1 *N* NaCl using 1 gram of wool to 60 ml of the solution.

5.4 Lead Acetate Test

Change in the oxidation state of sulfur was determined by using lead acetate staining as described by Weaver (1984) and Garner (1966). A solution of 6 g/l of lead acetate in 500 ml distilled water was acidified with 4 ml of glacial acetic acid. Each 0.6 g sample of wool was boiled for 45 minutes in 60 ml of acidified lead acetate. Staining occurs when lead acetate reacts with R-SSH Harris & Smith, 1937).

$$R\text{-}SSH + Pb(CH_3COO)_2 \longrightarrow PbS_{ppt} \quad \text{(dark brown stain)}$$

The extent of staining was measured with the Hunterlab Tristimulus Colorimeter. Staining was evaluated visually.

5.5 Allwörden reaction

The Allwörden (Herbig) reaction was used to determine the integrity of the epicuticle. The Meeuse bromine test method was used (Garner, 1966). A solution of bromine water (1:20 [$Br:H_2O$]) was prepared in the fume hood. A glass slide was prepared with ether

rinsed wool fibers and a drop of reagent. The appearance of raised bubbles along the fiber was observed using a light microscope. Discreet bubbles formed on the fiber within 2-5 min.

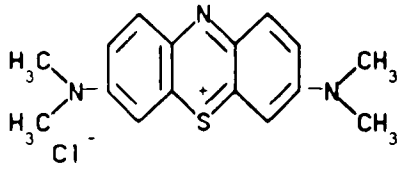
5.6 Krais-Viertel

The Krais-Viertel test as described by Merkel (Weaver, 1984) and Mercer and Golden (1953) was used to determine the reactivity of the fibers in alkaline medium, and to produce the Allwörden reaction. Ether cleaned fibers cut to 0.2 mm in length placed in Krais-Viertel solution (20g KOH in 50ml concentrated NH_4OH) on a glass slide were examined with a standard light microscope. The rate of dissolution and formation of swollen bubbles were compared with undamaged wool of the same mean diameter. Bubble formation within 10 to 30 minutes indicates acid damage. Acid damaged fibers usually disintegrate more rapidly than undamaged fibers. Although bromine water is a better reagent for observing the epicuticle the Krais-Viertel reagent is a better indicator of chemical reactivity in an alkaline medium.

5.7 Methylene Blue Staining

Absorption of the basic dye Methylene Blue (C. I. Basic Blue 9) was used to determine the extent of oxidative and cuticle damage. A modification of the Kornreich FB/3 (acidified saturated solution) and Meeuse FB/4 (0.002% dye:H_2O) methods described by Garner (1966) was used for microscopic investigation of oxidative damage. A stock solution (1g dye:100 ml H_2O) acidified with 5 ml 3*N* H_2SO_4 was

prepared. Colorometric analysis was estimated by using 1:10 ratio of wool in a 1:100 dilution of Kornreich stock (Garner, 1966) warmed to $60^{0}C$. The fibers and Methylene Blue solution were agitated for 60 minute on a Burrell Wrist Action Shaker Model 75. The color difference was measured as described in Sec 5.1.



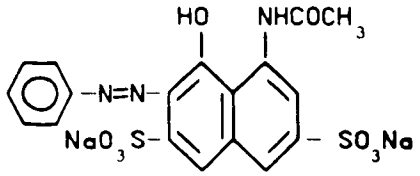
Methylene Blue

5.8 Kiton Red Staining

Kiton red staining with the acid dye, C.I. Acid Red 1, was carried out using the method proposed by Merkel (Weaver, 1984). A solution of 0.152 g dye in 500 ml H_2O acidified with 25 ml 0.1 *N* HCl was prepared. The wool fibers (0.6g) were pretreated for 10 minutes with 50 ml borax solution (10g borax in 500 ml H_2O) and steeped in 0.1% acetic acid for 5 minutes before rinsing and staining. The pretreated fibers were placed in sealed flasks and agitated with 100 ml of Kiton Red for 1 h on the Burrell Wrist Action Shaker. The color differences of wool stained with Kiton Red were measured on a Hunterlab Tristimulus Colormeter.

Fibers were examined with a microscop to determine the

uniformity of fibers and to locate areas of localized damage. Since the coloration of undamaged wool is proportional to the diameter of the wool, preliminary tests on damaged wool fibers were carried out in parallel with known undamaged fibers of the same mean diameter.

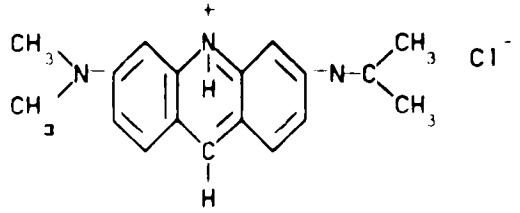


Kiton Red: C.I.-18050 Acid Red 1

5.9 Acridine Orange

The basic fluorescent dye, C.I Basic Orange 14, was used to determine if localized acid damage occurs during fumigation. The method used was a modification of Nossar's method (Nossar *et al.*, 1973) and the procedures described by Zhao for localized acid damage in carbonized wool (Zhao *et al.*, 1986). The procedure used for dyeing is described by Appleyard (1965). Wool fibers (0.3 g) were boiled (3 min) in 10ml of dye solution (0.1%) and air dried. Specimens were mounted in Clearmount and cured on a CCGE Slide Warmer at 45°C for 18 hours. A Zeiss III RS microscope with a fluorescent light source, and standard 25X and 40X objectives was used. Tungsten

160 film was used for light and fluorescent photomicrographs. A Nomarski interferometer increased contrast (Fatou, 1978).



Acridine Orange (C.I. Basic Orange 14)

5.10 Scanning Electron Microscopy

5.10.1 SEM

Specimens were mounted on aluminium stubs with 2-way stub tape and placed in a silica gel desiccator for 24 hours. The dried specimens were Au sputter coated in the SEMPREP 2 at 1500 volts for 2 minutes and examined in a Cambridge Stereoscan 250. Micrographs were obtained using Ilford or Kodak Plus X-pan 125 ASA film.

5.10.2 X-ray Microanalysis

Elemental content of phosphorus and chlorine in wool fibers was determined by using energy-dispersive spectroscopy (EDX). The analysis was conducted on the Cambridge Stereoscan 250 electron microscope using the software packages M-scan, and Microscan. The

first specimens analysed were mounted on aluminium stubs with 2 way stub tape or plastic rings. To avoid interference subsequent specimens were mounted on precoated carbon stubs with rings. Sorption samples were mounted and directly placed in the vacuum chamber. Desorption specimens were allowed to desorb in air for one week before analyses or washed in methanol or distilled water and dried before mounting. The uncoated specimens were placed in the vacuum chamber where the atmospheric pressure was reduced to 10^{-6} torr. The wavelength dispersive spectra was run on K-line.

The first stage in determining the content of chlorine and phosphorus was to obtain values for the x-ray intensities of sulfur which was assumed to be constant in all wool specimens. A baseline for sulfur established at 7.5 to 7.7 units where 1 unit equals one division on the EDX graph. Values for chlorine and phosphorus were determined by reading their peak heights directly from the EDX grids of the graph when sulfur was 7.5 to 7.7 units. When the sulfur peak did not equal 7.5 to 7.7 units, the proportion of chlorine or phosphorus relative to sulfur at 7.5 units was calculated.

The adhesive was determined to be the primary source of zinc or copper and the stub, a source of aluminum. Carbon stubs were used to reduce interference and the rings replaced adhesives.

The literature reports considerable variation in the sulfur content of wool along the fiber (Lindley, 1977), in adjacent anatomical locations (Marshall, *et al.*, 1985b), among species

(Włochowicz, 1985), with nutrients available during differentation (Marshall *et al.*, 1985a), and with processing (Fuchtenbusch & Baumann, 1985). Since the take-off angle is unkown due to the round, curved, long morphology, rough surface and heterogenous anatomy of wool fibers, the results are not absolute (Hess, 1980; de Bruijn *et al.*, 1987; Fiori *et al.*, 1979). Scattering and secondary excitation of rough specimens makes EDX analysis problematic (Hanlan, 1975; Blakey & Mickelthwaite, 1978).

5.11 Statistical Analysis

5.11.1 Tensile Properties

$SPSS_x$ statistical package was used for analysis of variance on the tensile properties. The Scheffe test was used to identify groups which differed significantly at the 0.05 level.

5.11.2 Color Difference

UANOVA on L^*, a^*, b^*, was used to find significant differences of group means and Scheffe multiple range test was used to identify significant differences of means at the 0.05 level.

6. RESULTS AND DISCUSSION

6.1. Interpretation

Wool's structural and chemical heterogeneity has long stimulated discussion and debate. There are problems with assigning a specific reason or theory to explain chemical and mechanical changes because the biochemistry of wool is complex (Crewther, 1975; Fraser *et al.*, 1972, 1977; Kulkarni & Bradbury 1975; McPhee, 1975). Exposure to reactive chemicals can have a profound effect on the fibers (Bradbury, 1973; Lindley, 1959, 1977; Blakey *et al.*, 1971; Fraser *et al.*, 1980; Gillespie & Frenkel, 1973).

A summary of the test results is given in Table 6.1.1. Detailed results of individual tests and discussion of results are presented in this chapter.

6.2. pH of Aqueous extract

The pH of the aqueous extract of wool fibers is recorded in Table 6.2. Increased exposure to DDVP decreased the pH from pH 5.8 (7 days) to pH 4.4 (21 days) (Figure 6.2.1). A dramatic decrease to pH 3.4 occurred in methanol/Shurgain scoured fibers exposed to the fumigant for 7 days (Figure 6.2.1). The pH of methanol scoured fibers decreased more than the pH of Shurgain scoured fibers. Possible sources of acid during fumigation are dimethyl phosphoric acid, and chlorinated peracetic and acetic acid.

Table 6.1.1 Overview of Test Methods, Purpose of Tests, and Changes in Fibers Exposed to S.W.A.T.® in a Dark Oven at 50°C for 7, 21, and 35 Days.

Test	Test Indicates	Observed change
Load-extension	inter, intra bond change	extension change tensile strength inconclusive
Color change	chemical change	yellowing
pH extract	ionic balance	increased acidity
Lead Acetate	cystine oxidation	oxidation of -SSH
Bromine Water	epicuticle integrity	epicuticle damage
Krais-Vier[illegible]l	acid damage, epicuticle integrity	acid damage epicuticle damage
Methylene Blue	mechanical, acid damage chlorination	change in dye uptake change in ionic charge surface damage
Kiton Red	oxidation, mechanical damage	change in dye uptake oxidation/acid damage
Orange 14 Fluorescence	acid damage	localized acid damage
EDX	presence of elements	sorption of chlorine and phosphorus
SEM	topographical changes	fiber degradation

Table 6.2 pH of Aqueous Extract for Wool Fibers Exposed to Dichlorvos (S.W.A.T®) Resin Strips at 50°C in a Desiccator in the Dark.

Scour	Time (Days)	pH of Aqueous Extract[a] Control[b]	Fumigant
Shurgain scour	7	6.8	5.8
	21	6.3	4.9
	35	5.9	4.4
Methanol/ Shurgain scour	7	6.4	3.4

[a]average of eighteen readings
[b]varibility may be effect of heat, changes in pH of distilled water, and use of Merino fibers for 35 day and 7 day methanol.

Wool fibers have the capacity to absorb 810 to 860μmol g^{-1} of acid at the isolectric point measured by titration with HCl (0.02M) (Maclaren & Milligan, 1981). The acid sorption values agree with the aspartic acid, glutamic acid, and C-terminal content of wool. Electrostatic interactions such as salt links contribute to the cohesion of wool. By breaking electrostatic interactions, increased acid sorption reduces the amount of work required to stretch fibers (Maclaren & Milligan, 1981; Speakman & Hirst, 1933; Lipson, 1975). An increase in acid sorption also affects the dyeing capacity of acid and basic dyes or stains (Bird, 1972; Whewell *et al.*, 1971; Gurr *et*

al., 1974; Peters, 1975). Changes in pH are discussed in the appropriate sections.

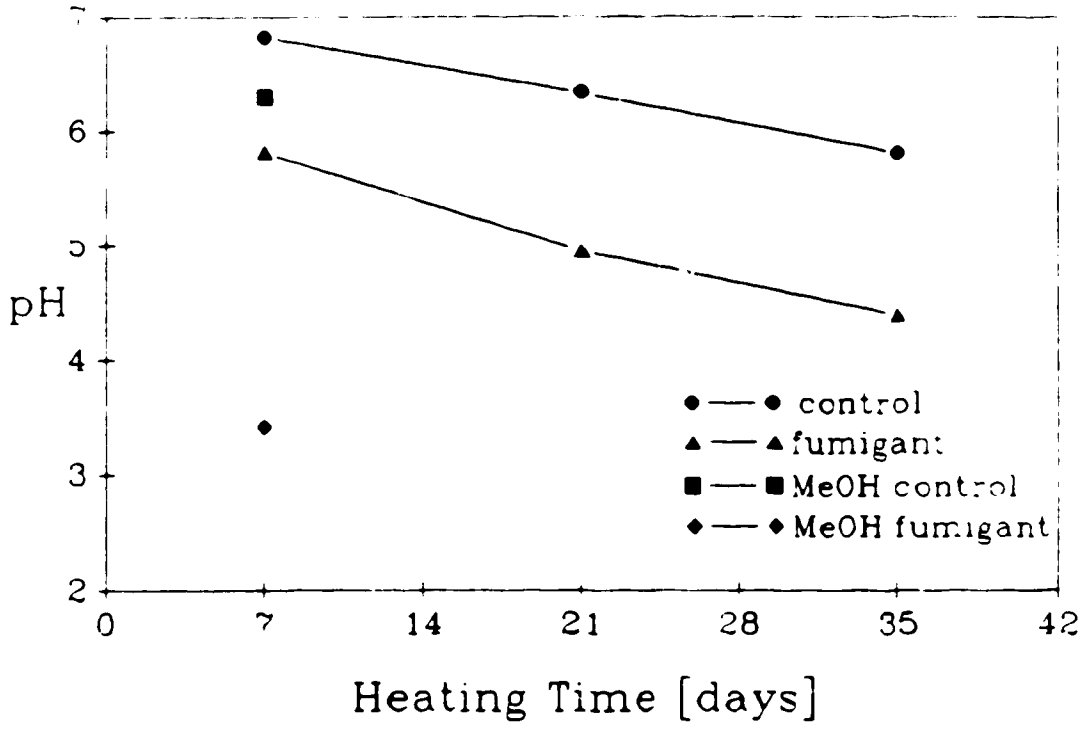


Figure 6.2.1 The pH of Aqueous Extract of Shurgain and Shurgain/Methanol Scoured Fibers Fumigated with S.W.A.T.® at 50°C in a Dark Oven.

6.3 Color Change

Color differences of wool fibers fumigated at 50° C for 7, 21, and 35 days in the dark are recorded in Table 6.3.1. Wool fibers exposed to a saturated concentration of dichlorvos at 50°C showed a significant change in color at the α=0.05 level (ΔE 4.43 to ΔE 28.08 CIELAB units). Visible changes were observed within 2 days of exposure. A slight color difference (ΔE 2.04 to ΔE 3.68 CIELAB units) occurred in the control samples heated without dichlorvos in a desiccator for 7, 21, and 35 days (Figures 6.3.1. & 6.3.2).

The fumigated wool's color changed from a creamy white to a soft yellow ochre within 7 days. A plot of b* (Appendix 1) color change ($blue^-/yellow^+$) during fumigation indicates that the fumigant tends to increase yellowness with time (Figure 6.3.3). The slight decrease in yellowness at 35 days may be due to increased wool acidity. Norton and Nicholls (1960) report that low pH tends to decrease yellowness.

Fumigated fibers given a methanol pre-soak to remove surface contaminants yellowed more rapidly than Shurgain scoured fibers (Figure 6.3.4). More yellowness occurred in 2 days (ΔE 11.84 CIELAB units) than occurred in 21 days (ΔE 8.09 CIELAB units) with the Shurgain wash and distilled water rinse (cf. Figure 6.3.3 to 6.3.4). Although the methanol/Shurgain scoured fibers exposed to the fumigant changed color rapidly (ΔE 11.84 CIELAB units in 2 days) the heated

control exhibited only slightly detectable color change (ΔE 1 32 CIELAB units) in 21 days.

Table 6.3.1 Color Change (ΔE) in CIELAB Units for Wool Fibers Exposed to Dichlorvos Impregnated (S.W.A.T.®) Resin Strips at 50°C in a Desiccator in the Dark.

		Colour difference (CIELAB units)		
Treatment	Time (days)	No heat	Heat[a]	S.W.A.T.®[b]
Shurgain	7	0	2.59	4.43
Scour	21	-	3.68	8.09
	35	-	2.04	6.73
Shurgain/	2	0	-	11.84[c]
Methanol	7	-	-	21.54[c]
Scour	14	-	-	26.79[c]
	21	-	1.32	28.08

[a] ΔE = difference between no heat and heat
[b] ΔE = difference between heat and S.W.A.T.®
[c] ΔE = difference between no heat and S.W.A.T.

Scouring of wool in methanol/Shurgain increased the degree of yellowness of fumigated fibers and decreased the degree of yellowness in the heated control (cf. Fig. 6.3.3 to 6.3.4). Methanol extractable surface contaminants including grease, suint, and other constituents of the proteinaceous contaminant layer (PCL) influence the degree of color change (Anderson,1983; Anderson & Christoe, 1984). Washing the wool fleece with only Shurgain did not remove all of the grease.

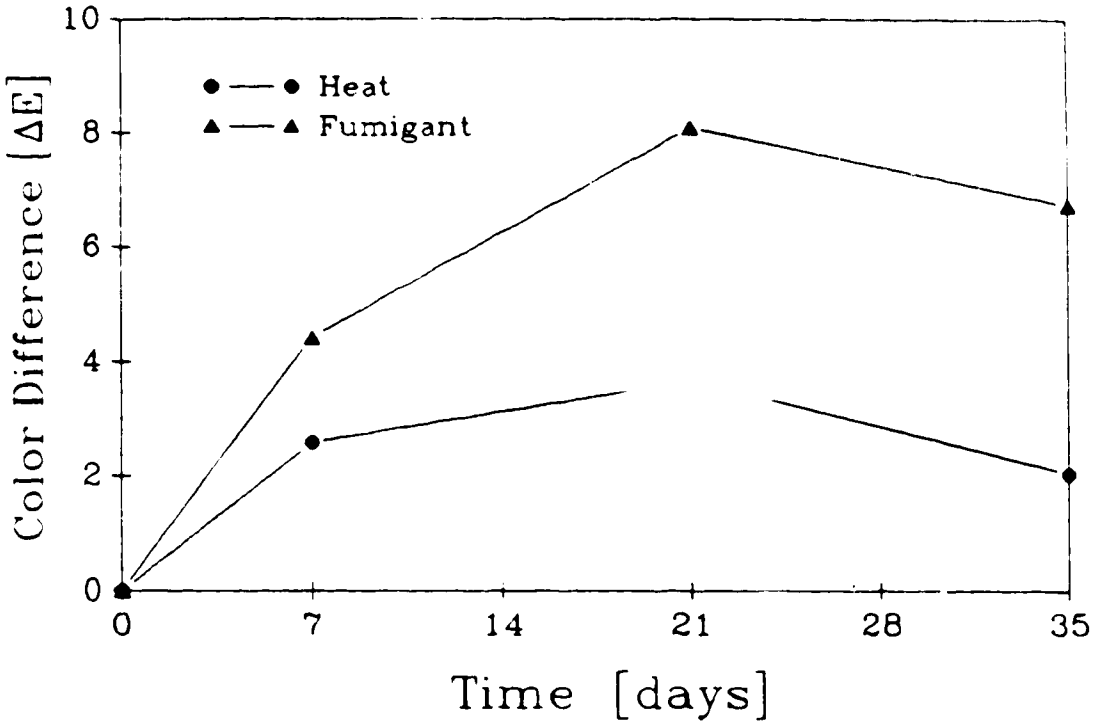


Figure 6.3.1 Total color difference of Shurgain scoured fibers fumigated with S.W.A.T.® at 50°C compared with the heated control.

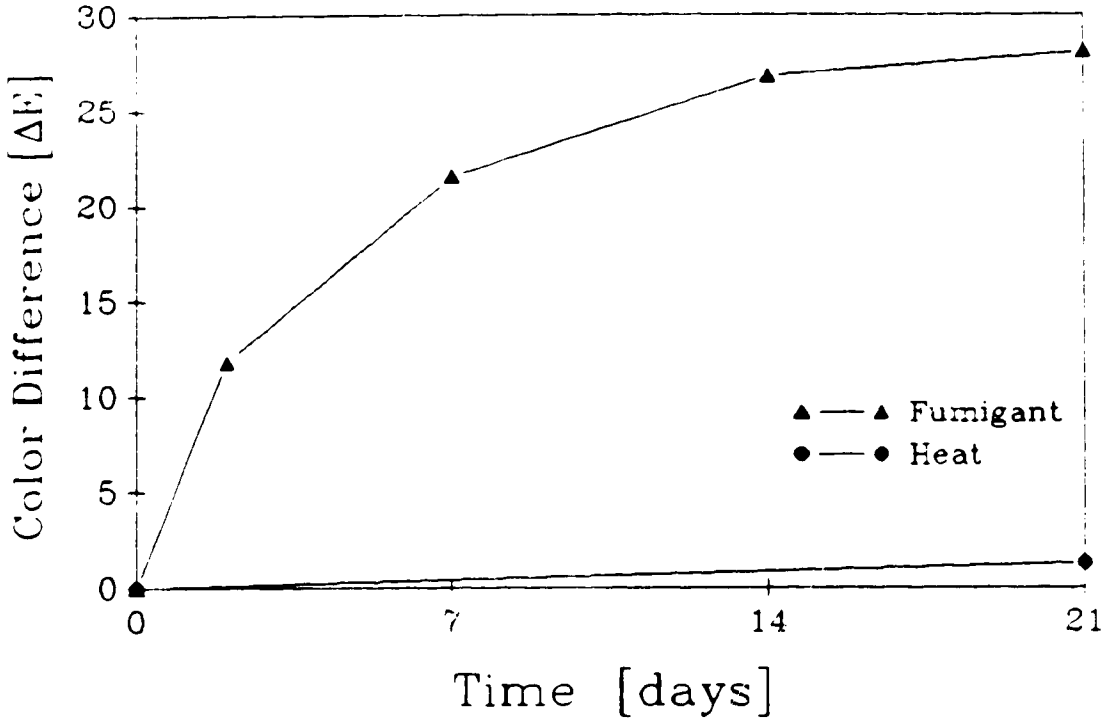


Figure 6.3.2 Total color difference of methanol/Shurgain scoured fibers fumigated with S.W.A.T.® at 50°C compared with fibers scoured in Shurgain.

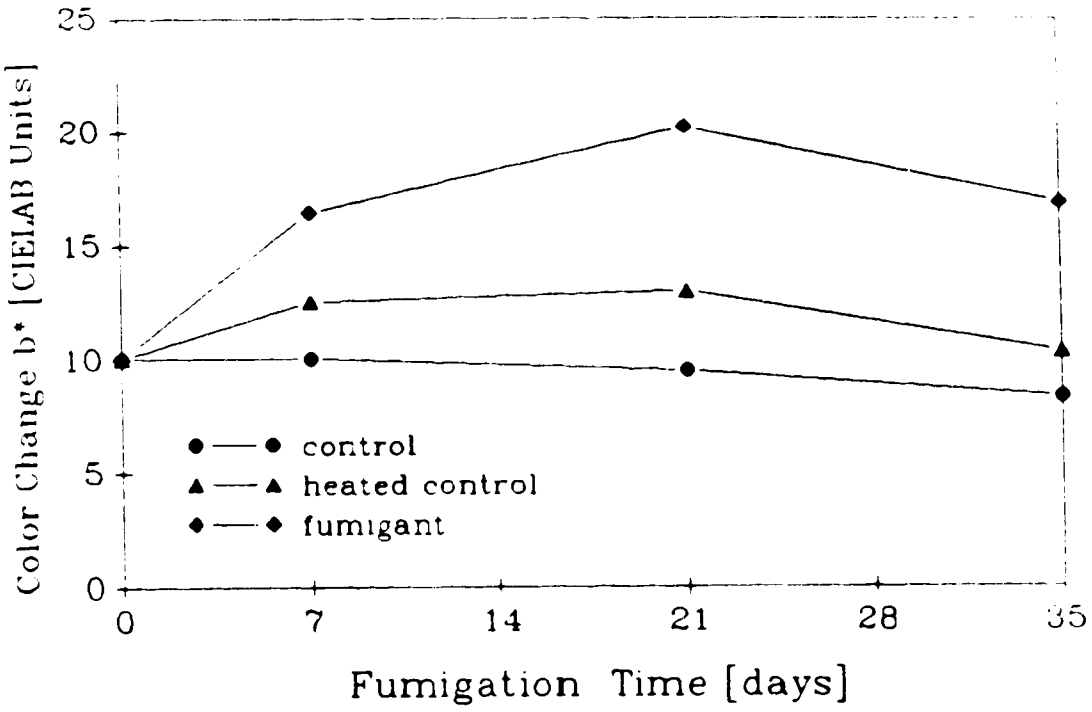


Figure 6.3.3 Changes in b^* (yellow$^+$/blue$^-$) of Shurgain scoured wool fumigated with S.W.A.T.® at 50°C.

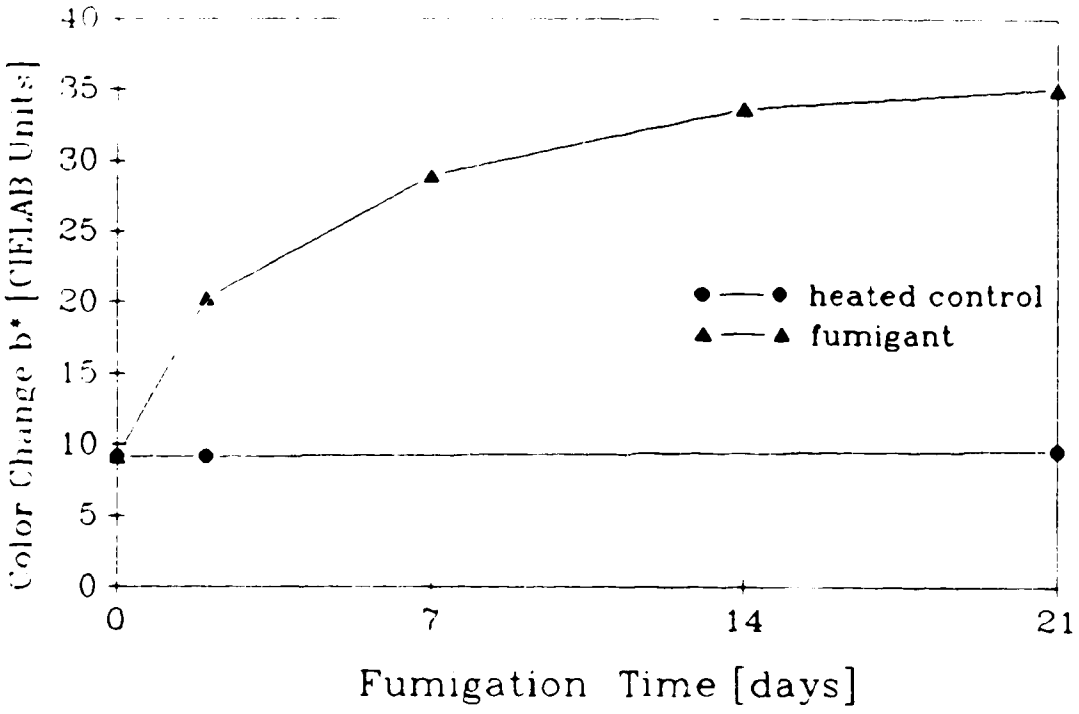


Figure 6.3.4 Changes in b^* (+yellow/-blue) of methanol/Shurgain scoured fibers fumigated with S.W.A.T.® at 50°C.

Oxidation of the greasy surface (Inglis & Lennox, 1965) contaminants probably caused the heated controls to change colour (ΔE 3.68 CIELAB units). The coloring agent in suint responsible for yellowing is methyl 10-(2,5-dihydroxyphenyl)-decanoic acid which is associated with nitrogenous β-ketone (Fraser & Truter, 1960). Post fumgiation washing with Shurgain removed most yellowness from the heated control fibers. However, the yellowness observed in fumigated fibers was not removable by Shurgain or solvents.

Many factors contribute to color changes in wool fibers. Weathering (Veldsman,1966a,b), heat (Launer & Black, 1971), photoyellowing (Lennox *et al.*, 1971; Leaver, 1985; Tucker & Whewell, 1975), chlorine retention (Mansour *et al.*, 1988), and peroxide induced yellowing (Cegarra & Gacen, 1971; 1983). Yellowing reactions occurring in the dark are promoted by moisture and heat (Maclaren & Milligan, 1981).

Temperature is an important parameter in yellowing. The many studies on the effects of heat and moisture during processing indicate that most of the fibers are not *excessively* damaged at temperatures below 50°C (Maclaren & Milligan, 1981). These studies do not include the long term effects of heating at 50°C.

The first chemical changes are detected when wool, heated to 55°C, liberates hydrogen sulfide (Maclaren & Milligan, 1981) Although dry heat causes less damage than than wet heat (Bell *et al.*,

1960) the control samples in this study exposed to dry heat at 50°C exhibited detectable color change (ΔE 3.68 in 21 days). A closed system (in the desiccators) generally mimimizes oxidative damage (Lennox, 1960), however, confined pyrolysis products may promote degradation (McLaren & Milligan, 1981). Chemical damage increases the rate of yellowing or color change (Lennox, 1960; Norton & Nicholls, 1960; Milligan *et al.*, 81-82 CSIRO; Maclaren & Milligan, 1981; Lenin, 1983). Chemically damaged fibers were yellowed more rapidly than undamaged fibers.

Studies of yellowing indicate that the amino acids most affected by light and oxidants (Bradbury & Leeder, 1971) are cystine (Tucker & Whewell, 1975) and tryptophan (Holt *et al.*, 1975; Savige, 1975), and to a lesser extent histidine, methionine, and tyrosine (Inglis & Lennox, 1965; Lennox *et al.*, 1971; Holt *et al.*, 1975). Large differences in the tyrosine content of wool may influence the degree of yellowing (Gillespie & Frenkel, 1975). Merino wool contains up to 12% and Lincoln less than 1% tyrosine (Maclaren & Milligan, 1981). Parallel tests with Merino and Corriedale wool were not conducted in this study. Increased temperature produces yellowing and increases the amount of dityrosine (Röper & Finnimore, 1985; Mattyus, 1975; Savige, 1975).

Although attention has focused on changes in amino acids (Earland *et al.*, 1960) the role of lipids or carbohydrates in yellowing should not be ignored (Inglis & Lennox, 1965). The

observation that heated wool fibers soaked in methanol were not thermally yellowed suggests that several factors contribute to yellowing (Norton & Nicholls, 1960; Lennox, 1960; Earland *et al.*, 1960).

Protection of amino groups by esterification, acylation, and acetylation or other mechanisms markedly reduces yellowing (Lennox *et al.*, 1971). Alcholic esterification may reduce yellowing. Other test results suggest that acylation and esterification of the wool fibers may have occurred (Sec 6.4, 6.8,6.9).

Speakman (1930) found that wool has a pronounced affinity for alcohol. Besides altering the elastic properties, color changes occur in wool which are attributed to acetaldehyde in methanol (Speakman, 1930). The marked increase in yellowing observed in the 7 day methanol/Shurgain fumigated samples may have been enhanced by the interaction of dichlorvos with residual methanol.

Chlorine species are postulated to contribute to yellowing. The energy dispersive x-ray analysis indicates a substantial increase in chlorine during the first few days of exposure to the DDVP resin strip at 50^{0}C (Sec 6.11). The oxidizing chlorine species was not determined.

Tyrosine and tryptophan, known contributers t are modified by chlorine species in an acidic environment Milligan, 1981; Mansour, *et al.*, 1988). Chlorinatio causes more extensive yellowing than at lower pH (Mak

Although the observed decrease in pH in this study may have retarded yellowing, the 50^{0}C temperature and concentration of DDVP by-products may have increased yellowing (Veldsman & Swanepoel, 1971, Lennox, 1960; Makinson, 1979). When wool is treated with chlorine in the presence of peracid or hydrochloric acid, 3-chloro and 3,5,-dichloro tyrosine derivates are formed (Maclaren & Milligan, 1981).

At temperatures above 50^{0}C the amino acids methionine, tryptophan and tryosine are known to be transformed to different oxidized species than at low temperatures (Szilard, 1973). Chlorine dioxide in acid solutions oxidizes tyrosine and produces a brown color (Szilard, 1973). According to some researchers oxidized tyrosine may be a precursor to melanin (Earland *et al.*, 1960). Melanin is a protective pigment and may play a significant role in providing protection to the internal components of the wool fiber.

Cegarra & Gacen (1983) suggest that H_3PO_4 releases H_2O_2. It is possible that peroxides formed during fumigation could change the degree of yellowing. Oxidation of cystine with H_2O_2 in dilute HCl produces small quantities of lanthionine sulfoxide, sulfone, and the major end product cysteic acid (Maclaren & Milligan, 1981). At pH 4-5 maximum modification of cystine, cysteic acid and modification of tyrosine and tryptophan are reported (Maclaren & Milligan, 1981). These modifications may cause yellowing.

6.4. Tensile Properties

The peak load and strain at break (elongation) are found in Table 6.4.1. The strain in the heated yarns (40.6-45.5%) and heated fumigated yarns (41.9-49.3%) were significantly different (α=0.05) from the unheated control (33.7%). At 35 days the elongation of the heated control (40.6% strain) and fumigated yarns (41.9% strain) decreased (Figure 6.4.1).

There was a slight increase in the peak load of yarns fumigated for 7 days (0.541 kg) and 21 days (0.531 kg). The slight increase in load was followed by a decrease in peak load after 35 days (0.507 kg) of exposure to S.W.A.T at 50°C (Figure 6.4.2).

TABLE 6.4.1 Yarn Tensile Strength and Strain at Break in Wool Fumigated with Dichlorvos Resin Strip (S.W.A.T.®) at 50°C.

	Peak Load (kg)		Strain (%)	
Time (days)	Heat	S.W.A.T.®	Heat	S.W.A.T.®
7	0.509	0.541*	44.3*	46.3*
21	0.499	0.531*	45.5**	49.3**
35	0.480	0.507	40.6**	41.9**

Each tabulated value is the mean of 45 samples
* Scheffe test significantly different (α=0.05)
** Subset different from unheated control.
Unheated control: mean peak load = 0.496kg (n=135); S.D.=0.043kg

A standard deviation (0.043kg) for the peak load of unheated

yarns and elongation range of 18-50% (unheated yarns) indicates there is sufficient variability in the wool to exercise caution in interpreting tensile strength and elongation changes. Previous studies indicate that tensile strength (load) is an insensitive measure for assessing degradation (Gregory, 1968; Maclaren & Milligan, 1981). Spivak *et al.*, (1981) found that the breaking strength of fabrics exposed to Vapona® was higher in some cases than unexposed fabrics. The increase in tensile strength in this study is consistent with their findings.

Both the heated controls and fumigated yarns exhibited significant changes (α=0.5) in elongation (44.26% to 46.29%) from the unheated controls (33.7%) in 7 days. The increase in strain after and 21 days of fumigation with S.W.A.T.® and subsequent decrease after 35 days indicate that yarn strain was changed. The fumigant has a greater influence on yarn elongation than heat, and perhaps catalyzes the effects of heat (Figure 6.4.1).

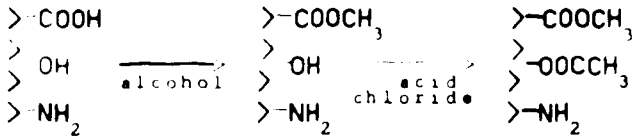
Yarns are stretched during spinning and processing (Schutz & Blin, 1971; Ross *et al.*, 1986). Therefore, differences in elongation between heated specimens and unheated specimens may be partially attributed to stress relaxation during heating and conditioning. Although both the unheated and heated control yarns were conditioned before measuring tensile properties only the heated and fumigated yarns were conditioned twice.

Shaw (1985) found that reversible dimensional changes occur

with variations in temperature (Schutz & Blin, 1971; Szucht, 1971; Feughelman & Robinson, 1967). Yarns are heated and stretched during processing (Von Bergen, 1970). Hysteresis may affect subsequent treatments because some energy is lost during the cycle of deformation and recovery (Watt, 1975; Makinson, 1979; deGraaf, 1980; Tao & Postle, 1985). Studies with wool exposed to acids indicate that contraction is followed by elongation exceeding the original length (Crewther & Dowling, 1959). Perhaps the increase in acidity caused by sorption of DDVP by-products contributed to elongation. Heating wool at temperatures similiar to this study produces crosslinking between lysine and glutamic or aspartic acid (Asquith & Otterburn, 1971; Lipson, 1975). Heating decreases strength and elongation (Maclaren & Milligan, 1981) and decreases acid and basic groups of wool (Menefee & Yee, 1965).

According to Speakman (1985) esterification of aspartic acid and glutamic acid side-chains or alkylation of basic amino acid residues breaks salt links which makes fiber extention easier (Speakman, 1985). Esterification may have occurred during fumigation. Wolfram and Milligan (1975) found selective esterification (at 60^{0}C) of aspartic and glutamic acid to the corresponding methyl esters and "a little conversion of amide to ester group" drastically lowers the stress required to extend wool and increases contraction in boiling water (Wolfram & Milligan, 1975). Esterification and acylation destabilize the fiber by

disrupting electrostatic forces between acidic and basic groups a: ! disrupting hydrogen bonds between hydroxyl and carboxyl side chains (Wolfram & Milligan, 1975).



Weigmann & Dansizer (1971) suggest that both temporary and permanent deformities in tertiary structure of keratins are dominated by disulfide bonds. Under stress the bonds easily change positions. Removal of SH-groups increases extensibility (See 6.5). Replacement of disulfide bonds with more stable cross-links decreases extensibility (Weighmann & Dansizer, 1971). Both heat and chemicals affect disulfide bonds (Sakamoto *et al.*, 1985; Meichelbeck & Spei, 1975). The lead acetate test for -SSH bonds indicates that SH-groups were removed by oxidants (See 6.5).

Speakman (1960) suggests that the effects of intermediate oxidation products of cystine can be removed with 0.05M hypophosphoric acid at 50^{0}C for a few hours (Haly & Feughelman, 1960). Removing the cystine oxidation products establishes new crosslinks (Feughelman, 1960). Although the phosphorus species formed in the degradation of dichlorvos are unknown, dimethyl phosphoric acid is one of the principal by-products of dichlorvos degradation and may produce phosphorus species capable of contributing to tensile changes.

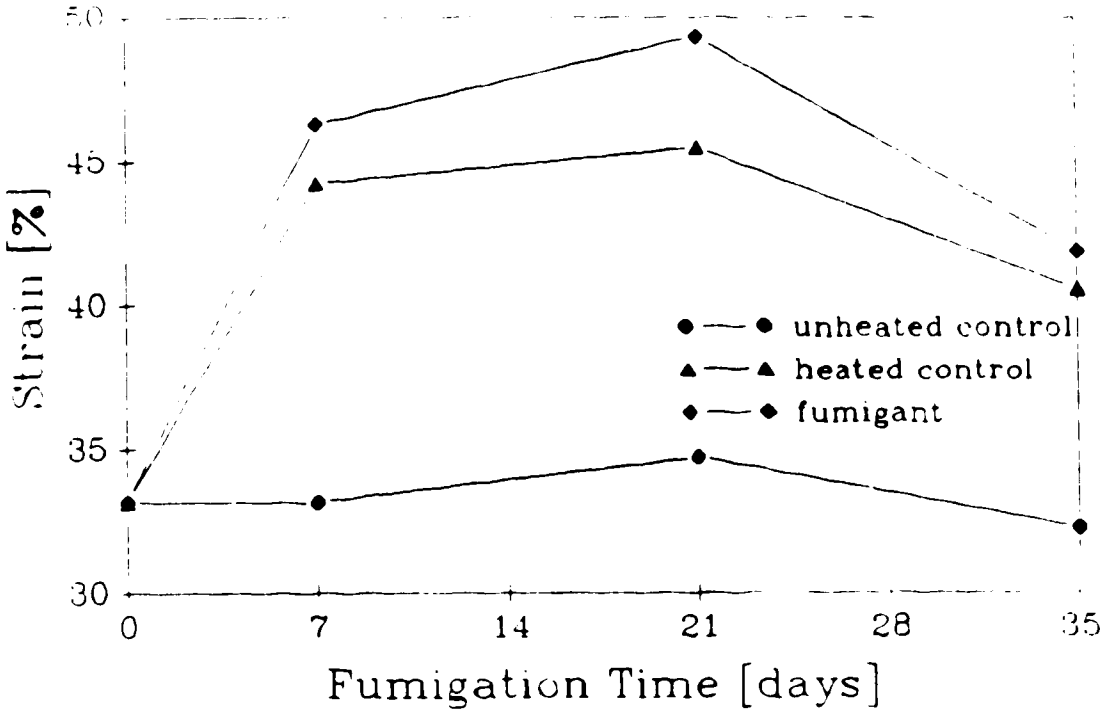


Figure 6.4.1 Strain(%) of Merino 2/22 wool yarns fumigated with S W.A.T.® at 50°C compared with heated and unheated wool yarns.

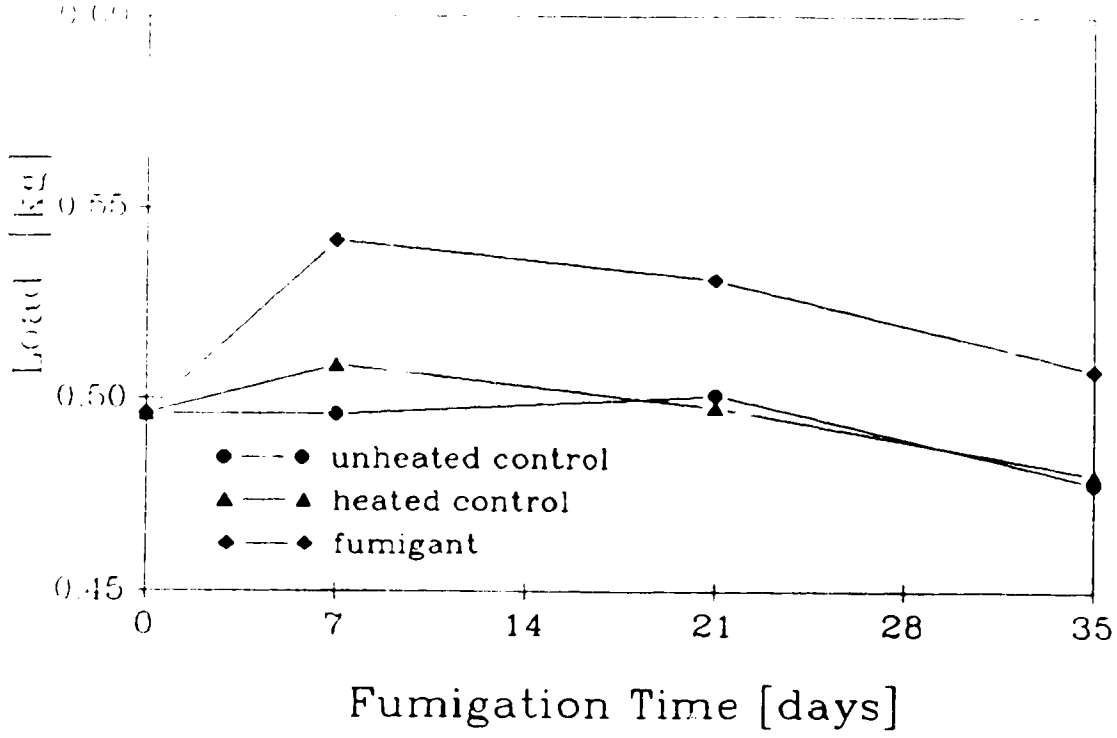


Figure 6.4.2 Load (kg) of Merino 2/22 yarns fumgiated with S.W.A.T.® at 50°C compared with heated and unheated yarns.

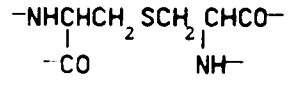
The high tyrosine component of wool forms part of the cortical matrix and CMC (Crewther, 1975). The matrix is known to govern mechanical properties (Spei, 1985). The yellowing of fibers exposed to DDVP implies tyrosine changes. Tyrosine forms self-crosslinks (di- and trityrosine) at the 3,5 position, and ether links by hydrolysis of alcoholic substituents. Ester crosslinks may be formed by condensation of tyrosine residues with aspartic and glutamic acid residues (Maclaren & Milligan, 1981). Tyrosine also couples with histidine and lysine. Changes in the mechanical properties of wool may reflect changes in tyrosine.

Wool modified by crosslinking agents such as chlorine and aldehydes shows changes in elongation (Koenig & Friedman 1977; Watt, 1971; Speakman, 1985). Evidence for crosslinking is indirect and often ambiguous because changes in properties may resemble changes introduced by monofunctional reagents that do not produce crosslinks. Koenig & Friedman (1977) found a large difference in extensibility in wool treated with monoacid chloride and diacid chloride crosslinking agents. Both treatments reduced the stress at break. Although the extension of monacid chloride treated fibers was approximately twice as great as di-a chloride both reagents reduced extension at break (Koenig & Friedman, 1977). It is unlikely that substantial crosslinking occurred during the earlier fumigation periods because elongation at 7 and 21 days increased substantially (Figure 6.4.1). However, the increase was followed by a decrease in elongation after

35 days of exposure. A similar decrease (45-40% strain) was observed in heated yarns at 35 days. The literature indicates that heat alone will form cross-links (Maclaren & Milligan, 1981). Crosslinking is usually accompanied by an increase in the breaking load. In this study longer heating periods reduced the breaking load.

New amino acid residues are known to form during heating at temperatures less than 70°C. Heated wool absorbs oxygen causing a variety of reactions. The formation of lanthionine, lysinoalanine crosslinks and cysteic acid residues accompanies the destruction of cystine residues. Lanthionine (16.5 $\mu mol.g^{-1}$) and a trace of lysinoalanine residues are formed at 60°C (Maclaren & Milligan, 1981). Crosslinks between the carboxyl groups of glutamic and aspartic acids and lysine form the isopeptides ϵ-(γ-glutamyl)lysine (Glu $\overbrace{\text{Lys}}$), and ϵ-(β-aspartyl) lysine in wool heated (60°C) for 48 hours (Maclaren & Milligan, 1981). Formation of crosslinks would reduce elongation at break. During the first 24 hours of heating (42-80°C) the isopeptide Glu $\overbrace{\text{Lys}}$ is stable. However, prolonged heating (96 h) at 80°C results in significant formation of Glu $\overbrace{\text{Lys}}$, and decrease in hydrolysis.

lanthionine



6.5. Lead Acetate Test

Lead acetate staining depends on the presence of hydrodisulfide (-SSH). Changes in the state of sulfur can be measured by observing a reduction in the formation of lead sulfide precipitate. Hydrodisulfide groups reacting with lead acetate form lead sulfide, a brownish black precipitate, on untreated wool. Other oxidation products of cystyl residues on the surface of wool fibers are unable to form complexes with lead acetate. Lead acetate color differences of heated and fumigated fibers are found in Table 6.5.1. Although the ΔE values indicate a color change, the actual fiber samples were unevenly stained, with underlying yellowing in the fumigated fibers that distorted ΔE results. Heated wool stained less than unheated controls and fumigated wool stained less than heated wool.

Heat caused some oxidation of the sensitive cystine residues. Heat plus DDVP oxidized the -SSH groups to an even greater extent. The greatest change in reactivity with lead acetate (ΔE = 17.93) was observed in the 7 day methanol/Shurgain scoured fibers which showed much less staining than the heated control.

Lead acetate test results show that limited oxidation of hydrodisulfide groups occurred when Shurgain scoured wool was exposed to dichlorvos. The removal of methanol soluble components of the protein contaminant layer (PCL) and greasy lanolin or tightly bound lanolin wax with methanol appeared to increase the rate of oxidation

resulting in less staining.

Table 6.5.1 Lead Acetate Color Difference (ΔE) in CIELAB Units for Wool Fibers Exposed to Dichlorvos (S.W.A.T.®) Resin Strips at 50°C in a Desiccator in the Dark.

		Color difference (CIELAB units)	
Treatment	Time (days)	Heat[a]	S.W.A.T.[b]
Shurgain Scour	7	8.98[c]	7.47[d]
	21		3.98[d]
	35		6.58[d]
Shurgain/ Methanol Scour	7	3.85[c]	17.93[d]

[a] ΔE - difference between unheat and heated fibers
[b] ΔE - difference between heated and fumigated fibers
[c] ΔE - less staining than unheated contols
[d] ΔE - less staining than heated controls

Uneven staining of the controls and 7 day Shurgain exposures is postulated to be a result of ageing, weathering, and uneven attack which result in different oxidation states of sulfur (Bell *et al.*, 1960). Heat alone oxidizes wool with a high cystine content (Norton & Nicholls, 1960). Oxidation of cystine by heating probably contributed to a decrease in lead sulfide precipitates (Table 6.5.1). Chlorine species, present in the fumigant, are also known to contribute to the observed uneven staining (Maclaren & Milligan,

1981). Fibers exposed to the fumigant stained less than the control fibers. This is indicative of increased hydrodisulfide oxidation.

Studies of oxidative chemicals used in wool processing indicate that several intermediate cystine species are formed (Baumann & Setiawan, 1985a; Ashrenazi, 1984). Although acidic solutions of hydrogen peroxide, performic acid, and sodium hypochlorite produce the cystine oxidation intermediates, cystine monoxide, cysteine sulfinic acid ($-SO_2H$), sulfonic acid ($-SO_3H$) and cysteic acid residues, the sensitive hydrodisulfide (-SSH) group is not detected (Baumann & Setiawan 1985). Wool oxidized with these agents will not stain darkly in the lead acetate test. Some oxidants were likely present during fumigation. Although mild conditions oxidize cysteine sulfinic acid to the more stable cysteic acid, over oxidation can convert cysteic acid to sulfate (Kearns *et al.*, 1977). Uneven topochemical attack, which occurs in acid chlorination, is confirmed by different ESCA (Electron spin capture analysis) sulfur spectra on different areas of the fiber (Baumann *et al.*, 1986; Baumann & Setiawan, 1985). Uneven staining of the fibers exposed to S.W.A.T.® is congruent with acid chlorination (See 6.1, and 6.10).

Although the principal target of oxidizing agents is cystine several other amino acids are modified or destroyed (Schumacher-Hamedat *et al.*; 1985; Baumann & Setiawan; 1985). Since lead acetate test results indicate that cystine was oxidized during fumigation other amino acids may be attacked. Methionine may be

converted to sulfoxide or sulfone and tryptophan and tyrosine modified or destroyed (Mazingue *et al.*, 1971 p. 209; Maclaren & Milligan, 1981). Some oxidants induce free radical formation causing main chain cleavage (Davies, 1961).

Known breakdown by-products of dichlorvos resin strips include oxidants (Williams *et al.*, 1984). Peracids, such as perchloric, peracetic, and dichloroacetic, peroxide, and chlorine are suspected by-products. Peracetic acid destroys some of the tryptophan, arginine, lysine and histidine residues (Lindley, 1959; Maclaren & Milligan, 1981). The effects of peracids are complex because radical intermediates are formed. It is possible that synergistic reactions of peracids with other known oxidants such as peroxides and chlorine occurred during fumigation.

Although hydrogen peroxide damage of wool occurs mostly in alkaline conditions, cystine oxidizes under acidic conditions. Maclaren and Milligan (1981) suggest that oxidation of cystine by peroxides is controlled by the rate of diffusion and sorption (See 6.11). Methanol scouring is suggested to increase the rate of sorption in wool which resulted in increased oxidation of hydrodisulfide groups that caused reduced staining with lead acetate.

.6 Allwörden/Herbig Reaction

The Herbig reaction with bromine was found to be the most

reliable test for measuring the integrity of the epicuticle. A freshly prepared bromine solution must be used to obtain consistent results. A summary of observations is found in Table 6.6.1.

The literature indicates that Allwörden sac development is a sensitive measure of changes in the cuticle. Al-Hariri & Rattee (1983) found that sac development in processed samples was 15 seconds for scoured, 22-29 seconds for scoured and carded, and 32-39 seconds for scoured carded and spun wool fibers. By using unprocessed wool with tippy ends trimmed off, it was possible in this study to

Table 6.6.1 Herbig Reaction for Epicuticle Integrity in Wool Fibers Exposed to S.W.A.T.® Resin Strips at 50°C in a Glass Desiccator in the Dark.

Treatment	Time (days)	Observation of Sac Formation	
Heat	7	normal formation of sacs[a]	<≅ 3 min
	21	normal formation of sacs	
	35	normal formation of sacs	
(CH_3OH scour)	7	normal formation of sacs	
S.W.A.T.®	7	sac formation appears normal	
	21	areas with no sac formation[b]	
	35	areas with no sac formation,	
(CH_3OH scour)	7	areas with no sac formation, epicuticle damage and etching of surface observed[c]	

[a] formation of discrete and apparent epicuticle sac

minimize effects of weathering and avoid the effects of manufacturing. The formation times given in the literature for the characteristic globular excrescences (bubble-like sacs) on the fiber surface, are too short to distinguish between wetting or interface activity and discrete sacs. Makinson (1979) reports that Herbig sacs, formed with bromine, are more variable and slower to form than Allworden sacs, formed with chlorine, because Herbig sacs consist of the epicuticle with various amount of material from other cuticle layers (Makinson, 1979). In this study separation of the epicuticle from the underlying cuticular protein layers with the formation of obvious Herbig sacs took ≈ 3 min for normal sac formation. Fumigated fibers tended to produce Herbig sacs more rapidly than unfumigated fibers.

Bradbury & Leeder (1971) found several conditions governing the formation of Allwörden (or Herbig) sacs (Bradbury & Leeder, 1971). Sacs may fail to form when insufficient protein dissolves to separate the epicuticle from the cuticle, when osmotic pressure from oxidation of disulphide bonds to cysteic acid residues is insufficient, or when the epicuticle membrane is not intact (Bradbury & Leeder, 1971; Lorimer, 1979). Makinson (1979) suggests that delay in sac formation may be due to previously degraded protein of low molecular weight diffusing out of the cell too quickly for swelling to occur (Makinson, 1979).

The hydrophobic epicuticle is part of the membrane system on

the surface of the fiber (King & Bradbury, 1968). Although the lysine- rich cuticle and cortex membrane complex form a continuous network throughout the fiber, amino acid analyses reveal that cuticle resistant membranes differ from the cortical CMC resistant membranes. Resistant membrances in the cuticle contain citrulline and ornithine, have a lower lysine and higher serine content, and have more cystine/cysteic acid than the cortical cell resistant membranes (Peters & Bradbury, 1976). The extreme chemical inertness and hydrophobicity of the epicuticle are attributed to ϵ-(γ-glutamyl) lysine crosslinks and the presence of small amounts of lipids (Röper *et al.*, 1982; Peters & Bradbury, 1976). Lysine-glutamic acid crosslinks contribute to ine. ness by removing the hydrophilic carboxyl and amino groups (Peters & Bradbury, 1976, p. 47). The unique characteristics of the epicuticle and wool fiber are strongly influenced by cysteic acid/cystine residues.

SEM micrographs (6.12) show cuticle damage and EDXA (6.11) indicates large amounts of chlorine in fumigated fibers. Inhibition of sac formation may be attributed to the formation of $-CH_2Cl$ and $-CH_2SO_2Cl$ groups from $-CH_2S-S-CH_2-$(disulfide bond) (Bradbury & Leeder, 1972). Less solubilizing at low pH may be due to lack of sulphonic a id anions in the *A* layer. If disulfide bonds are blocked or stabilized by conversion to lanthionine, preventing sulfonic acid groups from forming, Herbig sacs are smaller (Makinson, 1979). Cysteic acid, 3,5-dichlorotyrosine and α-aminoadipic acid from lysine

oxidation are present in acid hyrolysates of Allwörden membranes from chlorinated wool (Maclaren & Milligan, 1981). Exposure to S.W.A.T.® produced smaller Herbig sacs in the 7 day methanol fibers than in Shurgain fumigated fibers. Apparently the consistency of the dissolved proteins and amount of oxidizable material available for sac formation was decreased during fumigation.

6.7 Krais-Viertel Test

The Krais-Viertel test supposedly differentiates acid damaged wool from reduced or alkali damaged wool (Merkel, 1984; Garner, 1966). Garner (1966) reports that bubbles form on the surface of undamaged fibers in about 10 minutes. Merkel (1984) suggests that the reagent is a good indicator of acid damage. Acid damaged wool is reported to form larger bubbles in less time than undamaged wool. Cuticle removal by chlorination is reported to cause generallized swelling of the fiber.

Although the Krais-Viertel test is reported to be similiar to the Allworden reaction, it was found to differ considerably. The entire fiber is observed undergoing dissolution in the highly alkaline medium. Discrete bubbles observed in the Allwörden or Herbig reaction are not formed with Krais-Viertel reagent. Bulges are formed that give the fiber the appearance of a swollen segmented worm. In contrast to fibers placed in water, which usually become straighter and less crimped (Makinson, 1979), fibers immersed in Krais-Viertel

reagent twist, wriggle and curl into coils. By comparing unheated and heated controls with treated fibers it was apparent that the strongly alkaline Krais-Viertel reagent reacted more rapidly with the fumigated fibers. Fumigated fibers scoured with Shurgain detergent were less reactive than fibers exposed in the laboratory to 1M H_3PO_4 for 24 hours at room temperature. Methanol/Shurgain fumigated fibers reacted more rapidly than fibers soaked in 1M H_3PO_4 for 24 hours at room temperature.

The differential reactivity of the ortho and para cortex was easily observed with Krais-Viertel reagent (Kulkarni *et al.*, 1971). The orthocortex swelled more rapidly than the paracortex causing the fibers to coil.

Although the literature indicates that the reagent is stable for 4-6 weeks the difference in reactivity between 1 or 2 days was noticeable. The noxious, choking, corrosive reagent (KOH + NH_4OH) is difficult to use without adequate ventilation. The extreme variablity in time (minutes to hours) to produce the bulging sacs curtailed attempts to accurately record the time for sac formation.

Many interpretations for the swelling properties of fibers are found in the literature. Speakman and Hirst (1933) refer to lamellar micelles with the long axes parallel to the length of the fiber. Observing that micelle subdivision occurred in formic acid, the researchers concluded that subdivision to simple peptide chains was improbable. By using several acids at different concentrations

Speakman and Hirst (1933) concluded that acids were capable of separating micelles of wool. In this classic study Speakman & Hirst (1933) suggest that all acids cause a reduction in the resistance to swelling. The acidic methanol scoured fumigated fibers were the most reactive in the Krais-Viertal reagent. Some of the variability in reaction time may be due to crosslinking or an undetermined competing factor such as increased oxidation coupled with increased acidity that complicates chemical reactivity.

The Krais-Viertel is essentially an alkaline solubility test Lees *et al.*, 1960). In this study the increased speed of fiber dissolution in the Krais-Viertel test correlated with the decrease in lead acetate staining in fumigated fibers. Maclaren and Milligan (1981) suggest that chlorination of wool increases alkali solubility. High chlorine sorption (See 6.11) may have contributed to an increase in reaction rate of the fumigated fibers with the Krais-Viertal reagent. Knott and Zahn (1971) found that an acid content of greater than 1% (< pH 4) caused greater alkali solubility and attributed to N—>O peptidyl shift during storage.

6.8 Methylene Blue Staining

Staining of wool with Methylene Blue is used to detect oxidative damage (Schefer, 1978). Extensive oxidation under acidic conditions produces cysteic acid residues and sulfate, and modifies many amino acids. Anionic sites provide negative charges which

attract the cationic Methylene Blue dye. According to Maclaren and Milligan (1981) excessive oxidation of cystine residues to cysteic acid residues increases the uptake of Methylene Blue

The results of color difference measurements (ΔE) on Methylene Blue stained wool are recorded in Table 6.8.1. Fairly rapid dye uptake in fibers heated for 7 days (ΔE 6.79 CIELAB units) indicates oxidative heat damage. However, a decrease in staining occured in the 35 day heated control (ΔE 3.41 CIELAB units). According to Menefee and Yee (1965) oxidative damage at temperatures greater than 60°C causes acid and base groups to decrease.

Table 6.8.1 Methylene Blue Color Change (ΔE) in CIELAB Units for Wool Fibers Exposed to (S.W.A.T.®) Resin Strips at 50°C in a Desiccator in the Dark.

		ΔE Color difference (CIELAB units)	
Treatment	Time (days)	Heat[a]	S.W.A.T.®[b]
Shurgain Scour	7	6.79	2.37[c]
	21	6.93	14.03[c]
	35	3.41	15.05[c]
Shurgain/ Methanol Scour	7	15.08	29.80[c]

[a]ΔE = difference between no heat and heat
[b]ΔE = difference between heat and S.W.A.T.®
[c]ΔE = less dye pick-up than control

Heated controls, scoured with methanol/Shurgain, stained a deeper blue (ΔE 15.08 CIELAB units) after a 7 day heating period than the 35 day heated controls scoured only in Shurgain (ΔE 3.4 CIELAB units). Wool grease affects the uniformity of dyeing (Bocykowski, 1971). Merino wool, a much greasier wool than Corriedale (Lipson, 1975) was used for the 35 day and Shurgain/methanol samples. The observed difference in reactivity infers that lanolin contaminants provide protection. Methanol removed some of the grease layer from the fibers and increased the rate of chemical activity by directly exposing the fiber to the fumigant.

The color differences (ΔE 2.37 to 29.80 CIELAB units) of all fumigated samples represent a *decrease* in staining compared to the heated controls. The difference in dyeability of fibers washed in methanol solvent is evident in the 7 day fumigation periods. The color difference in fibers washed in Shurgain and fumigated in S.W.A.T.® for 7 days (ΔE 2.37 CIELAB units) is much less than the color difference (ΔE 29.80 CIELAB units) for Shurgain/methanol scoured fibers fumigated for the same length of time. Methanol/Shurgain scoured fibers fumigated for 7 days (ΔE 29.80 CIELAB units) resisted staining with Methylene Blue more than other exposures. Either the fumigated fiber repels the dye, or the sites normally occupied are blocked chemically or physically (Whewell *et al.*, 1971; Ebert, 1971). The resistance to dye absorption suggests, lack of oxidation, decreased pH, or chemical blocking of anionic

sites during fumigation. Since other tests suggest some oxidation, a low pH, or chemical blocking hypotheses are the most credible explanations.

Although Merkel (1984) suggests that Methylene Blue can be used to detect severe acid damage other researchers disagree. Methylene blue absorption is very pH dependent (Whewell, *et al.*, 1971). The nature and number of polar groups in the fiber, the physical and chemical modification, and accessibility of the dye-attracting acidic sites influence Methylene Blue absorption (Whewell, *et al.*, 1971; Peters 1975). Wool boiled in neutral solutions of the oxidant dichromate showed a decrease in Methylene Blue dye absorption (Whewell *et al.*, 1971). Apparently negative sites are removed by complexing with oxidants such as dichromate.

Dissociation of acids, such as acetic acid, produces free H^+ ions which compete with dye cations for the electronegative sites and retard dyeing (Ayyangar & Tilak, 1971). Below pH 6 the absorption of dye decreases and is minimal at pH 4.5 (Ayyangar & Tilak, 1971). Evidently the acidic by-products of S.W.A.T.® provide free hydrogen ions that retard dyeing. The more acidic fumigated fibers absorbed less dye. At least some of the decrease in dye absorption is attributed to a decrease in pH.

The rate of Methylene Blue diffusion into wool fibers is recognized as a reliable estimate of the epicuticle's condition, especially after chlorination (Whewell *et al.*, 1971; Merkel, 1984).

Since the cuticle provides an effective barrier to dye diffusion, the dye molecules are postulated to enter the fiber though the interstices of cuticle cells (Bradbury & Leeder, 1971; Leeder, 1986). Bradbury and Leeder (1971) suggest that the CMC, rather than the cuticle, is the barrier to dyes and acids. Areas of oxidative damage on fibres are usually stained deep blue (Merkel, 84). Under the light microscope the margins between intact cuticle cells appeared to have stained more deeply than adjacent surfaces. Wool fibers with a mechanically disrupted surface structure absorb more dye supporting the assumption of intercellular diffusion of dye between cuticle cells (Kondo, *et al.*, 1971; Leeder *et al.* 1985; Joko *et al.*, 1985).

Although mechanical and chemical attack usually increases the rate of dye uptake (Leeder & Lipson, 1963) the fibers most damaged by fumigation (methanol/Shurgain exposures) resisted both acid and basic dyes. Although progressive reduction of pH in the dyebath and fiber usually decreases absorption of basic dyes, some acids introduce a number of sulfonic groups which strongly attract basic dyes (Ebert, 1971; Baumann, 1971; Whewell *et al.*, 1971).

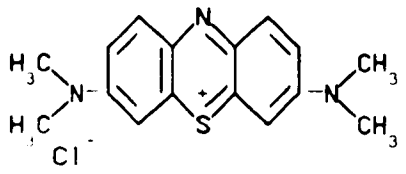
Maclaren and Milligan (1981) explain that despite the presence of sulfonic acid groups, the ion-exchange properties of oxidized wool are more similiar to cationic-exchange resins containing carboxylic acid side-chains than to sulfonic acid side-chains. Apparently the anionic sulfonic acid groups are compensated by basic groups within the fiber leaving the weakly

anionic carboxylic acid groups available for exchange with the cationic dye (Whewell, *et al.*, 1971).

While decreased dye uptake in fibers exposed to the fumigant for longer periods can be explained by pH sensitivity of Methylene Blue, a decrease in the dye uptake of heated wool (ΔE 6.79 to 3.41 CIELAB units) is unexplained. The decreased lead acetate reactivity and slight yellowing in the heated controls suggest oxidation that according to Merkel (1984) should increase sorption of Methylene Blue (Savige, 1975). The increase in dye absorption in the heated controls, especially the 7 day control also suggests oxidation. To complicate matters chromatic changes in the dye were observed in heated (methanol/Shurgain) and fumigated wool fibers after staining. The decreased absorption with increased oxidation and chromatic changes requires an explanation. Aqua tones were observed in the 21, 35, and 7 day methanol/Shurgain stained fumigated fibers and mauve tones in the 35 day and 7 day methanol/Shurgain heated controls.

Ayyangar and Tilak (1971) found that some bases produce a spectral shift towards red, attributed to interaction of Methylene Blue with basic ions which reduces the positive charge shared by the terminal positions. Although the conditions in this study are not basic it is apparent that a reaction exists that produces a mauve tone in the stained fiber.

Methylene Blue is a widely used in biochemistry as a hydrogen acceptor and vital stain because of its metachromatism (Gurr *et al.*).



Methylene Blue

In this study a spectral change to aqua was accompanied by reduced staining.

In all tests the depth of staining in fumigated fibers was less than fibers exposed to the oxidants, sodium hypochlorite, or H_2O_2. Wool soaked in 1*M* perchloroacetic acid, 1*M* dichloroacetic, and 1*M* trichloroacetic acid for 24 h at room temperature stain similiarly (aqua color and decreased staining) to the fumigated fibers. It appears that decreased pH prevents the cationic dye from coloring the fumigated fibers.

Methlyene Blue is used as a diagnostic technique to determine chemical composition, reactivity, histochemical components, and mechanical damage (Whewell,*et al.*, 1971; Kondo *et al.*, 1971; Orwin *et al.*, 1984). Methlylene Blue is used to determine changes in the high-tyrosine orthocortex, which is usually stained with Methylene Blue (Hewish *et al* , 1984; Hewish & French, 1986; Fraser *et al.*, 1972). When fumigated fibers were stained with the concentrated stock solution of Methylene Blue and examined under the light microscope the orthocortex appeared to resist staining, suggesting

some modification of tyrosine. The observed yellowing suggests that tyrosine residues are modified during fumigation.

An increase in oxidation of cystine residues to cysteic acid usually results in an increased uptake of Methylene Blue (Maclaren & Milligan, 1981; Whewell *et al*., 1971). Since a substancial increase in chlorine, a strong oxidant, was detected in EDX analyses (Sec 6.10) the reduction in staining suggests that either the normally available sites are occupied or the charge on the fiber is radically altered (Whewell *et al*, 1971). However, caution in postulating theories must be exercised because wool and dye chemistry are complex

Several theoretical models have been developed to explain the adsorption of basic (cationic) dyes from an acidic bath and in the pr sence of electrolytes (Peters, 1975). In a weakly acidic dyebath amino groups are protonated and bear a positive charge ($-^{+}NH_3$) and carboxyl groups a negative charge ($-COO^{-}$). In acidic dyebaths the carboxyl groups are protonated. The breakdown of dichlorvos produces chlorine and phosphorus ions which may react with the amino or carboxyl groups. Evidently chlorine changes both the pH and the absorption of acid and basic dyes. If positively charged phosphorus ions from dichlorvos hydrolysis were present in the dye bath, their influence would be negligable due to the acidity of the fumgiated fibers. However, phosphorus species may have reacted with the wool fibers in a more complicated manner.

Whewell *et al.* (1971) suggest that a decrease in Methylene Blue absorption may be caused by replacement of carboxyl groups by carboxyl complexes which repress absorption of dye cations. Dichlorvos is known to react with several amino acid groups such as cysteine, serine, and perhaps tyrosine (See 2.2).

6.9 Kiton Red Staining

The color differences (ΔE) of fibers stained with Kiton Red (C.I. Acid Red 1) are recorded in Table 6.9.1. An increase in staining is usually attributed to epicuticle damage, oxidation, or extreme acid damage (Merkel, 1984; Garner, 1 a). Acid damaged fibers have a tendency to stain lighter than undamaged fibers.

An increase in the dyeing capacity of both 7 day fumigated samples were observed. The Shurgain scoured wool had a color difference of 7.44 CIELAB units and 7 day methanol/Shurgain scoured wool had a color difference of 17.33 CIELAB units. The a^* index (+ red/- green) of the 7 day samples was similiar (7 day, a^* 27.7; 7 day MeOH, a^* 26.1). The difference in the values of ΔE in the methanol/Shurgain scoured fibers is due to increased yellowing in the 7 day methanol/Shurgain fumigated fibers and a decrease in dye uptake by the heated control Total color difference (ΔE) is insensitive to chromatic variability (Warns, 1985). Reduced staining was observed in the Shurga red samples fumigated for 21 and 35 days. Reduced staining sistent with the observed decrease in

pH of the 21 and 35 day fumigated specimens. Acid damaged wool is known to absorb less Kiton Red than undamaged wool. The slight orange tinge accompanying decreased staining is attributed to fiber yellowing. An increase in yellowing is evident in the b^* CIELAB tristimulus readings.

Table 6.9.1 Kiton Red (CIE Acid Red 1) Color Difference (ΔE) in CIELAB Units for Wool Fibers Exposed to (S.W.A.T®) Resin Strips at 50°C in a Desiccator in the Dark.

		ΔE Colour difference (CIELAB units)	
Treatment	Time (days)	Heat[a]	S.W.A.T.®[b]
Shurgain Scour	7	10.97	7.44[c]
	21	12.30	13.74[d]
	35	6.45	22.06[d]
Shurgain/ Methanol Scour	7	15.32[f]	17.03[e,c]

[a]ΔE - difference between no heat and heat
[b]ΔE - difference between heat and S.W.A.T
[c]ΔE - more dye pick-up than heated control
[d]ΔE - less dye pick-up than heated control
[e]ΔE - most of the difference attributed to yellowing
[f]ΔE - less dye pick-up than unheated control

The affinity of wool for acid dyes usually increases with acid chlorination (Weigmann *et al.*, 1985). An increase in acid dye affinity is proposed to be caused by "preferential loss of proteins

containing acidic amino acid residues from the cuticle" (Maclaren & Milligan, 1981, p 68). This is accompanied by an increase in the permeability of the epicuticle by chlorination and easier dye access (Maclaren & Milligan, 1981).

The pH of the dyebath has a marked effect on the uptake of acid dyes (Bird, 1972). Decline in degree of fixation in acidic environments is due to a decrease in the number and reactivity of nucleophilic groups (Lewis bases) at low pH (Baumann, 1971). Bird (1972), using the Gilbert-Rideal theory, attributes strong acid absorption (HCl) to an *equipotential region* where H^+ is bonded to the COO^- sites and Cl on the N^+H_3 sites, restoring electrical neutrality (Bird, 1971). The reaction is reversible because of wool's low affinity for chloride ions. Dye anions readily replace chloride ions.

Extrapolation of chlorine sorption with Kiton Red a^* (+red/-green) suggests that chlorine may influence dye uptake and increase yellowing (Fig 6.7.3 & 6.7.4). The increase in dyeability is consistent with the severe cuticular damage observed in methanol/Shurgain scoured fumigated fibers (See 6.10).

The ability of hydrogen peroxide treated hair to bind the acid dye Orange II is reported to be markedly decreased (Milligan & Wolfram, 1971). Robbins *et al.* (1968) attribute decreased dye binding to solubilizaton of protein, neutralization of dibasic amino acids, and an increase in the oxides of cystine. The cysteic acid

groups (SO_3^-) formed during oxidation bind very strongly with basic groups ($^+NH_3$), preventing the Orange II dye anion from displacing the sulfonic acid groups.

A reduction in acid-combining capacity is also attributed to amino group acetylation (Mathieson *et al.*, 1964). Ivanov (1987) reports that acetylated wool binds significantly less dye. The reaction of wool with dichlorvos breakdown may have resulted in acetylation. Milligan and Wolfram (1971) suggest that decreased dye-binding must be due to acetylation of lysine, arginine, and histidine. The formation of ornithine and a decrease in arginine in acetylated wool decreases dye binding. Acetyl chloride presumably acetylates serine, threonine and tyrosine side chains, decreasing acid Orange II binding capacity (Milligan & Wolfram, 1971).

Although the acid binding capacity of chlorinated wools is genera. markedly decreased, acid chlorination usually increases wool's affinity for acid dyes (Maclaren & Milligan, 1981, Bird, 1972; Mathieson, 1964). The phenomenon is proposed to occur when the loss of acidic groups from the cuticle during chlorination increases the permeability of the epicuticle. The loss of residues increases the affinity of the fiber for acid dyes and increases the rate of diffusion throughout the matrix (Maclaren & Milligan, 1981; Leeder & Rippon, 1981). In this study a reduced uptake of Methylene Blue and Kiton Red generally accompanied increased chlorine sorption. However, fumigated methanol scoured fibers exhibited both an increase

in chlorine sorption and an slight increase in Kiton red staining compared to the 35 day fumigated Shurgain scoured wool. The assumption that chlorine *per se* increased the affinity of the fibers for dyes is questionable. However, the degradative effects of chlorine on the topography of the cuticle, especially the epicuticle, probably decreased the physical barriers to dye diffusion.

Tester and Makinson (1982) suggest that a double layer of charge at the tips of the oxidized scales creates a positive space which attracts negative ions. This would account for some of the increased dyeability of oxidized wool at scale edges. Chlorination or oxidative treatment softens the cuticle and increases the number of charged and soluble molecules in the cuticle. This provides sites for dye bonding (Makinson, 1979; Röper *et al.*, 1984).

Heating wool fibers for 7 to 21 days at 50^{0}C generally increased wool's affinity for Kiton Red (ΔE 10.97 to 12.30 CIELAB units). Although the Shurgain scoured heated controls were usually stained more than unheated controls the 35 day control was stained less than the 7 and 21 day controls. The decrease in staining (ΔE 6.45 CIELAB units) in the 35 day heated control is probably due to the greasiness of Merino wool used for this heating period. Heat oxidation apparently increases dye uptake.

Methanol presoaking changes more than the protective PCL and F-layer of the fiber. The resistance to dye uptake in the methanol/Shurgain heated control is an enigma because lead acetate

staining indicated that the methanol control was oxidized.

Some sorption of Kiton Red was also expected in the 21 and 7 day fumigated wool because the lead acetate test showed slight oxidation and the oxidized methanol scoured fumigated fibers were stained. Although oxidation usually increases dye affinity other factors suppressed dye uptake.

Decreased Kiton Red affinity in fumigated wool may be caused by oxidized disulphide bonds being converted to $-SO_3^-$ which repel dye anions (Tester & Makinson, 1982). Acid damaged fibers stain less than undamaged fibers. Although a slight pH decrease caused an initial increase in dye uptake further decreases in pH lowered the dye uptake. Severe damage in the methanol scoured fumigated samples increased dye uptake.

It is apparent that dye uptake is more than a physical process. Because anionic or cationic dye molecules are bound to charged sites in the fibers, changes in the epicuticle and CMC alter the absorption of cationic Methylene Blue dye and anionic Kiton Red. Mechanisms governing dye affinity and resistance to dyeing in dichlorvos contaminated fibers are complicated.

6.10 Fluorescent Microscopy with Acridine Orange

Orange-red fluorescence on Acridine Orange (CI Basic Orange 14) dyed wool fibers indicates acid damage. Fumigated fibers dyed with Acridine Orange were acid damaged. Damage was first observed by

locating areas of orange-red fluorescence on the fibers which are normally green under ultra-violet light (Appleyard & Lees, 1964; Nossar *et al.*, 1973; Zhao & Johnson, 1986; Zhao & Pailthorpe, 1987). Zhao *et al.* (1986) suggest that the fluorescence is caused by the formation of serine and threonine hydrogen sulphate which absorb more basic dye than other areas of the fiber. Zhao *et al.* (1986) exposed fibers to H_2SO_4 and indicated that serine and threonine hydrogen sulphate were products of the reaction between the amino acid residues and the acid. The only acid that reacts similiarly with hydroxyl groups in this experiment is H_3PO_4. By absorbing more dye, the extra dyestuff "self-extinguishes its own fluorescence" by absorbing blue light and emitting orange/red light (Zhao *et al.*, 1986). Thus the green fluorescence is changed to red.

Acridine Orange is used to differentiate ribosomal RNA from DNA; absorption of the dye by RNA causes a red shift in the absorption spectra. According to theory, the dye and phosphate (1/1 ratio) form a complex which binds to negative sites in the RNA forming an aggregation of dye molecules (Gurr *et al.*, 1974). Bound phosphate groups from the hydrolysis of dichlorvos may provide sites for dye complexes to form. Another source of PO_4^- groups in wool may be the nuclear remnants (ENDO A1 fractions) in the endocuticle. If so, small pockets of red fluorescence from RNA enhanced by intercalated dye stacking could occur in damaged fibers. Garner (1966b) reports orange fluorescence in the untreated orthocortex,

which contains nuclear remnants and phospholipids, and red fluorencence in acid damaged fibers. Apparently macromolecular disorganization or degradation permits additional binding of dye molecules which increases red fluorescence (Gurr *et al.* (1974).

Orange-red fluorescence was observed to be concentrated along the periphery of the scales. The most severely damaged fibers tested with Acridine Orange were taken from a woolen hat, an artifact in the case study. These fibers had pronounced orange-red outlining on their distal and lateral scale edges. Similiar patterning was observed in fibers fumigated for 21 days. In other micrographs distinct spots of red-orange fluorescence in fibers from the woolen hat were similiar to the spots in fibers fumigated for 21 days (cf. Plate 3.1.4 to Plate 6.10.1). Photomicrographs suggest that dye diffusion occurs between the gaps of scales. The hydrophobicity of the fiber surface apparently causes uneven distribution of acid resulting in local accumulations of acid. The most vulnerable areas appear to be between the scales (Plate 6.10.2).The area beneath the scale tips provides a region of impingement where fumigant molecules could strike and rebound (Tester & Makinson, 1982). Capillary action could increase fumigant concentration at contact points between fibers, or in irregular sections of damaged fibers (Zhao *et al.*, 1986). The hypotheses are consistent with the fluorescent patterning observed in Plates 6.10.2 & 6.10.3.

Splaying of the cuticle (Plate 6.10.3) suggests that the

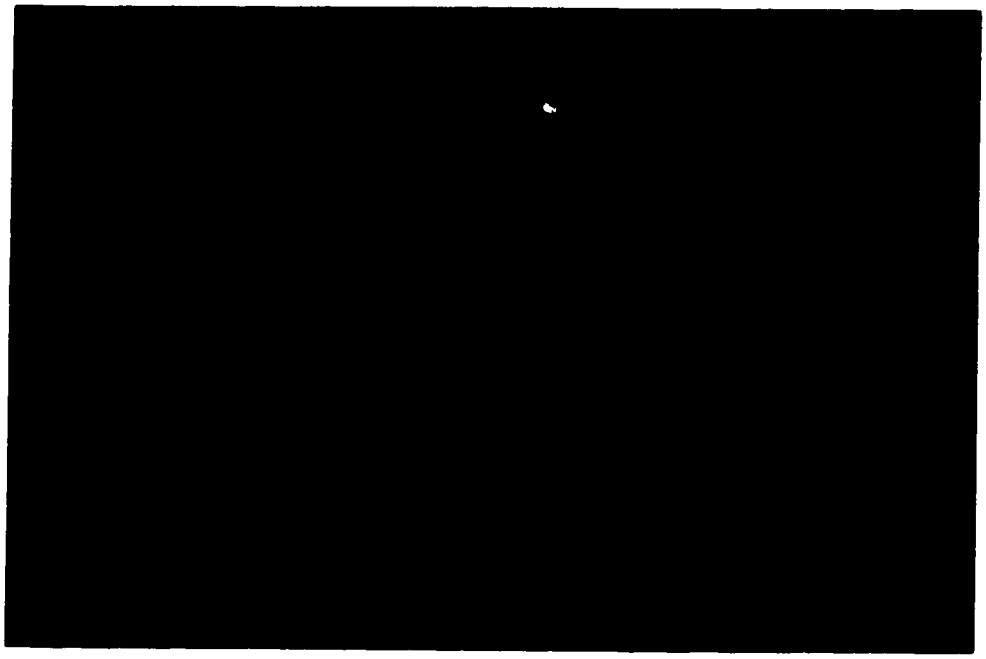


Plate 6.10.1 Dark red-orange spots in fluorescent photomicrograph of Wool Fiber Fumigated with S.W.A.T.® for 21 days at 50°C

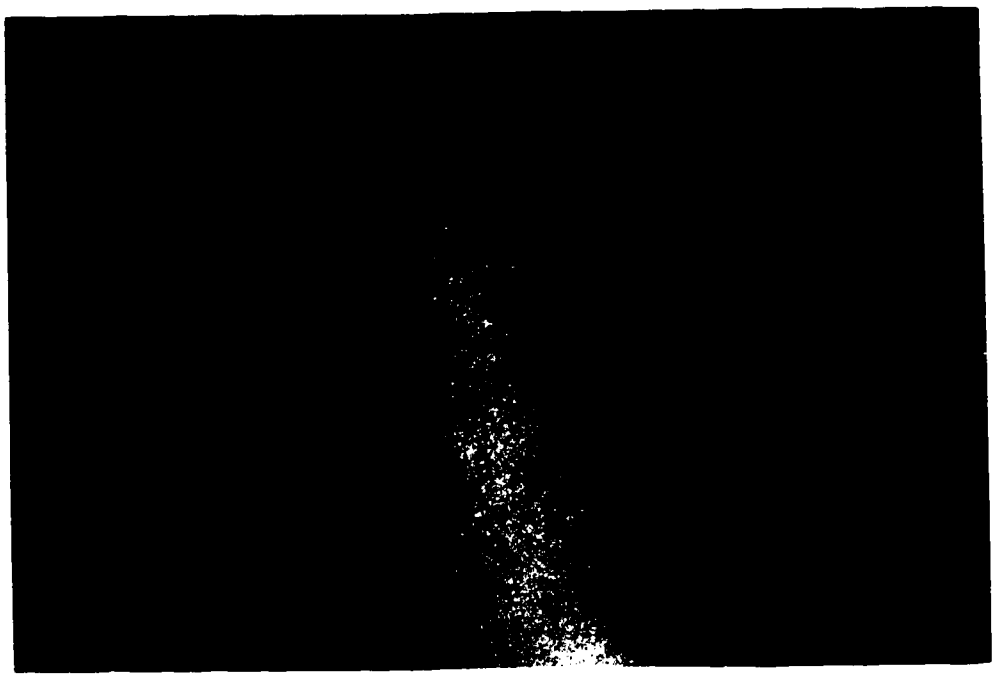


Plate 6.10.2 Fluorescent photomicrograph of Wool Fiber Fumigated with S.W.A.T.® for 35 days at 50°C.

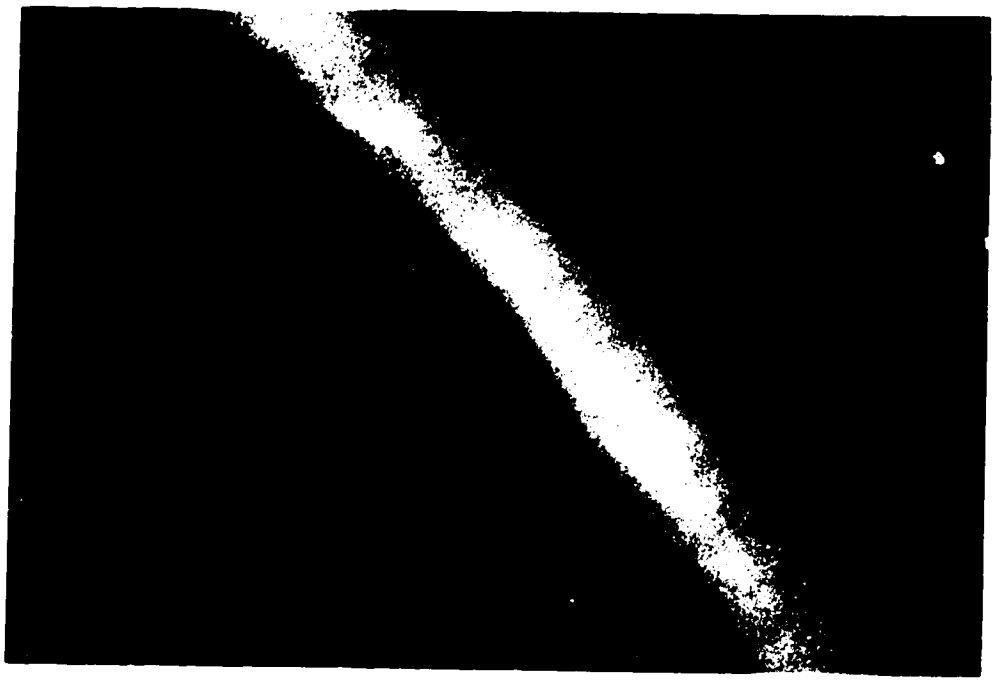


Plate 6.10.3 Protruding scales on fluorescent photomicrograph of wool fiber fumigated with S.W.A.T.® for 21 days at 50°C.

fumigant dissolves the intercellular cement which attaches the cuticle to the matrix (Makinson, 1971; Zhao, 1986). Damaged scales were broken and separated from the cortical sheath.

Protruding scales were observed on many fibers (Plate 6.10.3). By rotating or sliding the fibers along oppositely oriented fibers the scales were detached and visible with the microscope. Ley *et al.* (1985) found that detached scales could be dislodged by sliding a fiber against the scale tips of another fiber. The scales were also disrupted during dyeing with Acridine Orange and mounting. According to Makinson (1971) dry chlorination attacks the cementing matrix causing fragile shell-like scale fragments to detach (Makinson, 1971). Shell-like fragments were observed during microscopic examination.

Metachromatic fluorescence of the acid damaged fibers was achieved in spite of diminished dye affinity. Apparently severe acid damaged increases localized dye sorption. Longer fumigation periods may have increased red-orange fluorescence in severely damaged fibers.

Color difference tests were conducted with Acridine Orange (basic dye) to substantiate the Methylene Blue (basic dye) test. It is unusual for wool fibers to resist dyeing with both acid (Kiton Red) and basic dyes. Differences in the affinity of wool for Acridine Orange and Methylene Blue may be be attributed to the method of application and to the intrinsic nature of the dyes. Fibers dyed

with Acridine Orange were dyed at the boil. Heat increases diffusion of dye into the fibers.

Table 6.10.1 Acridine Orange (C.I. Basic Orange 14) Color Difference (CIELAB units) for Wool Fibers Exposed to S.W.A.T.® Resin Strips at 50°C in a Desiccator in the Dark.

		ΔE Color difference (CIELAB units)	
Treatment	Time (days)	Heat[a]	S.W.A.T.®[b]
Shurgain Scour	7	8.24	13.78[c]
	21	1.2	15.73[c]
	35	5.19	9.20[c]
Shurgain/ Methanol Scour	7	2.64	19.61[c,d]

[a]ΔE - difference between no heat and heat
[b]ΔE - difference between heat and S.W.A.T.
[c]ΔE - less dye pick-up than heated control
[d]ΔE - some of the difference attributed to yellowing

6.11 Chlorine and Phosphorus Sorption by EDXA

Qualitative EDXA was used to estimate the sorption and desorption of the fumigant. Chlorine and phosphorus, by-products of S.W.A.T.®, were detected in the EDX analysis (Fig. 6.11.1 & 6.11.2). Chlorine was rapidly absorbed by the Shurgain scoured wool during the first 7 days of fumigation with S.W.A.T.® at 50°C. Only small

amounts of chlorine were removed by washing the wool fibers in distilled water and methanol after fumigation (Fig. 6.11.1). The resistance of chlorine to removal by washing suggests that chlorine reacts with the wool fiber.

Phosphorus sorption increased steadily during the first 21 days of fumigation (Fig. 6.11.2). Washing with water and methanol did not remove appreciable amounts of phosphorus from the wool fibers (Fig 6.11.2). The increase in phosphorus during fumigation and apparent removal of phosphorus from the wool fibers (14 and 21 days) with distilled water may represent variability in measurements with EDX analysis (Ploem & Tanke, 1987; Morgan, 1980; Levin, 1986), or differences in the sorption and desorption of phosphorus. The desorption of phosphorus from fibers in air was similiar to sorption. Wool fibers retained chlorine and phosphorus.

The amount of chlorine and phosphorus absorbed by fibers fumigated at ambient temperatures and 50°C differ (cf. Fig. 6.11.3 with Fig. 6.11.4). The small phospho peak in Figure 6.11.3 indicates that the wool has absorbed a small amount of dichlorvos after 96 hours of fumigation at ambient temperatures. A small chlorine peak is visible in Figure 6.11 4 after 3 days of fumigation at 50°C. Phosphorus sorption continues to predominate at lower temperatures and chlorine sorption at higher temperatures after 3 or 4 weeks of fumigation (cf. Fig. 6.11.3 & 6.11.5 with Fig 6 11.4 & 6.11.6). Higher temperatures and lower pH are reported to increase

chlorine sorption (Kantouch & Fattah, 1971). This is unusual because sorption processes are exothermic and generally decrease at higher temperatures (Kantouch & Fattah, 1971; Gillett *et al.*, 1972). Similiar observations of increased sorption of halogens by wool at increasing temperature are reported (Kantouch & Fattah, 1971).

The mode of degradation and by-products released from dichlorvos resin strips are suggested to be governed by temperature. EDX analysis indicates that the rate of release of the fumigant at 50°C and sorption by wool is greater than the release of the fumigant and sorption at ambient temperatures (cf. Fig. 6.11.4 & 6.11.6). Earlier resin strip studies found that the rate of fumigant release was altered by changing the temperature and relative humidity (Gillett *et al.*, 1972). The EDX spectra from fumigated wool, and color changes in the strip, indicate that the decomposition of the strip and by-products varies with temperature and moisture. Heated resin strips turned green, whereas, the unheated strips remained yellow. The observed color change in the strips may suggest a different mode of degradation at 50°C.

The resistance of phosphorus and chlorine to removal by both a 20 minute wash in distilled water or methanol suggests that fumigant by-products are bound to the fiber. Only small quantities of chlorine and phosphorus were removed by distilled water and methanol (Fig. 6.11.1; 6.11.2).

The EDX graphs of wool fibers fumigated with Vapona® at

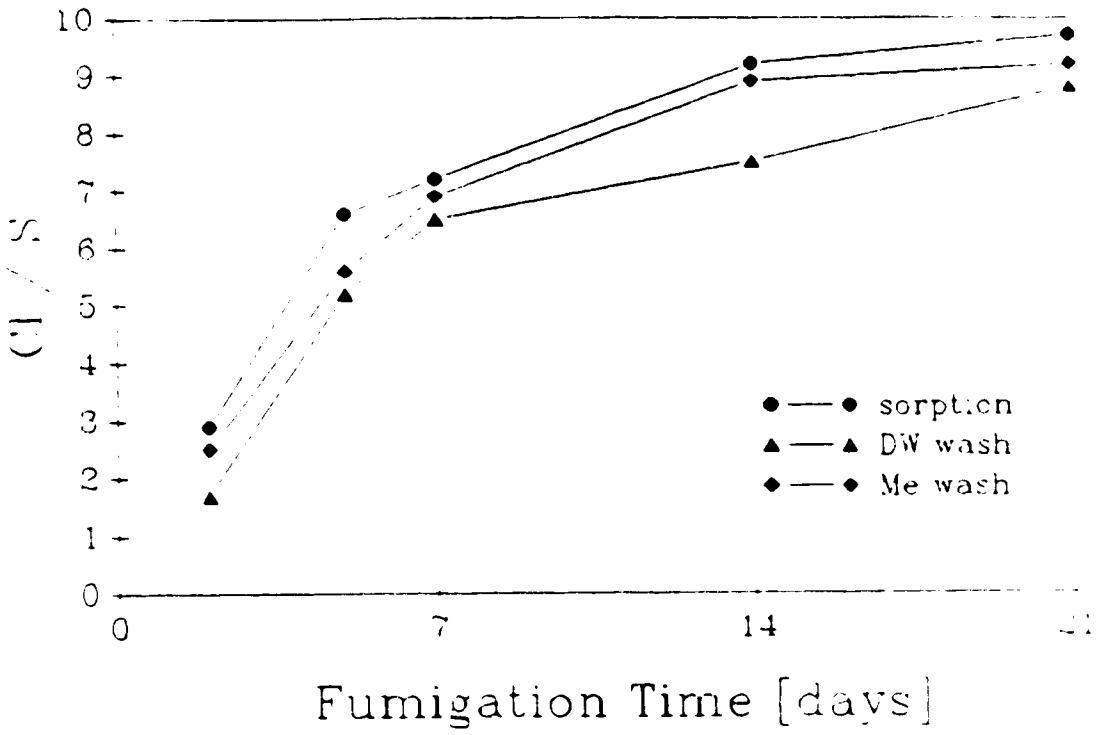


Figure 6.11.1 Chlorine sorption of S.W.A.T.® at 50 C° and desorption with distilled water and methanol

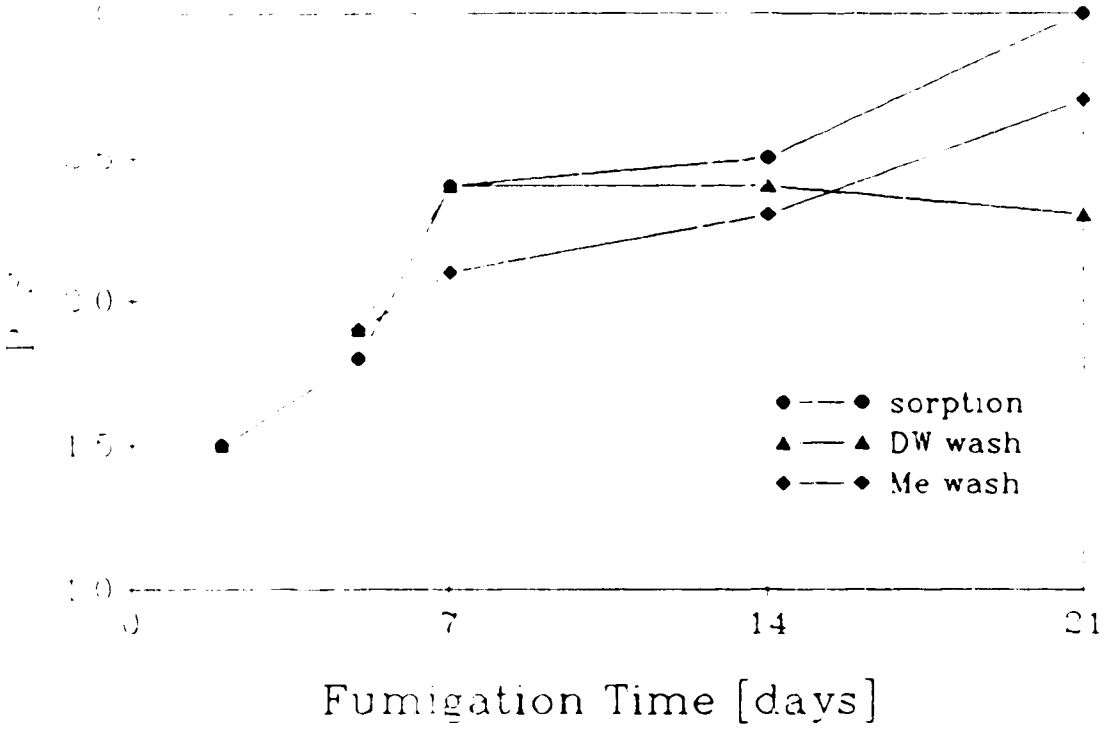


Figure 6.11.2 Sorption of p' phorous at 50°C and desorption with distilled water and methanol.

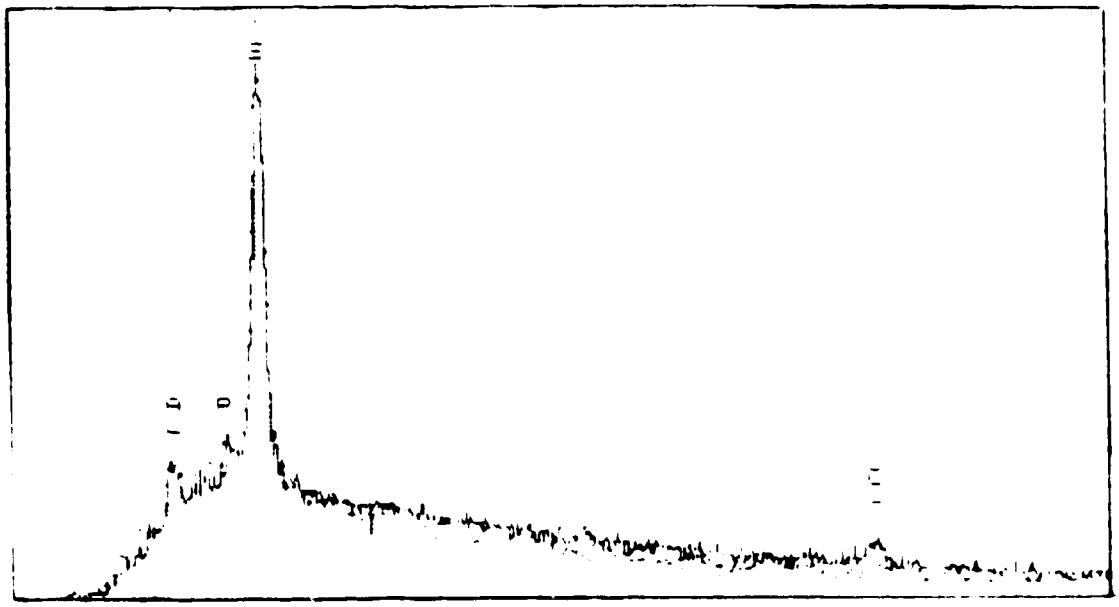


Figure 6.11.3 EDX graph of Shurgain scoured wool fibers fumigated with S.W.A.T.® at ambient temperature for 96 h.

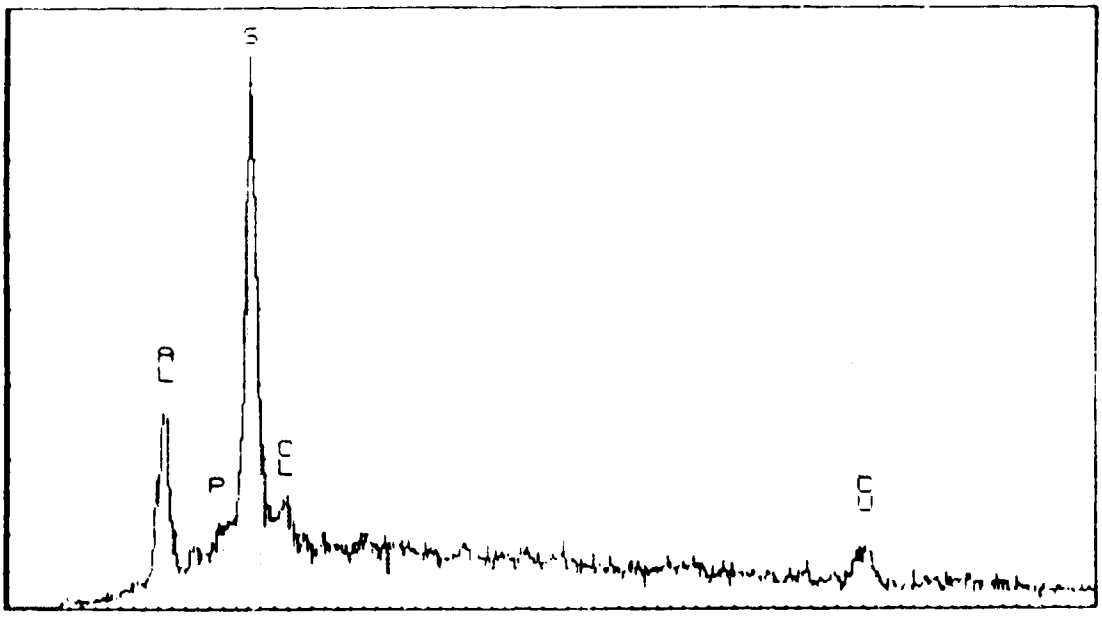


Figure 6.11.4 EDX graph of Shuttlin scoured wool fibers fumigated with S.W.A.T.® at 50°C for 72 h (3 days) in a dark oven.

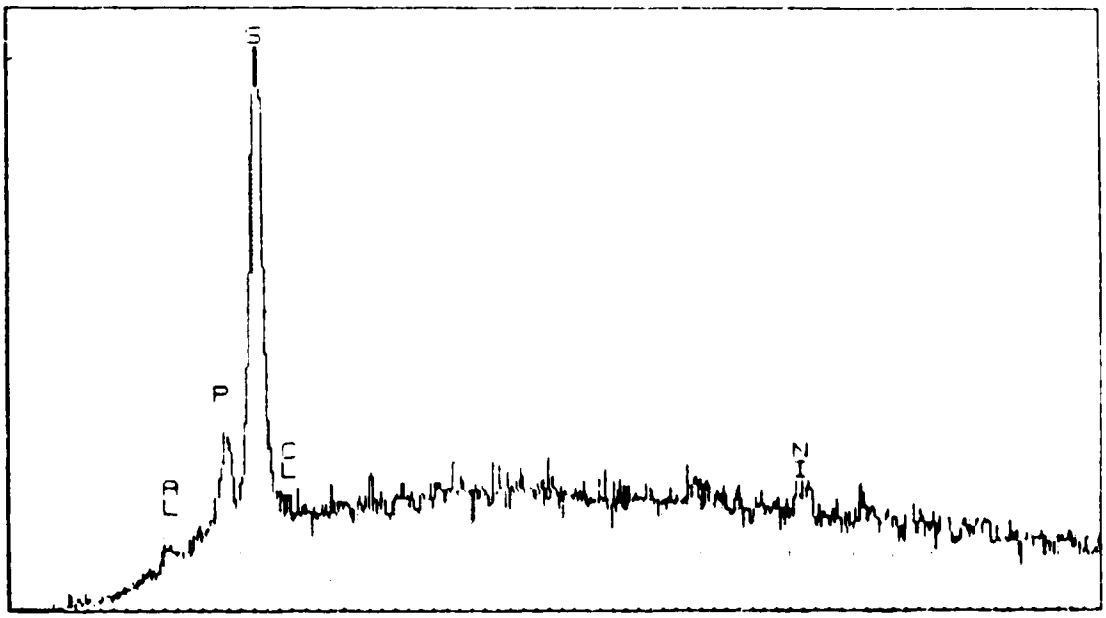


Figure 6.11.5 Sorption of chlorine and phosphorus at ambient temperature after 673 h (28 days) of fumigation with S.W.A.T.® in a desiccator in a fume hood.

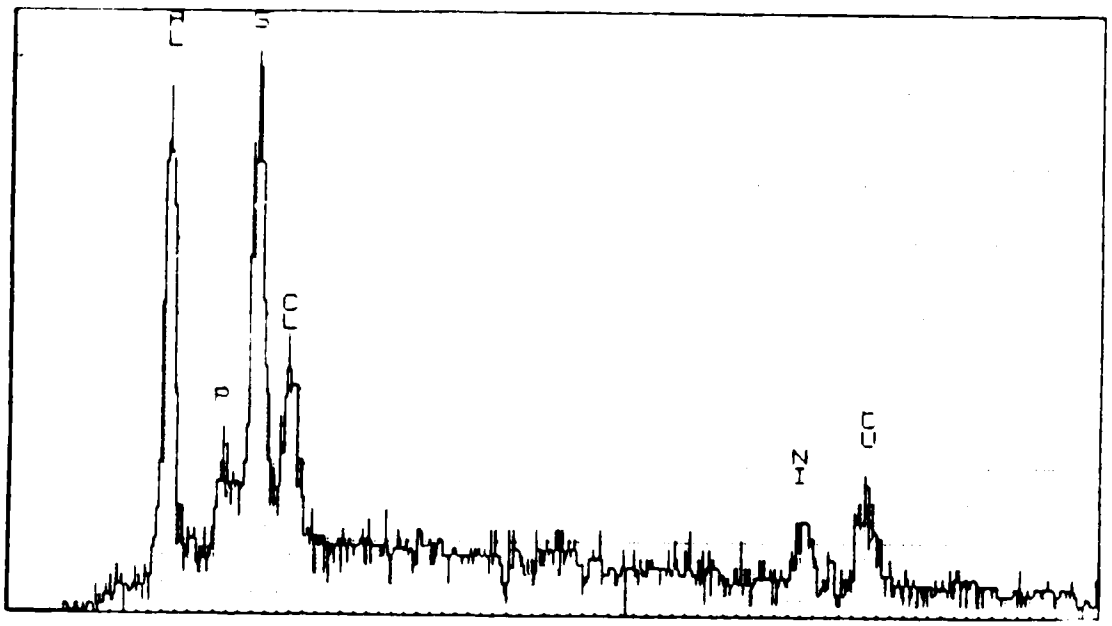


Figure 6.11.6 Sorption of chlorine and phosphorus at 50°C after 21 days of fumigation with S.W.A.T.® in a desiccator in a dark oven.

ambient temperature (Fig. 3.1.1) and S.W.A.T.® at 50°C (Fig. 6.11.7) are included to emphasize temperature dependent variability. Both graphs represent wool fibers with unknown pre-treatment histories. The ambient temperature sorption graphs were obtained from a woolen bonnet fumigated *in situ* (See 4.1) at the Edmonton collection centre. Although the preponderance of phosphorus may be an artifact, such as phosphate detergent residue, it is more likely fumigant residue. Figure 6.11.7 represents a commercially scoured handspun Merino yarn that was fumigated for 6 days at 50°C with S.W.A.T.®. The preponderance of chlorine indicates that commercially scoured wool fibers initially have the capacity to absorb more chlorine than fibers scoured in only a mild detergent. The sorption graphs from the woolen bonnet and the commercially scoured wool indicate that wool has a tremendous capacity to absorb both chlorine and phosphorus. The EDX graphs recorded in Figures 3.1.1 and 6.11.7 suggest that the sorption of chlorine and phosphorus in this study are only a small portion of wool's capacity for fumigant sorption Figure 6.11.8 shows the EDX graph of wool fibers prewashed in methanol/Shurgain and fumigated for 21 days.

Surface contaminants appear to have a profound effect on chlorine and phosphorus sorption of S.W.A.T.®. The removal of components of PCL such as wool grease during commercial scouring and with methanol increased chlorine and phosphorus sorption.

The sharp increase in phosphorus in the commercially scoured

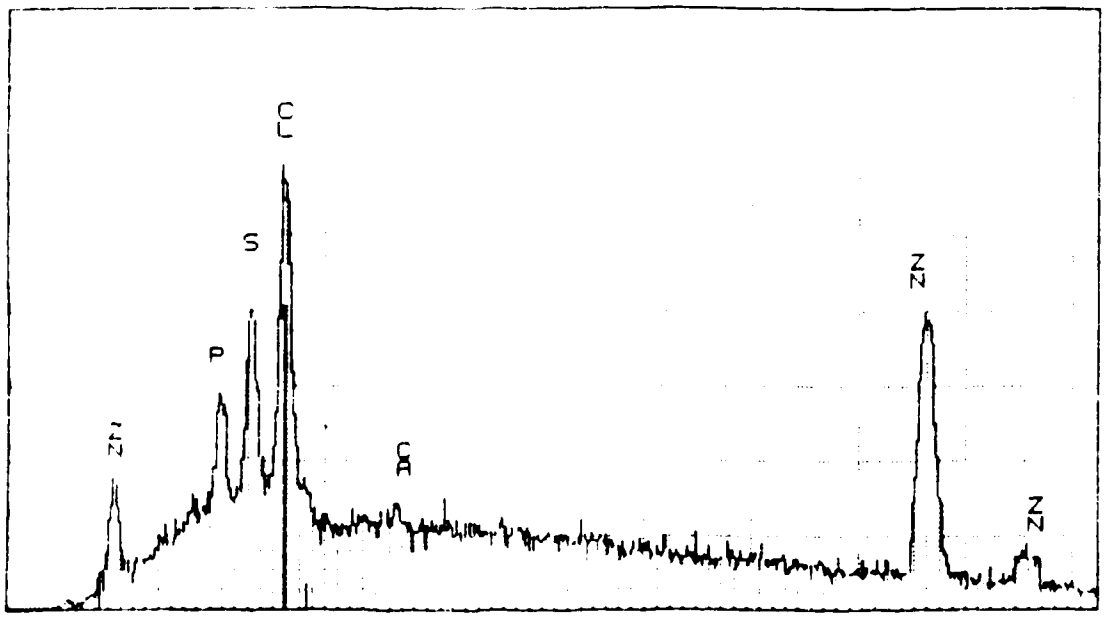


Figure 6.11.7 EDX graph of commercially washed Merino wool fibers fumigated with S.W.A.T.® at 50°C for 6 days.

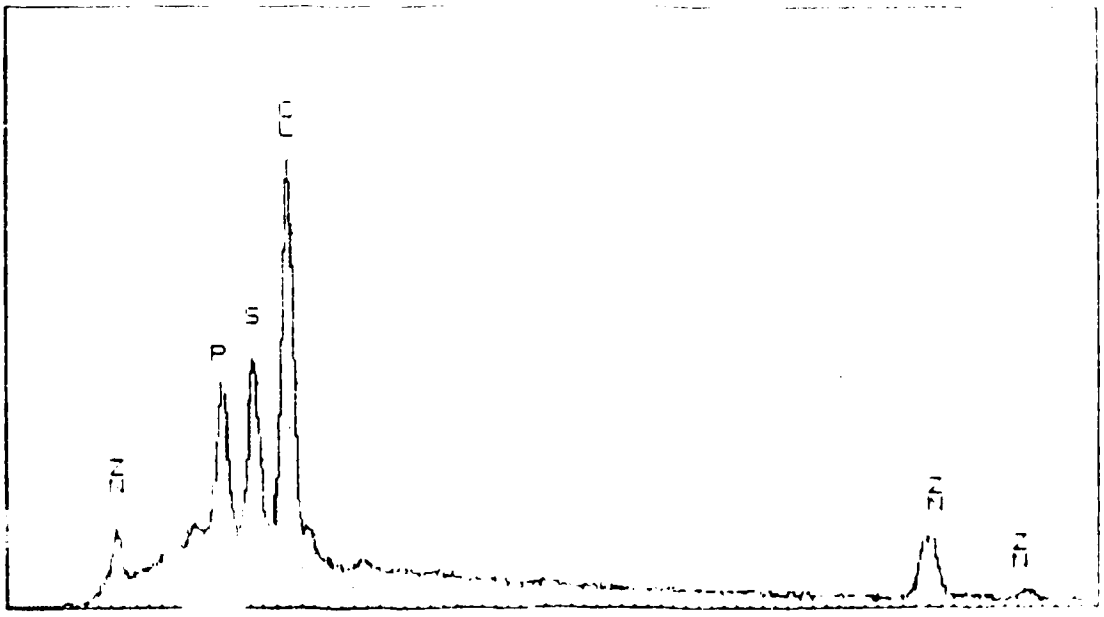


Figure 6.11.8 EDX graph of methanol/Shurgain scoured fiber after fumigation with S.W.A.T.® at 50°C for 21 days.

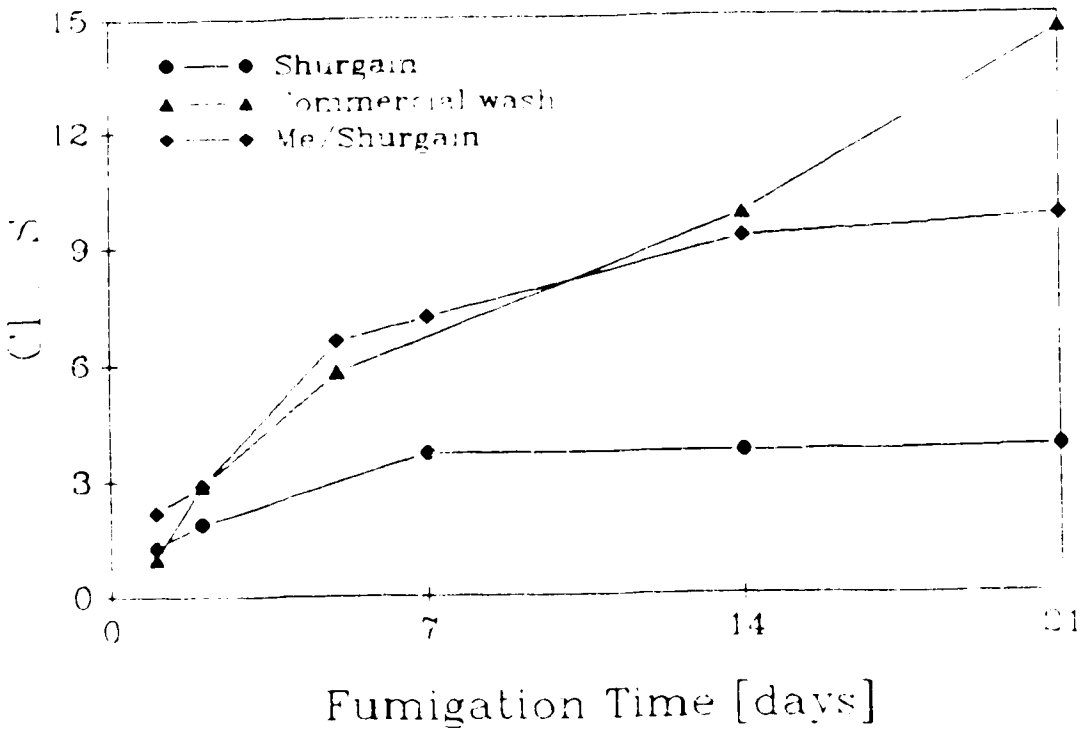


Figure 6.11.9 Chlorine sorption at 50°C on wool fibers after scouring with Shurgain, Shurgain/methanol, or a commerical wash.

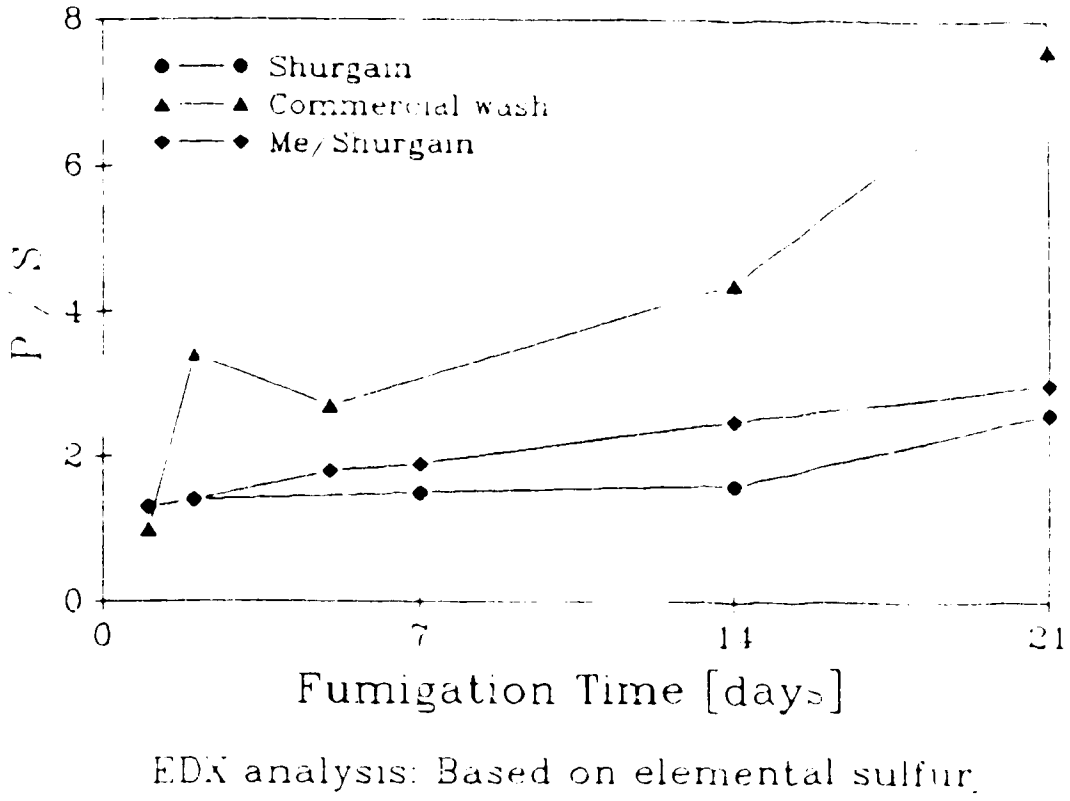


Figure 6.11.10 Phosphorus sorption at 50°C of wool fibers after scouring with Shurgain, Shurgain/methanol, or a commerical wash.

wool observed at 2 days is probably anomalous (Fig. 6.11.10). The occurrence of calcium phosphate (hydroxyapatite and brushite) inorganic crystals is common in wool (Blakey *et al.*, 1971). The crystals are generally located in cell pseudopodia and areas adjacent to convoluted cell walls. Calcium phosphate accounts for 25% of the mineral component of wool which is 0.5-1.0% of the mass (Blakey *et al.*, 1971). Small calcium peaks were also observed in some of the EDX spectra.

Other researchers have found that phosphate ester insecticides bind to wool (O'Loughlin, 1982). Jones (1983) found that only a small proportion of the insecticides were unfixed and removable by Soxhlet extraction with a mixture of dichloromethane, methanol and water. Although Jones (1983) found that spinning oil had little effect on the binding or sorption of phosphate and sulfur ester insecticides the results of this study suggest that wool lipids and PCL are an effective barrier. Conservators may be interested in long-term studies by Jones (1983) who predicts that the half-life of oxygen ester insecticides will be slightly less than the expected half-life of 15 years (20°C and 65% RH) of sulfur analogues. Hydrolysis of phosphate esters is suggested to be catalyzed by hydroxide ions and is, therefore, dependent on internal pH (Jones, 1983b). In this study aqueous extract pH indicates low fiber pH which retards the hydrolysis of phosphate esters (Eto, 1974).

Fumigation in open museum spaces would not decrease pH as

drastically as contained fumigation. Open space fumigation would favor hydrolysis of phosphate esters and phosphorus binding because the hydrolysis of dichlorvos is pH dependent (See 2.2).

Several investigations have found that chlorine absorption may be irreversible. Cacella & White (1955) found that a portion of gaseous HCl is irreversibly absorbed by wool.

Available experimental data on the effect of chlorine sorption induced changes in the physical and chemical properties of wool. Table 6.11.1 is a summary of changes correlated with increased chlorination.

Table 6.11.1 Effect of Chlorine Sorption on the Physical and Chemical Properties of Wool

Chlorine Sorption	Effect on Wool
Chlorine increases	pH of aqueous extract decreases
Chlorine increases	yellowing increases
Chlorine increases	basic and acid dye uptake changes
Chlorine increases	textile properties of yarns change
Chlorine increases	topographical degradation of fiber becomes more severe

Chlorine and its derivatives have a significant effect on the chemical reactivity and physical performance of keratin fibers. If wool is oxidized with NaOCl the oxidized species vary with pH

Hypochlorous acid predominates between pH 3-6, hypochlorite above pH 8, and free chlorine below pH 1.5 (Maclaren & Milligan, 1981). As a consequence chlorine damage to wool varies with pH

As stated previously, the degradation of dichloros resin strips degradation is postulated to produce a variety of chlorine derivatives capable of causing topographical, chemical and physical changes. The hydrolysis of DDVP, DMTP and Dipterx and subsequent oxidation produces mono-, di- and tri- chloroperacetic and acetic acids, and the degradation of the polyvinyl chloride carrier produces small amounts of HCl (Williams et al.)

6.12 Scanning Electron Microscopy

Topographic changes in the fumigated fibers were evident in the micrographs of Plate 6.12.1 to Plate 6.12.2). Generalized erosion and stuffing off of scales altered the appearance of fibers. A viscous-like coating adhered to the surface and appeared to bind fibers together (Plate 6.12.3 & 6.12.4). Although the nature of the viscous-like material was not determined it could be denatured cuticular protein, exudate from cellular or intercellular complexes, or a mixture of lipids and dissolved cuticular components. Quanitative analysis to determine the chemical composition of the coating was not performed.

The chlorination attacks the cementing matrix between cuticle scales causing the scales to become "visco-elastic or plastic".

these changes are the result of chemical degradation of part of the contents of the cuticular cells" (Makinson, 1971, p. 1084). The softening or plasticizing increases the area of contact between scales causing adhesion (Plate 6.11.4). Makinson (1971) suggests that hydrostatic pressure inside the cell after chlorination brings the CMC membranes together so that van der Waal's forces hold the scales together. The scales in Plate 6.11.4 appear to be firmly cemented together and coated with a viscous-like material. Micromanipulation during microscopy clearly indicated that the intercellular adhesion was disrupted. Tapping on the glass coverslip dislodged some of the cuticle scales.

By applying peroxide and chlorine to human hair in different sequences and for a varying number of cycles, Fair and Gupta (1985) were able to determine some synergistic effects of the reagents. Peroxide oxidation removed the scale structure of chlorinated hair leaving a smooth surface. Chlorination *per se* apparently produces a wrinkled surface appearance attributed to loss of proteinaceous material beneath the epicuticle (Fair & Gupta, 1985) and causes different changes in the cuticle of hair than applying both reagents (Fair & Gupta (1985). Both peroxides and chlorine are breakdown products of dichlorvos.

Localized lipids coating the fiber surface were visible with SEM and could be substantiated by the greasy handle of wool. In spite of the lipid layer the fumgiant modified the surface

and diffused into the fiber. Surface bubbles and surface pitting are evident on fumigated wool fibers in Plate 6.12.6.

Increased wettability during dye tests suggests alteration to the F-layer, the lanolin film, and PCL on greasy fibers (Carr *et al.*, [illegible], Arthur & Christoe, 1984). Fatty acids are bound covalently to the exocuticle via ester links to serine and threonine (Coder [illegible] *et al.*, [illegible]). The increased wettability suggests disruption of the hydrophobic surface layer (Griffith, 1967; Baumann *et al.*, 1986).

Researchers report that the cuticle is released prior to total cells (Kulkarni & Bradbury, 1974). Some of the micrographs indicate that the cuticle is separated from the cortex (Plate 6.12.9). In a moist environment axial cracks usually extend along cell membranes and macrofibril boundaries (Dobb & Murray, 1975). Electron microscopy studies confirm that the areas most susceptible to stress are the cell membrane complex, endocuticle and intermacrofibrillar regions, and to a lesser extent the CMC of the cortex (Orwin & Thomson, 1975; Dobb & Murray, 1975; Ito *et al.*, 1984; Dobb & Rogers, 1969; Dobb, 1966). The difference in susceptiblity among geographical regions in wool fibers is attrib[illegible] differences in chemical composition. Fumigation with dichlorvos appears to have exposed the resistant membranes of the cell membrane complex (Plate 6.12.9).

Nakamura *et al.* (1975) suggest that the cystine content in the intercellular material (δ-layer) between cuticle cells is lower

and the COOH content is higher than the intercellular material between the cuticle cells and cortex or cortical cells. Another finding is that cystine and COOH content of the δ-layer of the cuticle cells is similiar to the endocuticle (Nakamura *et al.*, 1975). Canals joining the cuticle δ-layer to the endocuticle form a network for diffusion through the epicuticle, and a backdoor entry for interstitial diffusion. The appearance of fibers in Plates 6.1.3 and 6.12.5 suggest dissolution of the cuticle and outward diffusion of denatured protein through the epicuticle and interstices between cuticular cells. It appears that there is total disruption of the CMC in the severely damaged fiber in Plate 6.11.9. Uneven fiber degradation produces voids where the internal structure has been destroyed (Swift, 1975).

Cuticle integrity is desirable because it stabilizes the cortex both mechanically and chemically. Variability in surface degradation is supported by the observations of other researchers. Studies of chemical modification using SEM microscopy indicate that fiber degradation is extremely variable (Kulkarni & Bradbury, 1972). Both native fibers and extensively damaged fibers were observed in the same field of SEM micrographs. Reagents such as formic acid and peracids are known to descale fibers (Kulkarni & Bradbury, 1972). Because of the extreme variability among wool fibers and within wool fibers, it is necessary to use several methods to detect damage.

Plate 6.12.1 Native Merino wool fibers washed in Shurgain and degreased in ether

Plate 6.12.2 Severely damaged wool fibers fumigated with S.W.A.T.® days at 50°C

Plate 6.12.3 Viscous-like coating adheres to fibers fumigated with S.W.A.T.® for 21 days at 50°C.

Plate 6.12.4 Dissolved cuticular proteins bind commercially [illegible] handspun Merino fibers together after 21 days [illegible] with S.W.A.T.® at 50°C.

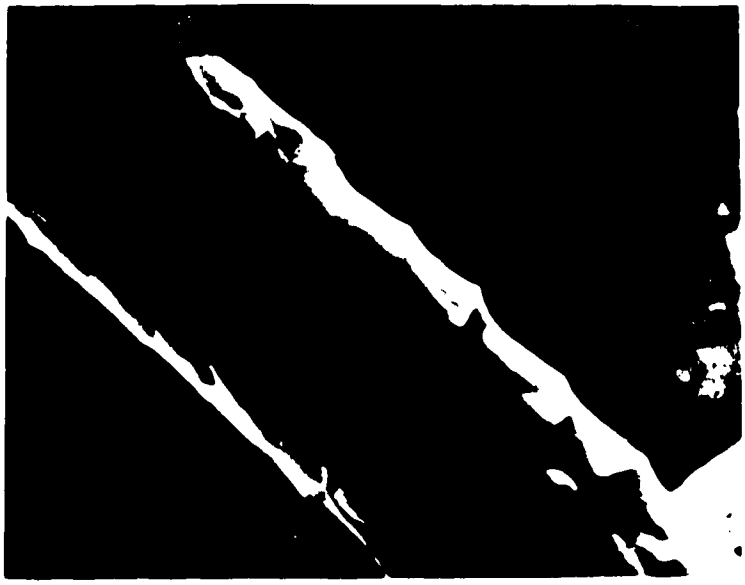


Plate 6.12.5 Eroded scales of methanol scoured Merino fiber after fumigation with S.W.A.T.® for 21 days at 50°C. Crack is an artifact from electron beam.

Plate 6.12.6 Bubbles and pits on surface of commercially scoured handspun Merino wool fumigated with S.W.A.T.® for 6 days at 50°C.

Plate 6.12.7 Severely eroded methanol scoured Merino fibers fumigated with S.W.A.T.® for 21 days at 50°C.

Plate 6.12.8 Wrinkling of cortical sheath in severely [illegible] methanol scoured fiber after 21 days of fumigation with S.W.A.T.® at 50°C.

Plate 6.12.9 Exposure of cortex in severely degraded methanol scoured Merino wool after fumigated with S.W.A.T.® for 21 days at 50°C.

Plate 6.12.10 Erosion of cuticle scales in methanol scoured Merino wool after fumigation with S.W.A.T.® for 7 days at 50°C

7. CONCLUSIONS

7.1 Review of the Problem Statement

Throughout this research answers to the problem statement: "Is dichlorvos a suitable space fumigant for ancient and historic museum collections containing keratin fibers?" were sought. An investigation of the effects of dichlorvos space fumigation on wool fibers partially filled a void. Since keratin fibers are often damaged by insects in museums it was appropriate to investigate the effects of dichlorvos on wool fibers (Pretty, 1976; Reagan (1982).

Dichlorvos is used throughout the world as a space fumigant in museums. About 40 per cent of the respondants in a museum pesticide status survey (Edwards *et al.*, 1981) reported using Vapona® for pest control. The suitability of DDVP space fumigation for artifacts was queried by a few authors. Health hazards and target species efficacy were queried by others. Dichlorvos is an extremely toxic acetycholinesterase depressant and a bioaccumulative xenobiotic that is a suspected carcinogen and teratogen. Long-term exposure t museum pesonnel could be harmful.

The paucity of information on the effect of dichlorvos on textile fibers justified the need and urgency for this study. A review of pertinent literatur and analyses by Williams *et al.* (1986) indicated that by-products of dichlorvos resin strip hydrolysis are potentially harmful. The Edmonton collection provided an *in situ* example of damage to fumigated wool textiles and furs.

Wool, a composite material consisting of a layered cuticule, segregated cortex, and in some cases a medulla, and minor histological components, is a heterogeneous protein fiber with complex histo-chemistry. Fumigation yellowed the wool fibers due to chlorine absorption and changed dye absorption and tensile properties. SEM micrographs of fumigated fibers showed cuticular and cortical damage. The degree of observed damage generally increased with fumigant exposure. However, the greater damage observed in methanol and commercially scoured fibers than in Shurgain scoured wool indicated that increased fumigant sorption was a more important parameter than fumigation time *per se*.

The pH of the aqueous extract decreased from pH 6.8 (control) to pH 3.4 in the methanol/Shurgain scoured fibers fumigated for 7 days at 50°C. Decreases in pH appear to be dependent on the amount of fumigant absorbed by wool fibers. Fluorescent photomicrographs and tests with Krais-Viertel and Herbig reagents indicated cuticular acid damage. The decreased affinity for Methylene Blue dye also suggested a decrease in pH.

The absorption of Methylene Blue, Acridine Orange and Kiton Red dyes generally increased in the initial period of fumigation and decreased with continuing exposure to dichlorvos. Changes in the dye uptake indicated that the fumigant reacts chemically with the fiber. Electrostatic, morphological, and chemical changes are implied. Oxidation, acylation and esterification of amino acid

residues may have occurred. Initial increases in dye uptake suggest oxidative damage while further changes are indicative of increased acidity and localized damage which decreased physical barriers to the dyes. A decrease in staining with lead acetate reagent was observed in fumigated fibers indicating oxidation of -SSH.

Apparently dichlorvos residues are not easily removed with standard treatments. Only a slight decrease in phosphorus and chlorine was detected in fibers washed in methanol or distilled water. The primary objective of this research was to investigate the damage to wool exposed to dichlorvos resin strips under controlled laboratory conditions. The results of the laboratory studies indicate that dichlorvos resin strips damage wool fibers.

7.2 Summary

In the literature and laboratory, dichlorvos was found to be an extremely toxic chemical. The author experienced physiological repercussions attributed to dichlorvos toxicity during the initial stages of experimental work. Headaches, nausea, palpitations, slight ataxia, and visual disturbances were experienced. The research was completed by limiting the time of exposure and wearing a respirator and full body covering while conducting laboratory experiments. Conservation students working at the Edmonton collection center also experienced headaches, nausea, and visual disturbances. Recent reports indicate that dichlorvos is a bioaccumulative xenobiotic.

This suggests that long-term exposure of museum personnel to dichlorvos could be harmful.

Some museum pests are resistant or acquire resistance to dichlorvos. Dichlorvos is not an effective biocide for all stages of an insect's life-cyle because as an acetylcholinesterase inhibitor, its toxicity is dependent on the developed nervous system and enzyme transmitter.

It is evident that fumigation with high concentrations of dichlorvos resin strips causes chemical, physical, and mechanical changes to wool fibers. Wool given a methanol soak prior to fumigation showed an increase in sorption of chlorine and color change; therefore, keratin fibers that have been subjected to commercial scouring, and routine fabric maintenance before their acquisition into museum collections have the potential for considerable damage if fumigated with dichlorvos. The degradative action of laundering, abrasion, air pollutants, and weathering causes progressive deterioration (Slater, 1985; Veldman, 1966a; Cardamone & Brown, 1986; Cardomone *et al.*, 1987; Masschelein-Kleiner, 1980; Roger & Hays, 1943; Spedding, 1969; Zeronian *et al.*, 1986; Wentz, 1985; Watt, 1964; 1971). Manufacturing processes, solvents, and weathering alter and remove fractions of the PCL and CMC that apparently protect wool fibers from harmful dichlorvos vapours. Although more laboratory experiments need to be conducted to substantiate the theory of increased susceptibility of ancient and historic textiles

and artifacts composed of keratin, it is evident that the methanol soluble components provided some protection. The damage incurred during short term fumigation in the laboratory was similiar to the damage observed in the case study textiles.

The validity of accelerated testing at $50^{0}C$ is questionable because high concentrations of the fumigant in closed chambers at elevated temperatures may produce different by-products than ambient temperatures and concentrations recommended by the manufacturer. It is evident that damage occurs in an extremely short period of time. The results of this research do not preclude the need for long-term testing at ambient temperatures using concentrations suggested by manufacturers, and the need for a critical examination of textiles exposed to long term fumigation.

The extensive literature review and experimental data indicate that DDVP is not an innocuous substance. The use of dichlorvos as a fumigant for wool in museums is not recommended.

8. Recommendations for Future Research

The need for more research in museums and controlled laboratory research is apparent. Throughout this research many topics for future research have been identified. A few of the areas in need of investigation are given below.

1. Development of test methods for the study of wool degradation.
2. Development of methods for removing dichlorvos residues from textiles.
3. Identification of amino acid residues modified by DDVP resin strip fumigation.
4. The role of lanolin as a protective barrier to volatile pollutants and fumigants.
5. The effects of moderate temperatures ($< 70^{0}C$) on keratin fibers.
6. Repetition of portions of this study with known concentrations of DDVP and controlled RH.
7. Studies on the effects of long term fumigation on wool at $20^{0}C$ with recommended concentrations of dichlorvos.
8. Studies of changes in the wool fiber preceeding overt degradation.
9. Identification of the role of protective mechanisms in the fiber.
10. Studies of the condition of objects subjected to long-term

dichlorvos fumigation.

11. Assessment and measurement of pesticide residues present in museum objects.

12. Survey of health problems of personnel working in areas fumigated with dichlorvos.

13. Development of a responsible a[illegible]ed pest management system for museums

REFERENCES

Abbey Newsletter. (1987). Events in the new. *The Abbey Newsletter Bookbinding and Conservation.*

Alam, I. (1983). Insects pests of musuem materials their deleterious effects and methods of control. *Journal of Indian Museums.* (pp 142-159).

Alberts, B., Bray, D., Lewis, J., Raff, M., Roberts, K., & Watson, J D. (1983). *Molecular Biology of the Cell.* New York: Garland.

Alexander, P., Hudson, R. F., & Earland, C. (1963). *Wool Its Chemistry and Physics.* London: Chapman & Hall.

Alexander, P., Fox, M., & Hudson, R. F. (1951). The reaction of oxidizing agents with wool. The oxidation products of the disulphite and the formation of a sulfphonamide in the peptide chain. *Biochemical Journal, 49,* (pp 129-138).

Algie, J. E. (1979). Dieletric properties of keratin and cellulose. In F. Happey (Ed.). *Applied Fibre Science, Vol. 2.* London: Academic Press.

Al-Hariri, D. K., & Rattee, I. D. (1983). The production of easily dyed wool: An evaluation of saline ammonia scouring. *Journal of the Society of Dyers and Colourists, 99,* 335-339.

Allen, C. F., Dobrowski, S. A., Speekman, P. T., & Truter, E. V. (1985). Evidence for lipid and filamentous protein in Allworden Membrane. In M. Sakamoto (Ed.), *Proceedings for the 7th International Wool Textile Research Conference, Vol. 1,* (pp. 143-151). Tokyo, Japan: Nichiei Kosen.

American Conference of Governmental Industrial Hygienists (ACGIH). (1986-87). Dichlorvos. *Documentation of Threshold Values.* Xerox copy.

Anderson, C. A. (1983). Mechanisms of raw wool scouring - The role of the protein contaminant layer. *Textile Research Journal, 53,* 741- 757.

Anderson, C. A., & Christoe, J. R. (1984). Characteristics of the protein contaminant layer on Merino wool. *Textile Research Journal, 54,* 378-382.

Anon. (1981). Close-up. *Pest Control,* December, 1981, 56.

Appleyard, H. M., & Lees, K. (1965). Observations on untreated and chemically treated wool fibers after staining with fluorescent stains. *Journal of the Textile Institute, 56,* T38-T43.

Arai, K., & Nagaoka, N. (1985). Cross-linking structures of wool. In M. Sakamoto (Ed.), Proceedings of the 7th International Wool Textile Research Conference, Vol. I. (pp. 105-114). Tokyo, Japan: Nichiei Kosen.

Armes, N. J. (1984). Aspects of the biology of the Guernsey carpet beetle, *Anthrenus Sarnicus*

84.13.1-84.13.3). Windsor, England: Reedprint.

Ashman, F., Wesley, D. J., Achillides, N. Z. (1974). Space treatment of warehouses using Dichlorvos. *EPPO Bulletin: European and Mediterrraean Plant Protection Organization, 4*,(4), 429-445.

Ashrenazi, B. S. (1984). Evaluation of oxidative damage to wool hair. *Colourage, 31*(11), 30-32.

Asquith, R. S., & Leon, N. H. (1977). Chemical reactions of keratin fibers. In R. S. Asquith (Ed.), *Chemistry of Natural Protein Fibers*. New York: Plenum.

Asquith, R. S., & Otterburn, M. S. (1971). Self-crosslinking in keratin under the influence of dry heat. *Applied Polymer Symposium, 18*, 227-287.

Astbury, W. T. (1933). Some problems in the X-ray analysis of the structure of animal hairs and other protein fibers. The structure of keratins. *Transactions of the Faraday Society, 29*,193-211.

ASTM. (1981). 2256-80 Strength, breaking load and elongation of yarn by the single strand method. *1981 Annual Book of ASTM Standards*. Easton, Md: American Standard

ASTM. (1981). Standard test method for pH of aqueous extracts of wool and similar animal fibers- D 2165-78. *Annual Book of ASTM Standards*. Easton, Md: American Standard

ASTM. (1985). Conditioning for testing D-1776-85. *Annual Book of ASTM Standards*. Easton, Md: American Standard.

Attfield, J. G., & Webster, D. A. (1966). "Dichlorvos". *Chemistry and Industry*. Feb. 12, 272-278. (Paper presented to the Pesticides Group of the Society of Chemical Industry in London on December 14, 1964).

Aubry, A., Ghermani, N., & Marraud, M. (1984). Backbone side chain interactions in peptides. *International Journal of Peptide and Protein Research, 23*, 113-122. QP 551 1615.

Ayyangar, N. R., & Tilak, B. D. (1971). Basic dyes. In K. Venkataraman (Ed.), *The Chemistry of Synthetic Dyes Vol. III*. New York: Academic Press.

Ballard, M. W. (1987). Is fumigation possible? *The International Journal of Museum Management and Curatorship, 6*(1), 83-86.

Ballard, M. W. (no date) *Mothproofing Museum Textiles*. Xerox copy.

Bauman, H. (1971). Isolation of dyed proteins from reactive dyed wool. *Applied Polymer Symposium, 18*, 307-315.

Baumann, H., & Setiawan, L. (1985a). New results in sulfur chemisty of wool based on ESCA. In M. Sakamoto (Ed.), Proceedings of the 7th International Wool Textile Research Conference, Vol. I (pp. 108-119). Tokyo, Japan: Nichiei Kosen.

Baumann, H., & Setiawan, L. (1985b). Topochemistry of dyeing and chemical processing of

wool. In M Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Vol. IV* (pp. 1-13). Tokyo, Japan: Nichiei Kosen.

Baumann, H., & Setiawan, L. D., & Gribbin, D. (1986). Surface studies of keratin fibres and related model compounds using ESCA. Intermediate oxidation products of cystyl residue on keratin fibre and surfaces and their hydrolytical stability. *Surface and Interface Analysis, 8*, 219-225.

Bauters, M. (1985). Study of the proteins extracted by formic acid from wool and mohair. In M. Sakamoto (Ed.), Proceedings of the 7th International Wool Textile Research Conference, Vol. I, pp. 162-170.

Bell, J. W., Clegg, D., & Whewell, C. S. (1960) The action of heat on wool *Journal of the Textile Institute, 51*, T1173-T1182.

Beloin, N. J. (1973). Fading of dyed fabrics exposed to air pollutants. *Textile Chemist and Colourist, 5*(7), 129-134.

Bendit, E. G. (1960). The ultraviolet absorption spectrum of solid keratin. *Journal of the Textile Institute Transactions, 51*, T544-T561.

Bendit, E. G. (1975). Structure and physics of wool. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. I* (pp. 423-438). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Bendit, E. G. (1975). The fine structure of keratin by X-ray and electron microscope studies. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. I* (pp. 487-492). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Benisek, L., Edmondson G. K., & Myers, P. C. (1985). Protection of wool against phototendering, photo-and thermal-yellowing. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference: Vol. I*, (pp. 79-89). Tokyo, Japan: Nichiei Kosen.

Bird, C. L. (1972). *The Theory and Practice of Wool Dyeing*. Leeds: Chorley and Pickersgill.

Blakey, P. R., Guy, R., Happey, F., & Lockwood, P. (1971). The effect of chemical modification on the morphological structure of keratin. *Applied Polymer Symposium, 18*, 193-200.

Blakey, P. R., & Mickelthwaite, B. (1978). Scanning electron microscopy and x-ray micronalysis. In F. Happey (Ed.), *Applied Fibre Science, Vol. 1*. London: Academic Press.

Blakey, P. R. (1979). Fine structure of keratin. In F. Happey (Ed.), *Applied Fibre Science, Vol. 3*. Toronto: Academic Press.

Blakey, O. M., Micklethwaite, B., Alfy, E., & Moses, I. (1985). The electron microscopy of plasma-etched fibres. In M. Sakamoto (Ed.), *Proceeding of the 7th International Wool Textile Research Conference: Vol. I*, (pp. 273- 282). Tokyo, Japan: Nichiei Kosen.

Boczkowski, D. (1971). The effect of natural grease content in wool on fabric wear properties. *Applied Polymer Symposium, 18*, 1143-1146.

Bogle, M. (1979). *Textile Conservation Center Notes*. North Andover, MA : Merrimack Valley Textile Museum.

Bogle, M., & Hueck, H. J. (1979). Technical data on moth-proofing. *Textile Conservation Center Notes, 10*. North Andover, MA: Merimack Valley Textile Museum.

Bradbury, J. H. (1973). The structure and chemistry of keratin fibers. In C. B. Anfinsen, J. T. Edsall, & F. M. Richards (Eds.), *Advances in Protein Chemistry. Vol. 27*. New York: Academic

Bradbury, J. H. (1975). Morphology and chemistry of wool. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. 1* (pp. 439-450). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Bradbury, J. H., Chapman, G. V., King, N. L., & O'Shea, J. M. (1970). Keratin Fibres. Amino acid analyses of histological components. *Australian Journal of Biological Sciences, 23*, 637-643.

Bradbury, J. H., & Leeder, J. D. (1971). The cell membrane complex of wool. *Applied Polymer Symposium, 18*, 227-236.

Bradbury, J. H., & Leeder, J. D (1972). Keratin fibers: Mechanism of the Allworden reaction. *Australian Journal of Biological Sciences, 25* 133-138.

Bradbury, J. H., & Ley, K. F. (1972). The chemical composition of wool: Separation and analysis of exocuticle and endocuticle. *Australian Journal of Biological Sciences, 25* 1235-1247.

Breuer (1975). Thermoelasticity of human hair. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. 1* (pp. 510-522). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Brooks, J. H., & Rahman, M. S. (1986). Surface energy of wool. *Textile Research Journal, 56*, 164-171.

Brooks, T. W., Ashare, E., & Swenson, D. W. (1977). Hollow fibers as controlled vapor release devices. In J. C. Arthur (Ed.), *Textile and Paper Chemistry and Technology: ACS Symposium Series, 49*. Washington, DC: American Chemical Society.

Bueche, F. (1962). *Physical Properties of Polymers*. New York: Wiley & Sons.

Bull, D. L. (1972). Metabolism of organophosphorus insecticides in animals and plants. In F. A. Gunther & J. D. Gunther (Eds.), *Residue Reviews: Vol. 43*. New York: Springer-Verlag.

Bussagli, M. (1980). *Cotton and Silk Making*. New York: Rizzoli.

Butler, G. L. (1977). Algae and pesticides. In F. A. Gunther & J. D. Gunther (Eds.), *Residue Reviews: Vol. 66*. New York: Springer-Verlag.

Byrne, K. M. & Shaw, T. (1981). Moothpoofing of wool with permethrin: Industrial, environmental and toxicological aspects. *Journal of the Society of Dyers and Colourists, 97*, 404-410.

Cacella, A. F., & White, H. J. (1955). Interaction of acid-treated wool and silk with water vapor and water. *Textile Research Journal, 25*, 506-515.

Canadian Centre for Occupational Health & Safety (CCOH&S). (Sept., 1987). *Trade Names Data Base*. CCOHS MSDS Number: 21842. (print-out)

Cardamone, J. M., & Brown, P. (1986). Evaluation of degradation in museum textiles using property kinetics. In H.L. Needles, & S. H. Zeronian (Eds.), *Advances in Chemistry: Vol. 212. Historic Textile and Paper Materials Conservation and Characterization* Washington, DC: American Chemical Society.

Cardamone, J. M., Gould, J. M., & Gordon, S. H. (1987) Characterizing aged textile fibers by fourier transform infrared photacoustic spectrospcopy part I: Comparison of artifical and natural ageing in cotton. *Textile Research Journal, 57*(4), 235-239. *The Textile Research Journal, 56*, 669-6 3.

Carr, C. M., Holt, L. A., & Drennan, J. (1986). Using electron microscopy, x-ray microanalysis to study wool morphology and composition. *Textile Research Journal, 57*, (4) 235-239.

Carr, C. M., Leaver, I. H., & Hughes, A. E. (1986). Detection and analysis by XPS of sulfhydryl groups at the surface of wool fibers. *Textile Research Journal, 56*, 216-217.

Carson, R. (1962). *Silent Spring*. Boston: Riverside Press.

Cavagna, G., Locati, G., Vigliani, E. C. (1969). Clinical effec of Exposure to DDVP (Vapona) insecticide in hospital wards. *Archives of Environmental Health, 19*, 112-123.

Cegarra, J., & Gacen, J. (1971). Optimization of the variables in bleaching wool by the pad-steam method. *Applied Polymer Symposium, 18*, 607-614.

Cegarra, J., & Gacen, J. (1983). The Bleaching of wool by hydrogen peroxide. *Wool Science Review, 59, September, 1983.*

Cegarra, J., Gacen, J., & Caro, M. (1983). The action of sodium laurylsulphaate in the bleaching of wool with hydrogen peroxide in an acidic medium. *Journal of the Textile Institute, 74*, 351-356.

Chapman, R. E. (1975). Electron microscopic and histochemical features of the formation of the orthocortex and paracortex in wool. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. II*, Aachen, (pp. 152-161). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

CHRIS. (June, 1985). *CHRIS: Hazardous Chemical Data*. Washington, DC: U.S. Coastguard.

Christian, G. D. (1971). *Analytical Chemistry*. New York: John Wiley & Sons.

Ciba-Geigy Canada. (1985). *Health and Safety Data Sheet*.

Coderch, M. L., Erra, P., Julia, M. R., Infante, M. R., & Garcia, J. (1985). Analysis of some morphological components extracted from wool subitted to shrink-resist treatments. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Tokyo, Vol. IV*, (pp. 252261). Tokyo, Japan: Nichiei Kosen.

Cook, J. R., & Fleishchfresser, B. E. (1984). Sorption of water-soluble anionic polymers by wool. *Textile Research Journal, 54*, 441-446.

Coughtrey, P. J., Martin, M. H., & Unsworth, M. H. (1987). *Pollutant Transport and Fate in Ecosystems*. Palo Atto: Blackwell Scientific.

Crawshaw, G. H. (1978). The role of wool carpets in controlling indoor air pollution. *The Textile Institute and Industry, 16*, 12-18.

Crewther, W. G. (1975). Primary structure and chemical properties of wool. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. I*, (pp. 1-102). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Crewther, W. G., & Dowling, L. M. (1959). Kinetic study of the supercontraction of wool fibers in solutions of salts, acids, and alkalies. *Textile Research Journal, 29*, 541-549.

Crewther, W. G., & Dowling, L. M. (1971). The preparation and properties of large peptides from the helical regions of the low-sulfur proteins of wool. *Journal of Applied Polymer Science: Applied Polymer Symposium, 18*, 1-20

Crewther, W. G., Dowling, L. M., Inglis, A. S., Sparrow, L. G., Strike, P. m., & Woods, E. F. (1985). Wool microfibrils and intermediate filaments have common structural features. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Tokyo, Vol. I*, (pp. 95-104). Tokyo, Japan: Nichiei Kosen.

Crewther, W. G., Fraser, R. D., Lennox, F. G., & Lindley, H. (1965). The chemistry of keratins. In C. B. Anfinsen, M. L. Anson, J. T. Edsall & F. M. Richards (Eds.), in *Advances in Protein Chemistry*. London: Academic Press.

Danilatos, G. D., & Brooks, J. H. (1985). Environmental SEM in wool research-present state of the art. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference: Vol. 1*. (pp 263-272). Tokyo: Nichiei Kosen.

Davies, A. G. (1961). *Organic Peroxides*. London: Butterworths.

de Bruijn, W. C., Koerten, H. K., Cleton-Soeteman, M. I., & Blok-van Hoek, C. J. (1987). Image analysis and x-ray microanalysis in cytochemistry. *Scanning Microscopy, 1*, (4), 1651-1667.

de Graaf, A. J. (1980). In F. Pertegamo (Ed.), *Conservation and Restoration International Conference, Como, 1980*. (pp. 54-61). Milan:ICOMO (Xerox copy).

de Jong, S., & Curiskis, J. I. (1985). The mechanical properties of wool as an engineering composite. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Tokyo, Vol. I*, (pp. 204-213). Tokyo, Japan: Nichiei Kosen.

Denton, M. J. (1982). The crimp curvature of bicomponent fibers. *Journal of the Textile*

Institute, 73, 253-263.

Desmarchelier, J. M., Wustner, D. A., & Fukuto, T. R. (1976). In F. A. Gunther & J. D. Gunther (Eds.), *Residue Reviews, Vol. 63.* (76-192). New York: Springer-Verlag.

Dickerson, R. E., & Geis, I. (1969). *The Structure and Action of Proteins.* New York: Harper & Row.

Dobb, M. G. (1966). Electron microscopy of wool keratin. *Wool Science Review, 28,* 1-11.

Dobb, M.G., & Murray, R. (1975). Aspects of keratin structure. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. II* (pp. 1-11). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Dobb, M. G., & Rogers, G. E. (1969). Electron microscopy of fibrous keratins. In W. G. Crewther (Ed.), *Symposium of Fibrous Proteins.* (pp. 267-278). Australia: Butterworths

Dobb, M. G., & Sikorski, J. (1971). Structure of alpha-keratin: High-resolution electron microscope and electron diffraction studies. *Applied Polymer Symposium 18,* 743-756.

Durham, W. F., Wolfe, H. R., & Elliott, J. E. (1972). Absorption and excretion of parathion sprayman. *Archives of Environmental Health, 24,* 381-387.

Dorough, H. W. (1976). Biological activity of pesticide conjugates. In D. D. Kaufman, G. G. Still, G. D. Paulson & S. K. Bandal (Eds.), *Bound and Conjugated Pesticide Residues: ACS Symposium Series 29.* (11-34). Washington, DC: American Chemical Society.

Earland, C. (1963). *Wool: Its Chemistry and Physics.* London: Chapman & Hall.

Earland, C., Alexander, P., & Hudson, R. F. (1963). *Wool Its Chemistry and Physics.* London: Chapman & Hill.

Earland, C., & Little, A. S. (1985). The bleaching of naturally pigmented cashmere with hydrogen peroxide. In M. Sakamoto (Ed.) *Proceedings of the 7th International Wool Textile Research Conference Tokyo, Vol. IV,* (pp. 138-140). Tokyo, Japan: Nichiei Kosen.

Earland, C., Stell, J. G., & Wiseman, A. (1960). The oxidative insolubilization of proteins. *Journal of the Textile Institute Transactions, 51,* T817-T840. London: Chapman & Hall.

Earle, R. H., Saunders, R. H., & Kangas, L. R. (1971). The chemistry of the chlorination/Hercosett resin shrinkproofing process. *Applied Polymer Symposium, 18,* 707-714.

Ebert, C. (1971). The chemical modification of reduced keratin. *Applied Polymer Symposium, 18,* 221-225.

Ebert, C., Ebert, G., & Knipp, H. (1977). On the introduction of disulfide crosslinks into fibrous proteins and bovine serum albumin. In M. Friedman (Ed.), *Advances in Experimental Medicine and Biology: Vol. 86A. Protein Crosslinking Biochemical and Molecular Aspects.* New York: Plenum. (silk serine OH reactions)

Ebert, G., & Zander, R. (1985). Solvation and structural stability of keratin fibers. In M Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Tokyo, Vol. I*, (pp. 293-302). Tokyo, Japan: Nichiei Kosen

Edwards, J. O. *Peroxide Reaction Mechanisms*. New York: Interscience.

Edwards, S. R., Bell, B. M., & King, M. E. (1981). *Pest Control in Museums. A Status Report (1980)* USA: ASC.

Elliott, A. (1952). The alpha-beta transformation in stretched hair. *Textile Research Journal, 22*, 783-786.

Eto, M. (1974). *Organophosphorous Pesticides: Organic and Biological Chemistry*. Cleveland: CRC.

Evans, D. J., Leeder, J. D., Rippon, J. A., & Rivett, D. E. (1985). Separation and analysis of the surface lipids of the wool fiber. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Vol. I*, (pp. 135-142). Tokyo, Japan: Nichiei Kosen.

Fair, N. B., & Gupta, B. S. (1985). The combined effect of chlorination and hydrogen peroxide bleaching on the surface properties of human hair. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Tokyo, Vol. IV*, (pp.163-172). Tokyo, Japan: Nichiei Kosen.

Fall, F. K. (1973). *Art Objects: Their Care and Preservation A Handbook for Museums and Collectors*. La Jolla: Laurence McGilvery.

Farnworth, A. (1964). The fine structure of wool keratin as revealed by X-ray diffraction. *Wool Science Review, 25*, 1-16.

Fatou, J. G. (1978). Optical microscopy of fibers. In F Happy (Ed.), *Applied Fibre Science, Vol. 1*, 69-155. London: Academic.

Feughelman, M. (1971). The relationship between structure and the mechanical properties of keratin fibers. *Applied Polymer Symposium, 18*, 757-774.

Feldtman, H. D., & Leeder, J. D. (1984). Effects of polar solvents on the abrasion resistance of wool fabric. *Textile Research Journal, 54*, 26-31.

Feldtman, H. D., & Leeder, J. D. (1985). Counteracting damage during processing. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Tokyo, Vol. IV*, (pp. 471-480). Tokyo, Japan: Nichiei Kosen.

Feughelman, M. (1960). The mechanical properties of set wool fibres and the structure of keratin. *The Journal of the Textile Institute, 51*, T589-T602.

Feughelman, M. (1978). The mechanical properties of fibres and fibre thermodynamics. In F. Happey (Ed.), Applied Fibre Science, Vol. 1. London: Academic Press.

Feughelman, M. (1980). Basic wool physics. *Proceedings of the 6th Quinquennial International Wool Textile Research Conference, Pretoria*, (pp. 35-59). Pretoria, South Africa: Heer.

Feughelman, M., & Reis, P. J. (1967). The longitudinal mechanical properties of wool fibers and their relationshop to the low sulfur keration fraction. *Textile Research Journal, 37*,(4), 334-336.

Feughelman, M., & Robinson, M. S. (1967). The relationship between some mechanical properties of single wool fibers and relative humidity. *Textile Research Journal, 37*,(6), 441-446.

Fiori, C. E., Myklebust, R. L., Newbury, D. E. (1979). A catalogue of artifacts observed in energy-dispersive x-ray spectrometry and their influence on analysis. In C. Lechene & R. Warner (Eds.), *Microbeam Analysis in Biology*. Toronto: Academic Press.

Fleischfresser, B. E., & Smith, G. C. (1988). Sorption of carboxylic acids by wool: Effect of solvents. *Textile Research Journal, 58*, 221-226.

Fletcher, J. C., & Buchanan, J. H. (1977). The basis of protein chemistry. In R. S. Asquith *Chemistry of Natural Protein Fibers*. New York: Plenum.

Florian, M. E. (1986). The freezing process-effects on insects and artifact materials. *Leather Conservation News 3*, 1-13.

Forslind, B., Li, H. K., Malmqvist, K. G., & Wiegleb, D. (1986). Elemental content of anagen hairs in a normal caucasion population studies with proton induced x-ray emission (PIXE). *Scanning Electron Microscopy, 1986, I*, (pp. 237-241).

Fraser, R. D., Jones, L. N., MacRae, T. P., Rowlands, R. J., & Tulloch, P. A. (1975). Studies on microfibrils isolated from alpha-keratin. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. II*, Aachen, (pp. 119-130). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Fraser, R. D., Jones, L. N., Macrae, T. P., Suzuki, E., & Tulloch, P. A. (1980). The fine structure of wool fibre. *Proceedings of the 6th Quinquennial International Wool Textile Research Conference, Pretoria, Vol. I* (pp. 1-34). Pretoria, South Africa: Heer.

Fraser, R. D., MacRae, T. P., & Rogers, G. E. (1960). Recent observations on the structure of alpha-keratin. Second Quinquennnial Wool Textile Research Conference, Harrogate. *Journal of the Textile Institute Transactions, 51*, T497-T505.

Fraser, R. D., MacRae, T. P., & Rogers, G. E. (1972). *Keratins: Their Composition, Structure and Biosynthesis*. Springfield: Charles C. Thomas.

Fraser, R. D., MacRae, T. P. (1973). *Conformation in Fiberous Proteins*. New York: Academic Press.

Fraser, R. D., MacRae, T. P., Rowlands, R. J., & Tulloch, P. A. (1975). Molecular structure of keratin. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. II*, Aachen, (pp. 80-89). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Fraser, R. D., & Rogers, G. E. (1955). The structure of resistant membranes isolated from oxidized wool. *Textile Research Journal, 25*, 235-241.

Fraser, I. E., & Truter, E. V. (1960). The constitution of suint. Part III, the coloring matter. *Journal of the Textile Institute Transaction, 51*, T863-T873.

Frear, D. S. (1976). Pestide conjugates-glycosides. In D. D. Kaufman, G. G. Still, G. D. Paulson, & S. K. Bandal (Eds.), *Bound and Conjugated Pesticide Residues*. (35-54). Washington, DC: American Chemical Society.

Freed, V. H. (1980). The university chemist-role in transport and fate of chemical problems. In R. Hague (Ed), *Dynamics, Exposure and Hazard Assessment of Toxic Chemicals*. Ann Arbor: Ann Arbor Science.

Fuchtenbusch, D., & Baumann, H. (1985). Systematical treatment of wool with amines. in M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Tokyo, Vol. IV*, (pp. 411-430). Tokyo, Japan: Nichiei Kosen.

Funder, J. V. (1972). Testing of insecticides in the laboratory. *Danish Pest Infestation Laboratory Annual Report, 1971*. Denmark: xerox copy.

Garner, W. (1966). *Textile Laboratory Manual, Vol. 1: Qualitative Methods*. London: Heywood Brooks.

Garner, W. (1967). *Textile Laboratory Manual, Vol. 5: Fibres*. London: Heywood Books.

Garner, W. (1967). *Textile Laboratory Manual, Vol. 6: Additional Methods*. London: Heywood Brooks.

Getty Conservation Institute. (1986). Effects of fumigants on Museum Collections. *The Getty Conservation Institute Newsletter, 3*, (p 8).

Gillespie, J. M., & Frenkel, M. J. (1973). The macroheterogeniety of type I tyrosine-rich proteins of Merino wool. *The Australian Journal of Biological Science, 27*, 617-627.

Gillespie, J. M., & Frenkel, M. J. (1975). The tryrosine-rich proteins of keratins: occurence, properties and regulation of biosynthesis. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. II*, Aachen, (pp. 265-276). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Gillet, J. W., Harr, J. R., Lindstrom, F. T., Mount, D. A., St. Clair, A. D., & Weber, L. J. (1972). Evaluation of human health hazards on use of dichlorvos (DDVP), especially in resin strips. In F. A. Gunther, & J. D. Gunther (Eds.), *Residue Reviews: Vol. 44*, (pp. 115-160). New York: Springer.

Gillet, J. W., Harr, J. R., St. Clair, A. D., & Weber, L. J. (1972). Comment on the distinction between hazard and safety in evaluation of human health hazards on use of dichlorvos, especially resin strips. In F. A. Gunther, & J. D. Gunther (Ed.), *Residue Reviews: Vol. 44*, (pp. 161-184). New York: Springer.

Glastrup, J. (1987). Insecticide analysis by gas chromatography in the stores of the Danish National Museum's ethnographic collection. *Studies in Conservation, 32*, 59-64.

Gosselin, R. (1984). *Clinical Toxicology of Commercial Products*. 5th Ed. Baltimore: Williams & Wilkins.

Grayson, M. (1978). Insect control technology. *Encyclopedia of Chemical Technology (Toxicology)*. 3rd Ed. New York: Wiley.

Gregory, C. J. (1968). Absorption of atmospheric sulfur-dioxide by natural and synthetic textile fibers (Doctoral dissertation, Rutgers University, 1968). *University Microfilms International, 68*, 17775.

Greven, R., Kaplin, I. J., & Zahn, H. (1986) Morphological origin of the S-Carboxymethyl kerateines of wool. *Textile Research Journal, 56*,. 523-526.

Griffith, J. C. (1967). The modification of wool fabric by certain organic sorbates. Part I: Factors relating to their equilibrium adsorption and the subsequent water uptake of the fabric. *Textile Research Journal, 37*,(3), 173-195. (check last page)

Gunther, F. A., Iwata, Y., Carman, G. E., & Smith, C. A. (1977). The citrus reentry problem: Research on its causes and effects, and approaches to its minimization. In F. A. Gunther & J. D. Gunther (Eds.), *Pesticide Reviews: Vol. 67. Residues of Pesticides and Other Contaminants in the Total Environment*. New York: Springer-Verlag.

Gurr, E., & Anand, N., Unni, M. K., & Ayyangar, N. R. (1974). Applications of synthetic dyes to biological problems. In K. Venkataraman (Ed.), *The Chemistry of Synthetic Dyes Vol. III*. San Francisco: Academic Press.

Haines, B. M. (1985). Hides and skins. In A. H. Rose (Ed), *Microbial Biodeterioration Economic Microbiology,6* (pp. 130-147). Academic Press.

Haly, A. R., & Feughelman, M. (1960). Stress changes at constant strain and hydrogen bonding in keratin fibres. *Journal of the Textile Institute Transactions, 51*, T573-T588.

Hanlan, J. (1975). The scanning electron microscope and microprobe: Applications to conservation and historical research. *ICCOM Committee for Conservation 4th Triennial Meeting, Venice*. Reprint.

Happey, F. (1978). General introduction. In F. Happey (Ed.), *Applied Fibre Science: Vol I*. London: Academic Press.

Happey, F. (1975). Wool research - the premise of molecular biology. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. II*, Aachen, (pp. 223-232). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Haque, R. (1980). *Dynamics, Exposure and Hazard Assessment of Toxic Chemicals*. Ann Arbor: Ann Arbor Science.

Harper, H. (1931). *Introduction to Textile Chemistry*. London: MacMillan.

Harrington, J., & Petherbridge, G. (1980-81). Chemical hazards and toxicology. In G. Petherbridge, & J. Harrington (Eds.), *Safety and Health in the Paper Conservation Laboratory: Vol. 5 & 6. The Paper Conservator Journal of the Institute of Paper Conservator*. Worthing: L & S.

Harris, M., & Smith, A. (1937). State of sulfur in oxidized wool. *Journal of Research of the National Bureau of Standards, 18*, (623-638).

Harris, M. (1954). *Handbook of Textile Fibers*. Washington, DC: Waverly.

Hartsuch, B. E. (1950). *Introduction to Textile Chemistry*. New York: Wiley & Sons.

Hattori, J., Oizumi, K., Sato, Y. Tsuda, K., Abe, T., & Harada, M. (1976). Biological properties of Sumithion. In F. A. Gunther, & J. D. Gunther (Eds.), *Residue Reviews: Vol. 60. Residue of Pesticides and Other Contaminants in the Total Environment* (pp. 39-101). New York: Springer-Verlag.

Hayat, M. A. (1974). *Principles and Techniques of Scanning Electron Microscopy, Vol. 2*. New York: Van Nostrand Reinhold.

Hearle, J. W. (1963). The development of ideas of fine structure. In J. W. Hearle & R. H. Peters (Eds.), *Fibre Structure*. (pp. 209-232). London: Butterworths.

Hearle, J. W. (1963). The fine structure of fibers and crystalline polymers. III. Interpretation of the mechanical properties of fibers. *Journal of Applied Polymer Science, 7*, 1207-1223.

Hearle, J. W., Chapman, B. M., & Senior, G. S. (1971). The interpretation of the mechanical properties of wool. *Applied Polymer Symposium, 18*, 775-794.

Herrling, J., & Zahn, H. (1985). Investigations on the competition of the cell membrane complex and its modification during industrial processing of wool. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Tokyo, Vol. I*, (pp. 181-193). Tokyo, Japan: Nichiei Kosen.

Hess, F. D. (1980). Influence of specimen topography on microanalysis. In M. A. Hayat (Ed.), *X-ray Microanalysis in Biology*. (pp. 241-261) Baltimore: University Park Press.

Hewish, D. R., Fraser, R., Frenkel, M. J, & Jones, I. N. (1983-84). *CSIRO Divsion of Protein Chemistry, Institute of Industrial Technology, Annual Report 1983-84*. Melbourne, Aust: CSIRO.

Hewish, D. R., Robinson, C., & Sparrow, L. (1984). Monoclonal antibody studies of a-keratin low-sulfur proteins. *Australian Journal of Biological Sciences, 37*, 17-23

Hewish, D. R., & French, P (1986). Monoclonal antibodies to a subfraction of Merino high-tryosine proteins. *Australian Journal of Biological Sciences, 39*, 341-351.

Hey, M. (1980-81). Paper conservation: Processes nazardous to Health. *The Paper Conservator, 5 & 6*, 5-10.

Higgerson, G. J., & Whiteley, K. J. (1985). Measurement of the crimp characteristics of greasy wool staples. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Tokyo, Vol. II*, (pp. 157-166). Tokyo, Japan: Nichiei Kosen.

Hojo, H. (1985). Improvement of wool fibers by removing exocuticle in the presence of metallic ions. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Tokyo, I*, (pp. 322-331). Tokyo, Japan: Nichiei Kosen.

Holt, L. A. (1984). Sorption of perchlorethylene and toluene by wool and other textiles:

Influence of co-solvents. *Textile Research Journal, 54*, 226-230.

Holt, L. A. (1985). Wool treatments from "non-swelling" solvents. In M. Sakamoto (Ed.), *Proceedings for the 7th International Wool Textile Research Conference, Vol. IV*. (pp. 89-98). Tokyo, Japan: Nichiei Kosen.

Holt, L. A., & Milligan, B. (1970). The esterification of wool. *Australian Journal of Biological Sciences, 23*, 165-173.

Holt, L. A., Milligan, B., & Rivett, D. E. (1975). The role of tryptophan in the photochemical crosslinking of peptides and proteins. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. I* (pp. 559-569). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Howitt, F. O. (1962). Wool keratin: Its chemical structure and reactions, Part 1. *Wool Science Review, 21*, 27-39.

Howitt, F. O. (1963). Wool keratin: Its chemical structure and reactions, Part 2. *Wool Science Review, 22*, 3-14.

Howitt, F. O. (1963). Wool keratin: Its chemical structure and reactions, Part 3. *Wool Science Review, 23*, 23-39.

Howitt, F. O. (1964). Wool keratin: Its chemical structure and reactions, Part 4. Wool Science Review, 24, 16-33.

Hueck, H. J. (1972). Textile pests and their control. In J. Leene (Ed.), *Textile Conservation*. Washington, DC: Smithsonian.

Huggins, M. L. (1975). Principles of the packing of helical molecules, such as keratin. K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. II* (pp. 109-116). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Hunter, L., & Gee, E. (1980). The effect of staple crimp, resistance to compression and fibre diameter and length characteristics on the physical properties of wool wortsted yarns. In *Proceedings of the 6th Quindquennial International Wool Textile Research Conference, Pretoria, 26 August3 September, 1980, Vol. I* (pp. 327-337). Pretoria, S.A:SAWTRI.

Hunter Assoc. (nd). *Hunterlab Tristimulus Colorimeter Instruction Mannual*. Reston, Va: Hunter Assoc. Laboratory Inc.

Inglis, A. S., & Lennox, F. G. (1965). Studies in wool yellowing. Part IX: Irradiation with different UV wavebands. *Textile Research Journal, 35*, 104-109.

Inglis, A. S., & Leaver, I. H. (1967). Some effects of peroxide oxidation of wool. *Textile Research Journal, 37*,(11), 995-997.

Ito, H., Sakabe, H., Miyamoto, T., & Ingaki, H. (1984). Fibrillation of the cortex of Merino wool fibers by freezing-thawing treatment. *Textile Research Journal, 54*, 397-402.

Ito, H., Sakabe, H., Miyamoto, T., & Inagaki, H. (1985). Isolation and characterization of the

orthocortical and paracortical cells from merino wool fibers. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Vol. I*, (pp 115-124). Tokyo, Japan: Nichiei Kosen.

Ivanov, V. B. (1987). *Reactive Dyes in Biology*. Chur, Switzerland: Harwood Academic.

Jaksic, D., & Jovanovic, R. S. (1985). Background of the X-ray methods for the determination of the crystalline fraction. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference: Vol. 1*. (pp. 253-262). Tokyo, Japan: Nichiei Japan.

Jeyaraj, V. (1983). Ethological collections, conservation problems and their solution in Governement Museum, Madras. *Journal of Indian Museums, 39*, 123-128.

Johnson, D. J., & Walton, C. J. (1975). Theoretical and experimental studies of meridional reflections in the X-ray diffraction patterns of alpha-keratin. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. II*, Aachen, (pp. 69-79). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Joko, K., Koga, J., & Kuroki, N. (1985). The interaction of dyes with wool keratin - The effect of solvent treatment of dyeing behaviour. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference: Vol. 5*, (pp. 23-28). Tokyo, Japan: Nichiei Japan.

Jones, F. W. (1983). Fibre-reactive organophosphorus esters suitable for insectproofing of wool. *Journal of the Society of Dyers and Colourists, 99*, 46-51.

Jones, F. W. (1983). The long-term stability of two fibre-reactive organophosphorus insecticides on wool. *Journal of the Society of Dyers and Colourists, 99*, 235.

Kaemmerer, K., & Buntenkotter, S. (1973). The problem of residues in meat of edible domestic animals after application or intake of organophosphate esters. In F. A Gunther, & J. D. Gunther (Ed), *Residue Reviews: Vol 46*. New York: Springer-Verlap.

Nakamura, Y., Kanoh, T., Kondo, T., & Inagaki, H. (1975). Some morphological and histochemical aspects on wool cuticle. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. II* (pp. 23-43). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Kantouch, A., & Abdel Fattah, S. H. (1971). Oxidation of wool with chlorine and some chlorinated compounds. *Applied Polymer Symposium, 18*, 317-323.

Kaplin, I. J., & Whiteley, K. J. (1985). The structure of keratin macrofibrils part 1: Keratins of high sulfur content. In M. Sakamoto (Ed.). *Proceedings of the 7th International Wool Textile Research Conference Vol. 1* . (pp. 95-104) Tokyo: Nichiei Kosen.

Katz, H. L. (1982). Fabric pests are back! *IPM Practioner: The Newsletter of Integrated Pest Management*.

Kaufman, D. D. (1976). Bound and conjugated pesticide residues. In D.D. Kaufman, G. G. Still, G. D. Paulson, & S. Bandal (Eds.), Bound and Conjugated Pesticide Residues. (pp. 1-10) ACS Symposium Series 29. Washington, DC: American Chemical Society.

Kawar, N. S., Bastita, G. C., & Gunther, F. A. (1973). Pesticide stability in cold-stored plant parts, soils, and dairy products, and in cold-stored extractives solutions. In F. A Gunther & J. D. Gunther (Eds.), *Residue Reviews: Vol. 48.* (pp 45-77). New York: Springer-Verlap.

Kearns, J. E., Maclaren, J. A., & Roxburgh, C. M. (1977). The use of oxidative hyrdolysis for estimating the extent of carbon-sulphur fission in modified wools. *Journal of the Textile Institute, 9,* 283-288.

Kidd, F. (1977). Other animal fibers. In R. S. Asquith (Ed.), *Chemistry of Natural Protein Fibers.* New York: Plenum.

King, N. L., & Wood, R. (1965). Temperature control of water-vapour sorption by keratin. *Journal of the Textile Institute, 56,* T215-T216.

King, N. L., & Bradbury, J. H. (1968). The chemical composition of wool, V. The epicuticle. *Australian Journal of Biological Sciences, 21,* 375-384.

Knott, J., & Zahn, H. (1971). Alkali solubility of carbonized wool as a function of storage time and acid content. *Applied Polymer Symposium, 18,* 601-605.

Koenig, N. H. &, Friedman, M. (1977). Comparison of wool reactions with selected mono- and bifunctional reagents. In M. Friedman (Ed.), *Advances in Experimental Medicine and Biology: Vol. 86A. Protein Crosslinking Biochemical and Molecular Aspects.* New York: Plenum.

Kondo, T., Tagawa, T., & Horio, M. (1971). Structure and properties of mohair in comparison with wool. *Applied Polymer Symposium, 18,* 879-886.

Kopke, V., & Nordby, H. A. (1971). The extension and recovery behaviour of single wool fibers under variable humidity conditions. *Applied Polymer Symposium, 18,* 935-946.

Kulkarni, V. G., & Bradbury, J. H. (1974). The chemical composition of wool. XII Further studies on cortical cells and macrofibrils. *Australian Journal of Biological Sciences, 27,* 383-396.

Kulkarni, V. G., & Bradbury, J. H. (1975). The chemical composition of wool part XIV: The microfibrils. K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. II* (pp. 139-147). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Kulkarni, V. G., Robson, R. M., & Robson, A. (1971). Studies on the orthocortex and paracortex of Merino wool. *Applied Polymer Symposium, 18,* 127-146.

Larose, P. (1955). The sorption of HCl by Textile fibers. *Textile Research Journal, 25,* 956-961.

Laughlin, J. M., & Gold, R. E. (1987). The vaporization of methyl parathion from contaminated cotton fabrics. *Textile Chemist and Colorist, 19*(5), 39-42.

Launer, H. F., & Black, D. (1971). Gases produced from wool by light and heat. Applied Polymer Symposium, 18, 347-352.

Lave, L. B., & Upton, A. C. (1987). *Toxic Chemicals, Health, and the Environment.* Baltimore: Johns Hopkins University Press.

Leaver, I. H., Marshall, R. C., & Rivett, D. E. (1985). Light-induced changes in the composition of wool and lysozyme. In M. Sakamoto (Ed.), *Proceedings of the 7th International Textile Research Conference Tokyo, IV,* (pg 11-20). Tokyo, Japan: Nichiei Kosen.

Leeder, J. D. (1986) The cell membrance complex and its influence on the properties of the wool fibre. *Wool Science Review: 63.* (entire issue)

Leeder, J. D., & Lipson, M. (1963). Sorption of liquids by wool. Part IV. Sorption of ethanol by modified wool. *Journal of Applied Polymer Science, 7,* 2053-2066.

Leeder, J. D., & Rippon, J. A. (1983). Some observations on the dyeing of wool from aqueous formic acid. *Journal of the Society of Dyers and Colourists, 99,* 64-65.

Leeder, J. D., & Rippon, J. A. (1981). Some observations on the dyeing of wool from aqueous formic acid. *Society of Dyers and Colourists Journal, 97,* 43-44.

Leeder, J. D., Rippon, J. A, & Rivett, D. E. (1985). Modification of the surface properties of wool by treatment with anhyrdrous alkali. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Tokyo: Vol. IV,* 312-321. Tokyo, Japan: Nichiei Kosen.

Leeder, J. D., Rippon, J. A., Rothery, F. E., & Stapleton, I. W., (1985). Use of the ransmission electron microscope to study dyeing and diffusion processes. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Tokyo: Vol. V,* (pp. 99-108). Tokyo, Japan: Nichiei Kosen.

Lees, K., Peryman, R. V., & Elsworth, F. F. (1960). The solubility of wool in urea-bisuphite solutions and its use as a measure of fibre modification-Part III. *Journal of the Textile Institute, 51,* T717-T725.

Lellinger, D. B. (1972). Dichlorvos and lindane as herbarium insecticides. *Taxon, 21,* 91-95.

Lennox, F. G. (1960). A spectrophotometric study of yellowing in wool fabric. *Journal of the Textile Institute Transactions, 51,* T1193-T1213.

Lennox, F. G., King, M.G., Leaver, I. H., Ramsay, G. C., & Savige, W. E. (1971). Mechanisms, prevention, and correction of wool photo-yellowing. *Applied Polymer Symposium, 18,* 353-369.

Levin, E. R. (1986). Principles of x-ray spectroscopy. *Scanning Electron Microscopy, 1986, 1,* (pp. 65-76)

Levin, R. (1983). Colour Measurements on raw, scoured, and bleached wool. *Textile Research Journal, 74,* 223-225.

Ley, K. F., Marshall, R. C., & Crewther, W. G. (1985). Further studies on the release and characterisation of cuticle and membranous components from wool. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference: Vol. 1,*

(pp. 152-170). Tokyo, Japan: Nichiei Kosen.

Light, A. (1974). *Proteins Structure and Function*. Englewood Cliffs, NJ: Prentice-Hall.

Lindley, H. (1959). The varying reactivity of the cystine of wool. In R. Bensesh (Ed.) *Sulfur in Proteins*. New York: Academic Press.

Lindley, H. (1977). The chemical composition and structure of wool. In R. S. Asquith (Ed.), *Chemistry of Natural Protein Fibers*. New York: Plenum.

Lippmann, M. (1987). Toxic chemical exposure and dose in target tissue. In L. Lave & A Upton (Eds.), *Toxic Chemicals, Health, and the Environment*. (pp. 114-141). Baltimore: Johns Hopkins University Press.

Lipson, M. (1975). Chemical technology of wool processing. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. 1*. (pp. 210-247). Aachen, Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Lorminer, J. W. (1979). Osmosis. In F. Happey (Ed.), *Applied Fibre Science, Vol. 2*. London: Academic Press.

Lotz, B., & Brack, A. (1979). The use of polypeptides as models in fibrous protein structure investigations. In F. Happey (Ed.), *Applied Fibre Science, Vol. 3*. Toronto: Academic Press.

Maclaren, J. A., Leach, S. J., & Swan, J. M. (1960). Study of some problems in protein chemistry using new (non-hydrolytic) methods for the determination of thiol and disulphide. *Journal of the Textile Institute Transactions, 51*, T665-T667.

Maclaren, J. A., & Milligan, B. (1981). *Wool Science: The Chemical Reactivity of the Wool Fibre*. CSIRO, Aust: Science Press.

Makinson, R. K. (1967). The swelling of merino-wool fibres by large anions and by water. *Journal of the Textile Institute, 58*, 1-17.

Makinson, R. K. (1971). Mechanisms involved in shrinkproofing by degradative treatments. *Applied Polymer Symposium, 18*, 1083-1096.

Makinson, R. K. (1975). Cuticle and cell membranes. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. 1* (pp. 493-494). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Makinson, R. K. (1979). *Shrinkproofing of Wool*. New York: Marcel Dekker.

Mansour, M. P., Cornell, H. J., & Holt, L. A. (1988). Photoyellowing of wool pretreated with chlorinated solvents. *Textile Research Journal, 58*, 246.

Marshall, R. C., & CSIRO (1983-84). Composition, structure and textile performance. *CSIRO Division of Protein Chemistry, Institute of Industrial Chemistry, Annual Report 1983-84*. Melbourne, Aust: CSIRO.

Marshall, R. C., Gillespie, J. M., McGuirk, B. J., Marler, J. W., Reis, P. J., Rogan, I. M., &

Whiteley, K. J. (1985). Investigation of the relationship of variations in fibre protein composition upon fleece properties and fabric performance. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Tokyo, Vol. II*, (pp. 36-44). Tokyo, Japan: Nichiei Kosen.

Marshall, R. C., Gillespie, J. M., & Gee, E. (1985). The effect of nutritional and physiological manipulation of sheep on the protein composition of wool. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Tokyo, Vol. II*, (pp. 26-34). Tokyo, Japan: Nichiei Kosen.

Markkula, M. (1973). Pesticide regulation in Finnland. In F. A. Gunther & J. D. Gunther (Eds.), *Residue Reviews, Vol. 48*. (117-140) New York: Springer-Verlap.

Mason, P. (1965). Thermal transition in keratin. Part III: Stress and temperature effects in relation to the alpha-beta transition. *Textile Research Journal, 35*, 483-490.

Masschelein-Kleiner, L. (1980). Conservation of very brittle textiles. In F. Pertegato (Ed.), *Conservation and Restoration of Textiles, International Conference, COMO*. Milan: CISST.

Matthew (c. 40 AD [Holy Bible]). In *Good News New Testament in Today's English Version of the Bible*. Toronto: Canadian Bible Society.

Mathieson, A. R., Whewell, C. S. (1964). Acid-base properties of fibers. A polyelectrolyte theory of the combination of fibers with acids and bases. *Journal of Applied Polymer Science, 8*, 2029-2048.

Mathieson, A. R., Whewell, C. S., & Williams, P. E. (1964). Acid-base properties of fibers. *Journal of Applied Polymer Science, 8*, 2009-2027.

Matsumura, F. (1985). *Toxicology of Insecticides*. New York: Plenum Press.

Mattyus, H., Nagy, L. G., & Gajari, J. (1975). Accessibility changes in wool brought about by thermal treatment. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. II*, Aachen, (pp. 468-476). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Mazingue, G., Ponchel, P., & Lubrez, J. P. (1971). The composition and properties of the wool cuticle. *Applied Polymer Symposium, 18*, 209-216.

McCrone, W. C., McCrone, L., & Delly, J. G. (1978). *Polarized Light Microscopy*. Ann Arbor: Ann Arbor Science.

McGaughey, W. H. (1973). Dichlorvos vapor for insect control in a rice mill. *Journal of Economic Entomology, 66*,(5), 1147-1151.

McGriffin, R. F. (1985). A current status report on fumigation in museums and historical agencies. Technical Report No. 4. Nashville: Historic Society.

McKenzie, J. A., & Whitten, M. (1984). Estimation of the relative viabilities of insecticide resistance genotypes of the Australian Sheep Blowfly *Lucilia cuprina*. *Australian Journal of Biological Sciences, 37*, 45-52.

McPhee, J. R. (1971). *The Mothproofing of Wool.* Watford, England: Merrow.

McPhee, J. R. (1975). Chemical technology of wool. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. I*, Aachen, (pp. 467-482). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Meichelbeck, H., & Speil, M. (1975). X-ray investigations of reduced and reduced-extended keratin fibres. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. II*, Aachen, (pp. 119-130). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Mendoza, C. E. (1972). Analysis of pesticides by the thin layer chromatographic-enzyme inhibition technique. In F. A. Gunther, & J. D. Gunther (Eds.), *Residue Reviews: Vol. 43*. New York: Springer-Verlap.

Menefee, E. (1971). Relation of keratin structure to its mechanical behavior. *Applied Polymer Symposium, 18*, 809-821.

Menefee, E. (1977). Physical and chemical consequences of keratin crosslinking, with application to the determination of crosslink density. In M. Friedman (Ed.), *Advances in Experimental Medicine and Biology: Vol. 86A: Protein Crosslinking Biochemical and Molecular Aspects*. New York: Plenum.

Menefee, E., & Yee, G. (1965). Thermally-induced structural changes in wool. *Textile Research Journal, 35*, 801-812.

Mercer, E. H. (1953). The heterogeneity of the keratin fibers. *Textile Research Journal, 23* 388-396.

Mercer, E. H., & Golden, R. L. (1953). A new method of producing the Allworden reaction. *Textile Research Journal, 23* 43-44.

Merck & Co., (1983). *Merck Index*. (see Windholz) Rahway, NJ: Merck & Co.

Merkel, R. S. (1984). A glossary of AATCC standard terminology or how to say what you mean and avoid confusion. *Textile Chemist and Colorist, 16* (5), 109-111.

Merkel, R. S. (1984). In J. W. Weaver (Ed.) *Analytical Methods for a Textile Laboratory.*

Meybeck, A., Galafassi, P. (1971). The effect of hydrocarbon substituents in azo dyes on wool dyeing. Applied Polymer Symposium, 18, 463-472.

Milligan, B. & CSIRO (1983-84). Fibre reactivity and textile processing. *CSIRO Division of Protein Chemistry, Institute of Industrial Technology, Annual Report 1983-84.* Melbourne, Aust: CSIRO.

Milligan, B., & Wolfram, L. J. (1973). The acetylation of wool. *Journal of the Textile Institute, 64*, 170-174.

Miyamoto, T., Sakabe, H., Ito, H., & Inagaki, H. (1985). An aspect for the mechanism of bilateral staining of merino wool fibers by basic dyes. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Vol. I*, (pp.

125-134). Tokyo, Japan: Nichiei Kosen.

Moncrieff, R. W. (1950). *Moothproofing*. London: Billing & Sons, Guildford and Esher.

Morgan, A. J. (1985). *X-ray Microanalysis in Electron Microscopy for Biologists*. Oxford: Alden Press.

Morcos, R. (1986). *Environmental Status Report 1979-1984: Vinyl Chloride Industry*. Ottawa: Environment Canada.

Morgan, A. J. (1980). Preparation of specimens: Changes in chemical integrity. In M Hayat (Ed.), *X-ray Microanalysis in Biology*. Baltimore: University Park. (65-165).

Morton, W. E., Hearle, J. W. (1975). *Physical Properties of Textile Fibers*. London: Heinemann.

Myers, G. H. (1952). *Rugs: Preservation, Display and Storage*. Washington, DC: Textile Museum.

Nakamoto, N. (1984). Insecticidal effect of a fumigation agent (DDVP). *Kiito Kensa Kenkya Hokoku, 38*, 75-83.

Nakamura, S. (1985). Attack of attagenus larvae against silk and chemical fibers (Dermestidae, Coleopera). In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Tokyo, Vol. IV*, (pp. 197-204) Tokyo, Japan: Nichiei Kosen.

Nakamura, Y., Kanoh, T., Kondo, T., & Inagaki, H. (1975). Some morphological and histochemical aspects on wool cuticle. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. II* (pp. 23-43). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Nakamura, Y., Tanaka, T., & Watanabe, A. (1975). Electrokinetic studies on the surface structure of wool fibre. K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. II* (pp. 34-43). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Nishiumi, S. (1971). The effects of scales on wool fiber surface properties and the feltability of man-made fibers. *Applied Polymer Symposium, 18*, 1057-1065.

Nogues, B., Coderch, L., Julia, R., & Parra, P. (1988). New advances in the internal lipid composition of wool. *Textile Research Journal, 58*, 338-342.

Norton, G. P., & Nicholls, C. H. (1960). The effect of heating wool containing alkali. *Journal of the Textile Institute Transactions, 51*, T-1183-T1192.

Nossar, M. S., Chaikin, M., & Datyner, A. (1973). The role of surfactants in reducing acid damage in carbonizing. *Journal of the Textile Institute, 64*, 490-494.

Nossar, M. S., & Chaikin, M. (1975). Carbonising of burry wool for combing. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. IV*, Aachen, (pp. 136-142). Aachen: Deutsches Wollforschungsinstitut and der Technischen

Hochschule.

O'Loughlin, G. J. (1982). Fiber-reactive insect-proofing agents for wool: "Phosphourus esters of 3-(Hydroxymethyl)-4-nitrophenol. *Journal of Agricultural Food Chemistry, 30*(6), 1056-1059.

Onions, W. J. (1962). *Wool an Introduction to its Properties, Varieties, Uses and Production.* New York: Interscience.

Orwin, D. R., & Thomson, R. W. (1975). The electron microscopy of two types of fibre damage and its relationship to some cell components. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. I* (pp. 173-183). Aachen: De ches Wollforschungsinstitut an der Technischen Hochschule.

Orwin, D. F., Woods, J. L., & Gourdie, R. G. (rtical ceil type and wool strength. In M. Sakamoto (Ed.), *Proceedings of the 7t a nal Wool Textile Research Conference, Tokyo, Vol. I*, (pp. 194-203). n: Nichiei Kosen.

Orwin, D. F., Woods, J. L., & Ranford, S. L. (1984 rtical cell types and their distribution in wool. *Australian Journal of Biological Sciences, 37*, 237-55. Fibers. New York: Plenum.

Paulson, G. D. (1976). Sulfate ester conjugates-their synthesis, purification, hydrolyis, and chemical and spectral properties. In D. D. Kaufman, G. G. Still, G. D. Paulson, & S. K. Bandal (Eds.), *Bound and Conjugated Pesticide Residues.* (86-102). Washington, DC: American Chemical Society.

Peltz, P., & Rossol, M. (1983). Safe pest control procedures for museum collections. *Center for Occupational Hazards*, 1-8. New York: Xerox

Perkinson, R. (1975). On conservation pH. *Museum News, 53*(6).

Pesce, A. J., Rosen, C. G., & Pasby, T. L. (1971). *Fluorescence Spectroscopy.* New York: Marcel Dekker.

Peters, R. H. (1975). *Textile Chemistry, Vol. III.* Oxford: Elsevier Scientific.

Peters, D. E., & Bradbury, J. H (1972). The chemical composition of wool: Material digested by trypsin from fibres and cortical cells. *Australian Journal of Biological Sciences, 25*, 1225-1234.

Peters, D E., & Bradbury, J. H. (1976). The chemical composition of wool: The cell membrane complex. *Australian Journal of Biological Sciences, 29*, 43-55.

Ploem, J. S., & Tanke, H. J. (1987). *Introduction to Fluorescence Microscopy.* Oxford: Alden Press.

Plenderleith, H. J., & Werner, A. E. (1971). *The Conservation of Antiquities and Works of Art: Treatment Repair and Restoration.* London: Oxford Press.

Pretty, G. (1976). The ethnographic collections in the South Australian Museum. In C. Pearson & G. L. Pretty (Eds.), *The Proceedings of the National Seminar on the*

Conservation of Cultural Material, Perth 1973. (pp. 107-124). Perth: Western Australian Museum.

Purdy J. (1987). Ciba-Geigy Canada Ltd. Agricultural Division, Chemist. (personal correspondence)

Purshottam, T. L., & Srivastava, R. K. (1984). Effect of high fat diets and toxicity of parathion and dichlorvos. *Archives of Environmental Health, 3*, 425-430.

Ramsden, W. (1938). Coagulation by shearing and freezing. *Nature, 142*, 1120-1121.

Reagan, B. (1982). Eradication of insects from wool. *Journal of the American Institute for Conservation, 21*,(2).

Reagan, B. B., Cheng, A., Perenich, T. A., & Caldwell, J., (Members of IPTC Committee). (1984). Effect of insecticides on the colorfastness of acid and disperse dyes on nylon and polyester. *AATCC, 16*(1), 25-37.

Rice, J. W. (1969). Requirements for bulk storage protection against insect damage. *Textile Museum Journal, 2*,(4), 31-33.

Rivett, D. E., & CSIRO (1983-84). Fibre surface and cell membranes. *CSIRO Division of Protein Chemistry, Institute of Industrial Technology, Annual Report 1983-84.* Melbourne, Aust: CSIRO.

Rivett, D. E., Holt, L. A., Jones, L. N., Ley, K. F., & Stapleton, I. W. (1984-86). Wool fibre surface and cell membranes. *CSIRO Divsion of Protein Chemistry, Institute of Industrial Technology, Biennal Report 1984-86*. Melbourne, Aust: CSIRO.

Robbins, C.R., Scott, G. V., & Barnhurst, J. D. (1968). A study of the causes of variation in the acid dye comging capacity of human hair. *Textile Research Journal, 38*, 1130-1135.

Robson, A. (1975). The chemical reactions of wool. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. I*, Aachen, (pp. 137-166). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Robinson, M. S. (1981/1983). Changes in some mechanical and chemical properties of keratin fibres with time. *Journal of the Textile Institute, 74*, 311. Abstract.

Roper, K., & Finnimore, E. (1985). Chemical structure of chromophores formed during photo-yellowing of wool. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, IV*, (pp. 21-30). Tokyo, Japan: Nichiei Kosen.

Roper, K., Fohles, J., Peters, D., & Zahn, H. (1984). Morphological composition of the cuticle from chemically treated wool. Part 1: Calculating endocuticle content in isolated cuticle from the results of amino acid analysis. *Textile Research Journal, 54*, 139-143.

Roper, K., Fohles, J., Peters, D., & Zahn, H. (1984). Morphological composition of the cuticle from chemically treated wool. Part 2: The role of the cuticle in industrial shrinkproofing processes. *Textile Research Journal, 54*, 262-269.

Roger, R. E., & Hays, M. (1943). Effect of storage on fabrics. *Textile Research Journal, 13*,6,

20-35.

Rosenburg, H. G. (1984). The home is the workplace. In W. Chavkin (Ed.), *Double Exposure: Women's Health Hazards on the Job and at Home*. New York: Monthly Review Press.

Ross, D. A., Carnaby, G. A., Lappage, J. (1986). Woollen-yarn manufacture. *The Textile Institute, 15(1/2)*.

Rottenbury, R. A., (1979). Staple strength: Appraisal and measurement. *Proceedings of a Seminar on Staple Length & Staple Strength of Greasy Wool*. Ryde, NSW, Australia: CSIRO.

Rowlands, D. G. (1975). The metabolism of contact insecticides in stored grains. III. 1970-1974. In F. A. Gunther & J. D. Gunther (Eds.), *Residue Reviews, Vol: 58*. (113-156). New York: Springer-Verlap.

Ryckman, R. E. (1969). Vapona for the control of museum pests. *Journal of Medical Entomology, 6*. 98.

Sakamoto, M. (1985). Japanese contributions in the science of wool and silk. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Vol. I* (pp. 1-26). Tokyo, Japan: Nichiei Kosen.

Sakamoto, M. (1985). Gas chromatographic and mass spectrometric study on the reaction silk and wool fibers with various epoxides. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Vol. IV*, (pp. 461-470). Tokyo, Japan: Nichiei Kosen.

Sakamoto, M., Kohara, N., & Nam, S. W.(1985). Setting Setting properties of disulfide-enriched wool. In M. Sakamoto (Ed.), *Proceedings for the 7th International Wool Textile Research Conference, Vol. IV*, (pp. 382-391). Tokyo, Japan: Nichiei Kosen.

Santodonato, J., Bosch, S., Meylan, W., Becker, J., & Neal, M. (1985). Monograph on human exposure to chemicals in the workplace: Dichlorvos. Prepared for *Division of Cancer Etiology National Cancer Institute Bethesda, MD*. Syracuse, NY: Center for Chemical Hazard Assessment.

Savige, W. E. (1975). Ny oxidation and photo-oxidation products of tryptophan. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. II*, Aachen, (pp.570-580). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Schefer, W. (1978). Nachweis und beurtellung chemischer schaden an wolfe. *Textilveredlung, 13*, 55-57.

Schumacher-Hamedat, U., Fohles, J., & Zahn, H. (1985). Oxidation intermediates of cystine during bleaching processes. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Vol. IV*, (pp. 120-129). Tokyo, Japan: Nichiei Kosen.

Schutz, R. A., & Blin, J. R. (1971). Contribution to the study of wool during drying. *Applied Polymer Symposium, 18*, 1131-1141.

Shaw, T. (1985). Thermal expansion of woven wool fabrics at constant moisture regain. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Vol. III*, (pp. 158-167). Tokyo, Japan: Nichiei Kosen.

Sikorski, J. (1963). The fine structure of animal and man-made fibers. In J. W. Hearle & R. H. Peters (Eds.), *Fibre Structure*. (pp. 269-301) London: Butterworths.

Sikorki, J. (1963). Surface structure. In J. W. Hearle & R. H. Peters (Eds.), *Fibre Structure*. (pp. 391-418) London: Butterworths.

Sikorski, J. (1975). Secondary and tertiary structure and physical properties of wool. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. I*, Aachen, (pp. 103-136). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Sikorski, J., Hepworth, A., & Buckley, T. (1971). Scanning electron microscope techniques in wool research and processing. *Applied Polymer Symposium, 18*, 887-893.

Slater, K. (1985). The characterisation of progressive deterioration in fabric properties. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Vol. III*. (pp. 100-108). Tokyo, Japan: Nichiei Kosen.

Smeltz, K. C. (1983). Why do white fabrics and garments turn yellow during storage in polyethylene bags and wrappings? *Textile Chemist and Colorist, 15*(4), 17-21.

Solomons, T. W. (1984). *Organic Chemistry*. Toronto: Wi ns.

Speakman, J. B. (1930). The elastic properties of wool in organic liquids. *Transactions of the Faraday Society, 26*, 61-69.

Speakman, J. B., & Hirst, M. C. (1933). The constitution of the keratin molecule. *Transactions of the Faraday Society, 29*, 148-172.

Speakman, P. T. (1985). Wool fibers. In M. Lewin, & E. Pearce (Eds.), *Handbook of Fiber Science and Technology (IV): Fiber Chemistry*. New York: Marcel Dekker.

Sparrow, L. G. (1984-86). Wool orthocortex-paracortex composition. *CSIRO Division of Protein Chemistry, Institute of Industrial Technology, Biennial Report 1984-86*. Melbourne, Aust: CSIRO.

Spedding, D. J. (1969). The fate of sulphur-35/sulfur dioxide in a laboratory. *Atmospheric Environment, 3*, 341-343.

Spei, M. (1975). The ordered matrix-facto or artefact? K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. II* (pp. 90-96). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Spei, M. (1985). Sorption behaviour and thermal stability on the microfibril-matrix-complex of alpha-keratins. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Vol. I* . (pp. 312- 318). Tokyo, Japan: Nichiei Kosen.

Spencer, E. Y. (1981). *Guide to the Chemicals Used in Crop Protection*. Publication 1093,

London, Ont: Agriculture Canada.

Spivak, S. M., Wood, F. E., & Worth, J. (1981). Assessing the effects of pesticidal chemicals on historic textiles. In W. E. Williams (Ed.), *Preservation of Paper and Textiles of Historic and Artisitc Value II*. ACS Adv. Chem. Ser. 193: 333-343.

SPSSx. (1986). *SPSSx User's Guide*. Toronto: McGraw Hill.

Stansfield, G. (1985). Pest control: A collection management problem. *Museums Journal, 85*,(2), 97-99.

Stearns, E. J., & Clemson, S. C. (1985). Calculation of tristimulus values and weights with the revised CIE recommendations. *Textile Chemist and Colourist, 17*, 53-59.

Stephani, G., & Zahn, H. (1985). Investigation of blends of fine animal hairs by two-dimensional SDS-polyacrylamide gelelectrophoresis. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool and Textile Research Conference Tokyo, Vol. II*. (pp.195-204). Tokyo, Japan: Nichiei Kosen.

Stoker, H. S., & Seager, S. L. (1976). *Environmental Chemistry: Air and Water Pollution*. Dallas: Scott Foresman.

Story, K. O. (1985). *Approaches to Pest Management in Museums*. Washington, DC: Conservation Analytical Laboratory, Smithsonian.

Story, K. O. (1985). Pest Control in Airplanes. *Pest Control Technology, 13*,(10), 104-107.

Stratton, W. O. (1967). The meaning of crystallinity when judged by X-rays. In F. P. Price (Ed.), Polymer Symposia, Jan. 18, 1966, *Journal of Polymer Science, 18*, 33-49.

Swift, J. A. (1971). Histological examination of keratin fibers by scanning electron microscopy. *Applied Polymer Symposium, 18*, 185-192.

Swift, J. A. (1975). The chemical composition of various morphological components isolated from human hair cuticle. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. I* (pp. 12-21). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Swift, J. A. (1977). The histology of keratin fibers. In R. S. Asquith, *Chemistry of Natural Protein Fibers*. New York: Plenum.

Szilard, J. A. (1973). *Bleaching Agents and Techniques*. London: Noyes Data.

Szucht, E. (1971). Recovery of wet wool fibers in air as determined the rate of release. *Applied Polymer Symposium, 18*, 947

Tanaka, Y., & Shiozki, H. (1985). Heterogeneous reaction of protein fabrics treated with epoxides. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool and Textile Research Conference, Vol. IV*, (pp. 441-460). Tokyo, Japan: Nichiei Kosen.

Taylor, D. S. (1985). Physical testing of textiles. *Textiles, 14*,(1), 21-28.

Taylor, D. S. (1985). Wool technologies-present and future. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool and Textile Research Conference, Vol. I*, (pp. 27-69). Tokyo, Japan: Nichiei Kosen.

Taylor, H. M. (1985). Physical testing of textiles. *Textiles. 14*,(1), 21-24.

Tao, X., & Postle, R. (1985). Influence of physical ageing on stress relaxation of single wool fibers and textile assemblies. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool and Textile Research Conference, Vol. I* , (pp. 233-242). Tokyo: Nichiei Kosen.

Tester, D. H. (1987). Fine structure of cashmere and superfine merino wool fibers. *Textile Research Journal, 57* (4), 213-219.

Tester, D. H., & Makinson, K. R. (1982). Transmission electron microscopy studies of the permanganate/salt shrinkproofing treatment of wool. *Textile Research Journal, 52*, 227-232.

Thomson, G. T. (1978). *The Museum Environment.* London: Butterworths.

Townsend, M. W. (198). Moths and wool. *Textiles.* Xerox copy.

UNESCO. (1979). Recommendations of the Pacific Regional Conservation Centre. *The Conservation of Cultural Materials in Humid Climates, UNESCO Regional Seminar, February, 1979.* Canberra College: Australia.

Trollip, N G., Maasdorp, A. P., & Van Rensburg, N. J. (1985). A study of mordant bleaching of karakul. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference Tokyo, Vol. IV*, (pp. 141-151). Tokyo, Japan: Nichiei Kosen.

Trotman, E. R. (1984). *Dyeing and Chemical Technology of Textile Fibres.* Worcester, England: Oxford.

Truman, B. B. (1976). *The Scientific Guide to Pest Control Operation.* (3rd Ed.) Purdue University: Harvest.

Truter, E. V. (1973). *Introduction to Natural Protein Fibres: Basic Chemistry.* London: Paul Elek.

Tucker, D. J., & Whenwell, C. S. (1975). The yellowing of chemically modified wool by light. In K. Zeigler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Aachen, Vol. I* (pp. 590-600). Aachen: Deutsches Wollforschungsinstitut an der Technischen Hochschule.

Tweedie, A. S., Mitton, M. T., & Fry, M. (1959). A study of the relationship between breaking time and breaking load for fabrics tested on CRE and CRT machines. *Textile Research Journal, 29*, 235-251.

Veldsman, D. P. (1966a). Weathering in wool, part 2. *Wool Science Review, 28*, 12-21.

Veldsman, D. P. (1966b). Weathering in wool, part 3. *Wool Science Review, 29*, 34-44.

Veldsman, D. P. (1975). Wet and dry finishing of wool and wool blends. In K. Ziegler (Ed.),

Proceedings 5th International Wool Textile Research Conference, Vol. 1, Aachen, (pp. 497-498). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Veldman, D. P., & Swanepoel, O.A. (1971). The influence of the rate of chlorination on the shrinkproofing of wool with DCCA. *Applied Polymer Symposium, 18*, 691-700.

Vettorazzi, G. (1976). The state of the art of the toxicological evaluation carried out by the Joint FAO/WHO Meeting on pesiticide residues: Carbamate and organophosphorus pesticides used in agriculture and public health. In F. A Gunther, & J. D. Gunther (Eds.), *Residue Reviews: Vol. 1. Residues of Pesticides and Other Contaminants in the Total Environment.* (pp. 1-44). New York: Springer-Verlag.

Vigo, T. L. (1980). Protection of textiles from biodeterioration. In F. Pertegamo (Ed.), *Conservation of Textiles International Conference*. (pp. 18-26) Milan, Italy.

Vigo, T. L. (1983). Protection of textiles from biological attack. In M. Lewin, Sello, S. B. (Eds.), *Handbook of Fiber Science and Technology: Vol II. Chemical Processing of Fibers and Fabrics: Functional Finishes Part A.* New York Marcel Dekker.

Von Bergen, W. (1963). *Wool Handbook, Vol I.* Toronto: Interscience.

Von Bergen, W. (1970). *Wool Handbook, Vol II.* Toronto: John Wiley.

Ward, P. R. (1976). Getting the bugs out. *Museum Methods Manual*. British Columbia Provincial Museum.

Warner, J. J. (1980). Continuous solvent refining of wool grease. In *Proceedings of the 6th Quinquennial International Wool Textile Research Conference, Pretoria 26 August-3 September, 1980.* Pretoria, SA:SAWTRI.

Watt, I. C. (1964). Sorption of organic vapors by wool keratin. *Journal of Applied Polymer Science, 8*, 1737-1750.

Watt, I. C. (1971). Modification of physical properties of wool fibers. *Applied Polymer Symposium, 18*, 905-914.

Watt, I. C. (1975). Importance of the wool-water relationship to fibre properties. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. II*, Aachen, (pp. 402-413). Aachen: Deutsches Wollforschungsinstitut and der technischen Hochschule.

Weatherburn, A. S., & Bayley, C. H. (1952). The sorption of synthetic reactive compounds by textile fibers. *Textile Research Journal, 22*, [illegible]

Weaver, J. W. (1984). *Analytical Methods for a Textile Laborat[illegible]*

Weigmann, H. D., & Dansizer, C. J. (1971). Effects of cross-linking [illegible]al properties of keratin fibers. *Applied Polymer Symposium, 18*,

Weigmann, H. D., Steeken, I., Moore, R. A., & Ruetsch, S. (1985). Analysis of dye transport phenomena in Hercosett treated wool by microspectrophotometry. In M. Sakamoto (Ed.), *Proceedings of the 7th International Wool Textile Research Conference, Tokyo,*

Vol. V, (pp. 141-149). Tokyo, Japan: Nichiei Kosen.

Wentz, M. (1985). Experimental studies on the effect of aqueous and nonaqueous treatments on historic textiles. In H. L. Needles, & S. H. Zeronian (Eds.), *Advances in Chemistry Series: Vol. 212. Historic Textiles and Paper Materials Conservation and Characterization.* Washington, DC: American Chemical Society.

Westrate, D. F. (1984). Warning: your collection may bugged. *American Philatelist, 98,* (9), 929-932.

Whewell, C. S. (1977). The Chemistry of Wool Finishing. In R. S. Asquith (Ed.), *Chemistry of Natural Protein Fibers.* (pp. 333-369)

Whewell, C. S., Stevens, C. B., & Amin, S. C. (1971). Observations on the dyeing behavior of chemically modified wools. *Applied Polymer Symposium, 18,* 387-408.

Whiteley, K. J., Campbell, M. C., Sui, C., & Kulkarni, V. G. (1975). Mechanical and chemical properties of wool fibers exhibiting extreme variations in chemical composition and crimp form. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. IV,* Aachen, (pp. 57-66) Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

WHO (World Health Organization) Geneva. (1986). *Organophoshorus Insecticides: A General Introduction.* (Finnland) Geneva: WHO

Wildman, A. B. (1954). *The Microscopy of Animal Textile Fibers.* Leeds, England: Wool Industries Research.

Williams, S. L., & Hawks, C. A. (1986) Considerations in the use of DDVP resin strips for insect pest control in biological research collections. In G.Llewellun & C.O'Rear (Eds.), *Biodeterioration 6th.* Washington, DC:

Williams, S. L. (no date). Unpublished report from author.

Williams, S. L., Laubach, R., & Genoways, H. H. (1977). A Guide to the management of recent mammal collections. *Carnegie Museum of Natural History Special Publications No. 4.* Pittsburgh: Carnegie Museum.

Williams, (1977). Metabolism of synthetic organic pesticides by anaerobic microorganism. In G. Gunther (Ed.), *Pesticide Reviews, 66.* New York: Springer-Verlap.

Windholz, M., Budavari, S., Blumetti, R., & Otterbein, E. S. (1983). *The Merck Index.* Rahway, NJ: Merck & Co.

Wlochowicz, A. (1985). Topography of changes in selected wool fibres parameters on sheep body. In M. Sakamoto (Ed.), *Proceedings for the 7th International Wool Textile Research Conference, Vol. II,* (pp. 55-84). Tokyo, Japan: Nichiei Kosen.

Wofram, L. J., & Milligan, B. (1975). Non-covalent bonding in keratins: the effect of esterification and acylation on the properties of wool. In K. Ziegler (Ed.), *Proceedings 5th International Wool Textile Research Conference, Vol. III,* Aachen, (pp. 242-252). Aachen: Deutsches Wollforschungsinstitut and der Technischen Hochschule.

Woods, E. F., Gillespie, J. M., Gruen, L. C., Inglis, A. S., Dowling, L. M., Marshall, R. C., Sparrow, L. C., Frenkel, M. J., Jones, L. N., Ley, K. F., & McMahon, T. W. (1979-80). Proteins of wool. *CSIRO Division of Protein Chemistry Annual Report.*

Woolman, M. S., & McGowan, E. B. (1913). Textiles: A Handbook for the Student and Consumer. New York: MacMillian. (1926 Ed.)

Worthing, C. R., & Walker, S. B. (8th Ed.). *The Pesticide Manual: A World Compendium.* British Crop Protection Council. (pp. 269-270). Xerox.

Young, G. (1987). Personel Correspondence. Canadian Conservation Institute, Ottawa.

Zahn, H. (1980). Wool is not keratin only. *Proceedings of the 6th International Wool Textile Research Conference, Pretoria.* (pp. 1- 45). Pretoria, SA:SAWTRI.

Zahn, H., Schallah, A., Scharff, D., & Meichelbeck, H. (1971). Crosslinking of wool during treatment with carboxylic acid chlorides. *Applied Polymer Symposium, 18,* 163-174.

Zavon, M. R., & Kindel, E. A. (1966). Potential hazard in using dichlorvos resin. In R. Gould (Ed.), *Organic Pesticides in the Environment: Advances in Chemistry Series 60.* 177-186 Washington, DC: American Chemical Society.

Ziegler, K., Melchert, I., & Lurken, C. (1967). N-(2-amino-2carboxyethyl)-ornithine, a new amino-acid from alkali-treated protein. Nature, 214, 404-405.

Zhao, W., & Johnson, N. A. (1986). A study of wool carbonizing Part I: Causes and prevention of localized acid damage in carbonized wool. *Textile Research Journal, 56(12),* 741-747.

Zhao, W., Johnson, N. A., & Willard, A. (1986). Investigating wool fiber damage by image analysis. *Textile Research Journal, 56,* 464-466.

Zhao, W., & Pailthorpe, T. (1987). A study of wool carbonizing Part II: Critical concentration of sulfuric acid and its effect on wool. *Textile Research Journal, 57,* 39-43.

Zhao, W., & Pailthorpe, M. T. (1987). A study of wool carbonizing Part III: Effects of the time delay in rapid carbonizing. *Textile Research Journal, 57(9),* 523-527.

Zeronian, S. H., Alger, K. W., Ellison, M. S., & Al-Khayatt, S. M. (1986). Studying the cause and type of fiber damage in textile materials by scanning electron microscopy. In H. L. Needles, & S. H. Zeronian (Eds.), *Advances in Chemistry Series: Vol. 212. Historic Textile and Paper Materials Conservation and Characterization* . Washington, DC: American Chemical Society.

Ziegler, K. (1977). Crosslinking and self-crosslinking in keratin fibers. In R. S. Asquith (Ed.), *Chemistry of Natural Protein Fibers.* New York: Pleum.

APPENDIX

TENSILE PROPERTIES DATA FILE

elongation divided by 250 times 60- speed

Load/ peak Instron id	group	peak/load	elongation	speed
all file checked				
control	unheated	7 day		
1	0	.4671	87.52	
2	0	.5369	97.98	
3	0	.5074	88 11	
4	0	.5691	73.34	
5	0	.5302	99.39	
6	0	.5893	124.5	
7	0	.5745	117.2	
8	0	.5745	102.8	
9	0	.5101	79.08	
10	0	.4846	92.62	
11	0	.5771	72.18	
12	0	.5530	100.6	
13	0	.5611	81.89	
14	0	.4940	56.68	
15	0	.5624	114.4	
16	0	.4819	82.52	
17	0	.4792	84.84	
18	0	.5476	64.73	
19	0	.5087	69.71	
20	0	.5275	81.83	
21	0	.4604	70.41	
22	0	.5007	88.62	
23	0	.4456	67.45	
24	0	.4188	48.23	
25	0	.4966	104.2	
26	0	.5329	107.4	
27	0	.5221	100.6	
28	0	.4872	74.39	
29	0	.5423	83.69	
30	0	.4805	68.18	
31	0	.4523	73.27	
32	0	.5007	66.27	
33	0	.4846	97.16	
34	0	.4564	64.84	
35	0	.5074	61.52	
36	0	.5329	86.89	
37	0	.5544	99.24	
38	0	.4725	64.64	
39	0	.5584	97.40	
40	0	.5302	80.11	

41	0	.4859	58.57	
42	0	.4899	58.97	
43	0	.4738	78.82	
44	0	.4188	70.05	
45	0	.4698	88.84	

7-day control heated

1	1	.5597	130.0	31.2
2	1	.5812	115.1	27.62
3	1	.4403	59.25	14.22
4	1	.5785	125.6	30.14
5	1	.5436	130.2	31.24
6	1	.4698	111.5	26.76
7	1	.5208	114.0	27.36
8	1	.4846	78.33	18.79
9	1	.3973	56.68	13.50
10	1	.4685	97.2	23.32
11	1	.5101	111.4	26.73
12	1	.5007	81.59	
13	1	.5020	88.34	
14	1	.5651	118.4	
15	1	.5047	121.3	
16	1	.4778	100.9	
17	1	.4872	102.4	
18	1	.4832	151.1	
19	1	.4523	108.7	
20	1	.4564	115.4	
21	1	.5020	138.6	
22	1	.4886	115.3	
23	1	.5557	153.4	
24	1	.5248	132.2	
25	1	.4658	103.7	
26	1	.5195	119.1	
27	1	.4591	95.96	
28	1	.5289	129.3	
29	1	.4980	126.7	
30	1	.4504	89.31	
31	1	.4886	86.97	
32	1	.4778	97.2	
33	1	.5503	119.7	
34	1	.5557	100.4	
35	1	.5208	115.0	
36	1	.5168	97.51	
37	1	.5517	128.5	
38	1	.5597	124.2	
39	1	.5195	110.8	
40	1	.5852	151.8	
41	1	.4953	92.44	

42	1	.5289	98.34
43	1	.5476	110.6
44	1	.4685	92.72
45	1	.5490	133.0

7 day Swap

1	2	.5248	99.49
2	2	.5195	134.4
3	2	.5423	112.4
4	2	.5772	113.2
5	2	.5356	90.85
6	2	.5329	85.5
7	2	.5557	101.8
8	2	.5436	91.04
9	2	.5557	107.9
10	2	.5812	116.6
11	2	.6054	137.8
12	2	.5235	109.7
13	2	.5517	111.5
14	2	.5906	114.6
15	2	.5570	98.26
16	2	.5047	100.3
17	2	.5423	92.60
18	2	.4805	97.04
19	2	.5570	123.0
20	2	.5047	113.4
21	2	.4926	124.9
22	2	.5007	117.3
23	2	.4765	98.84
24	2	.5839	141.5
25	2	.4778	103.7
26	2	5369	121.7
27	2	.5584	137.3
28	2	.5195	113.9
29	2	.4617	94.92
30	2	.4564	119.3
31	2	.5476	124.7
32	2	.5624	128.2
33	2	.5235	90.27
34	2	.6013	117.9
35	2	.5382	129.9
36	2	.5691	141.4
37	2	.5463	116.3
38	2	.5611	142.2
39	2	.6107	135.8
40	2	.5664	136.8
41	2	.5409	97.44
42	2	.5221	120.8
43	2	.5960	150.8
44	2	.6027	125.6
45	2	.5262	125.6

21 day control heated			
1	3	.5114	108.9
2	3	.5302	109.4
3	3	.5208	105.8
4	3	.5906	106.4
5	3	.5315	97.05
6	3	.4859	81.89
7	3	.5114	107.4
8	3	.5101	101.2
9	3	.5825	136.6
10	3	.5329	108.4
11	3	.4859	102.5
12	3	.5302	117.6
13	3	.5584	133.0
14	3	.5275	108.4
15	3	.5275	119.5
16	3	.4564	113.2
17	3	.4577	99.76
18	3	.4671	118.8
19	3	.4671	95.36
20	3	.5087	130.9
21	3	.4805	119.4
22	3	.4792	100.9
23	3	.4725	84.38
24	3	.4698	114.3
25	3	.5020	101.5
26	3	.5235	117.5
27	3	.4738	90.84
28	3	.4752	101.2
29	3	.5799	151.9
30	3	.5289	117.9
31	3	.4980	117.7
32	3	.4765	113.4
33	3	.4765	125.0
34	3	.5248	141.7
35		.4792	127.0
36		.4926	114.4
37		.5060	134.6
38	3	.4550	136.7
39	3	.4523	128.0
40	3	.4295	100.4
41	3	.4846	119.8
42	3	4510	112.6
43	3	.671	125.1
44	3	.4685	106.4
45	3	.4644	112.2
21 day SWAP			
1	4	.5866	134.4

2	4	.5329	89.33
3	4	.5248	121.0
4	4	.5476	114.1
5	4	.5141	110.2
6	4	.4899	116.0
7	4	.4886	88.02
8	4	.5987	158.8
9	4	.5342	120.8
10	4	.5409	101.8
11	4	.5638	130.1
12	4	.5651	119.2
13	4	.5356	108.1
14	4	.5825	132.4
15	4	.5315	118.3
16	4	.5181	112.2
17	4	.5476	118.2
18	4	.5611	119.2
19	4	.5799	150.4
20	4	.5879	138.1
21	4	.5423	117.1
22	4	.5289	124.9
23	4	.5705	127.0
24	4	.5329	125.5
25	4	.5772	134.5
26	4	.5101	118.0
27	4	.5570	143.7
28	4	.5503	144.8
29	4	.5262	131.7
30	4	.5409	142.7
31	4	.5302	146.9
32	4	.5356	119.8
33	4	.5168	185.6
34	4	.5289	112.4
35	4	.4403	95.46
36	4	.4510	93.28
37	4	.4819	99.81
38	4	.5369	122.1
39	4	.5181	113.1
40	4	.5329	141.4
41	4	.4792	114.7
42	4	.5342	141.6
43	4	.5020	121.4
44	4	.4537	94.55
45	4	.4886	138.4

2nd series unheated control for 21 day

1	5	.5315	114.4
2	5	.5369	98.34
3	5	.5369	91.89
4	5	.5369	80.24
5	5	.5262	88.14
6	5	.54[illegible]3	98.52
7	5	.53[illegible]2	80.37
[illegible]	5	.4685	68.26
9	5	.5087	64.89
10	5	.4899	57.03
11	5	.5195	91.19
12	5	.5396	88.78
13	5	.5342	70.23
14	5	.5168	93.69
15	5	.4685	63.08
16	5	.4658	86.55
17	5	.5168	102.5
18	5	.4913	87.59
19	5	.4725	101.9
20	5	.5127	86.41
21	5	.4778	95.7
22	5	.4752	94.59
23	5	.4523	64.69
24	5	.5007	92.04
25	5	.4	81.06
26	5	.4[illegible]	66.62
27	5	.575[illegible]	108.5
28	5	.4765	65.85
29	5	.5396	104.6
30	5	.5289	94.6
31	5	.5127	103.2
32	5	.4953	79.94
33	5	.5020	102.4
34	5	.4497	73.77
35	5	.4953	78.28
36	5	.5047	86.22
37	5	.5007	89.39
38	5	.4523	82.36
39	5	.4429	104.3
40	5	.5168	99.74
41	5	.4497	68.08
42	5	.5074	83.72
43	5	.4846	85.48
44	5	.4899	117.3
45	5	.5074	71.4

35 day control unheated checked (33 no br)

1	6	.4993	76.95
2	6	.4658	109.0
3	6	.4671	75.98
4	6	.4483	85.69
5	6	.4792	87.87
6	6	.4617	91.66
7	6	.4268	103.8
8	6	.4497	93.77
9	6	.4537	86.18
10	6	.4389	86.58
11	6	.4188	75.98
12	6	.4497	72.85
13	6	.4336	52.36
14	6	.4470	83.88
15	6	.4872	75.63
16	6	.4993	81.01
17	6	.4805	56.95
18	6	.4591	52.41
19	6	.4846	101.9
20	6	.4644	89.60
21	6	.4859	79.03
22	6	.4550	58.31
23	6	.4470	65.55
24	6	.4483	63.69
25	6	.5248	93.66
26	6	.5221	96.81
27	6	.5731	94.98
28	6	.5221	81.80
29	6	.5490	91.47
30	6	.4604	46.47
31	6	.4416	63.54
32	6	.4322	73.62
33	6	.4685	72.63
34	6	.5101	78.43
35	6	.5275	79.18
36	6	.5168	94.99
37	6	.5342	96.76
38	6	.4926	78.02
39	6	.5248	86.84
40	6	.4913	85.60
41	6	.4886	88.52
42	6	.4792	80.33
43	6	.4550	71.30
44	6	.4631	93.07
45	6	.4859	75.48

35 day heated control		checked	(34 no br)
1	7	.5114	110.2
2	7	.4886	97.47

	7	.4886	73.31
4	7	.4926	93.17
5	7	.4523	96.16
6	7	.5020	114.5
7	7	.5289	104.4
8	7	.4993	99.11
9	7	.5329	117.7
10	7	.4940	107.9
11	7	.4725	108.7
12	7	.4228	75.31
13	7	.4819	79.13
14	7	.4332	84.29
15	7	.4711	104.0
16	7	.4711	88.91
17	7	.3933	88.27
18	7	.4443	84.77
19	7	.4389	133.2
20	7	.5101	127.9
21	7	.4188	80.25
22	7	.4456	85.02
23	7	.4980	99.74
24	7	.5074	124.6
25	7	.4859	110.5
26	7	.4591	116.3
27	7	.4899	94.81
28	7	.4671	85.04
29	7	.4859	103.6
30	7	.4685	99.21
31	7	.4980	98.98
32	7	.5007	103.3
33	7	.4926	91.84
34	7	.5208	99.11
35	7	.4577	97.84
36	7	.5007	143.1
37	7	.4980	102.2
38	7	.4899	117.6
39	7	.5315	133.3
40	7	.4859	91.38
41	7	.4832	86.27
42	7	.4765	103.5
43	7	.4765	101.1
44	7	.4859	115.2
45	7	.4591	91.82

35	day SWAP	(20 no br)	checked	
1	8		.5007	99.18
2	8		.5906	140.1
3	8		.5330	128.9
4	8		.5490	90.01

5	8	.5731	121.9
6	8	.5289	111.1
7	8	.4953	117.9
8	8	.5557	123.5
9	8	.4685	90.52
10	8	.4859	107.0
11	8	.4792	97.57
12	8	.5060	138.0
13	8	.5436	97.27
14	8	.5168	102.6
15	8	.5517	139.9
16	0	.4899	122.3
17	8	.4658	108.8
18	0	.4617	101.9
19	0	.498[illegible]	115.0
20	8	.4711	91.06
21	8	.5342	104.9
22	8	.5530	119.1
23	8	.5033	97.30
24	8	.5007	91.34
25	8	.5127	110.6
26	8	.4685	61.19
27	8	.4886	95.44
28	8	.5208	113.8
29	8	.5181	105.0
30	8	.5275	103.9
31	8	.5020	90.26
32	8	.4792	94.26
33	8	.4886	134.6
34	8	.4617	76.40
35	8	.5020	90.44
36	8	.4993	81.48
37	8	.5074	137.8
38	8	.4886	108.1
39	8	.5168	88.60
40	8	.5221	120.7
41	8	.4336	75.43
42	8	.4510	83.29
43	8	.4631	72.48
44	8	.5020	100.2
45	8	.4819	111.4

RAW DATA FOR COLOR DIFFERNCE MEASUREMENTS

COLOR id	yellowness group	L*	a*	b*
unheat				
1	0	85 10	-2.73	8.40
2	0	85.44	-2.90	8.47
3	0	85.92	-2.47	8.27
4	0	85.98	-2.72	9.33
5	0	85.74	-2.69	9.31
6	0	84.36	-3.20	10.74
7	0	84.64	-3.19	10.81
8	0	84.56	-3.25	10.92
9	0	84.74	-3.24	10.82
10	0	84.60	-3.16	10.76
11	0	83.88	-3.08	11.18
12	0	82.45	-2.58	10.18
13	0	79.28	-1.55	10.18
14	0	83.98	-3.04	10.66
15	0	84.40	-3.14	10.68
7 heat				
1	1	83.83	-3.58	12.25
2	1	83.85	-3.56	12.60
3	1	84.22	-3.53	3.08
4	1	84.02	-3.56	12.29
5	1	84.33	-3.62	12.29
6	1	84.45	-3.48	12.78
7	1	84.53	-3.46	12.80
8	1	84.58	-3.46	12.81
9	1	84.68	-3.49	12.60
10	1	84.57	-3.54	12.64
11	1	85.28	-3.70	12.23
12	1	85.81	-3.67	11.39
13	1	85.02	-3.30	12.79
14	1	85.51	-3.63	12.69
15	1	85.89	-3.88	12.40
7 swat				
1	2	83.53	-2.70	17.28
2	2	84.08	-3.10	16.44
3	2	83.89	-3.22	16.34
4	2	83.96	-3.10	16.83
5	2	83.80	-2.97	17.04
6	2	82.99	-2.46	16.45
7	2	82.85	-2.42	16.51
8	2	82.85	-2.40	16.54
9	2	82.89	-2.39	16.52
10	2	82.66	-2.28	16.42
11	2	81.87	-2.50	16.06
12	2	81.96	-2.38	15.91

13	2	81.96	-2.38	15.91
14	2	82.12	-2.64	16.25
15	2	82.17	-2.63	16.27
unheat				
1	3	85.49	-2.73	9.39
2	3	85.50	-2.81	9.19
3	3	85.36	-2.81	8.71
4	3	86.68	-2.84	9.15
5	3	86.14	-2.87	9.27
6	3	86 09	-2.78	9.93
7	3	86.23	-2.68	9.61
8	3	86.18	-2.66	9.98
9	3	85.92	-2.83	9.65
10	3	85.97	-2.83	9.65
11	3	85.85	-2.67	10.17
12	3	86.23	-2.73	9.47
13	3	85.90	-3.01	8.99
14	3	84.61	-3.06	9.64
15	3	85.66	-3.11	9.83
21heat				
1	4	83.88	-3.02	13.17
2	4	84.20	-3.17	12.50
3	4	84.96	-3.31	12.62
4	4	84.05	-3.18	13.28
5	4	84.47	-3.28	13.19
6	4	84.47	-4.13	16.81
7	4	85.12	-4.21	14.77
8	4	84.74	-3.99	15.11
9	4	84.57	-3.91	15.23
10	4	84.58	-3.86	15.57
11	4	85.05	-3.37	10.50
12	4	85.18	-3.49	10.29
13	4	86.22	-3.31	10.10
14	4	85 16	-3.14	10.96
15	4	85.90	-3 18	10.54
21swat				
1	5	81.33	-2.08	20.69
2	5	81.57	-2.05	20.69
3	5	81.18	-1.92	20.90
4	5	81.09	-1.94	20.98
5	5	81.33	-1.96	21.18
6	5	80.94	-1.92	20.26
7	5	81.39	-1.65	20.36
8	5	81.54	-1.74	20.40
9	5	81.45	-1.57	20.54
10	5	80.80	-2.01	20.07
11	5	81.90	-2.74	19.23
12	5	82.25	-2.69	19.52
13	5	81.05	-2.70	19.25
14	5	81.44	-2.77	19.45
15	5	82.61	-3.07	19.56
unheat				

1	6	85.20	-2.63	8.60
2	6	85.29	-2.55	8.68
3	6	85.65	-2.69	8.68
4	6	85.37	-2.73	8.54
5	6	85.64	-2.73	8.47
6	6	84.93	-2.66	8.75
7	6	85.06	-2.76	8.70
8	6	85.22	-2.67	8.73
9	6	85.58	-2.55	8.35
10	6	85.75	-2.64	8.33
11	6	86.33	-2.49	7.06
12	6	85.83	-2.69	8.05
13	6	84.36	-2.50	8.50
14	6	84.18	-2.53	8.42
15	6	83.43	-2.28	7.76
35heat				
1	7	84.36	-3.26	10.35
2	7	84.78	-3.13	10.40
3	7	85.14	-3.08	10.53
4	7	84.07	-3.41	9.94
5	7	84.70	-3.52	9.99
6	7	85.51	-3.35	10.16
7	7	86.07	-3.41	10.42
8	7	85.94	-3.42	10.12
9	7	85.55	-3.36	10.47
10	7	85.98	-3.37	10.46
11	7	86.51	-3.19	10.82
12	7	85.93	-3.08	10.62
13	7	86.05	-3.12	10.58
14	7	86.08	-3.23	10.53
15	7	86.20	-3.31	9.78
35swat				
1	8	84.54	-2.41	16.69
2	8	84.58	-2.28	16.67
3	8	84.40	-2.53	16.41
4	8	83.91	-2.14	16.15
5	8	83.61	-2.17	15.97
6	8	83.70	-2.19	19.18
7	8	83.50	-2.27	19.11
8	8	83.67	-2.21	19.11
9	8	82.45	-2.62	17.98
10	8	83.36	-2.71	18.17
11	8	84.59	-2.69	15.44
12	8	84.65	-2.81	15.46
13	8	84.70	-3.05	15.50
14	8	84.34	-2.77	15.55
15	8	84.15	-2.85	15.53
methanol unheated				
1	9	85.80	-3.14	8.83
2	9	85.66	-3.07	8.81
3	9	85.39	-2.96	9.76
4	9	85.35	-2.89	9.11

5	9	85.90	-2.93	9.24
m48				
1	10	82.65	-.71	20.26
2	10	82.86	-.76	20.04
3	10	82.70	-.66	19.93
4	10	82.01	-.32	20.64
5	10	82.09	-.40	20.62
m7				
1	11	78.29	1.63	29.14
2	11	78.30	1.63	29.13
3	11	78.37	1.73	29.03
4	11	78.45	1.65	29.11
5	11	78.70	1.45	28.17
m14swat				
1	12	75.90	2.06	33.56
2	12	75.86	2.12	33.70
3	12	75.86	2.11	33.56
4	12	75.85	2.11	33.66
5	12	76.11	1.83	33.53
m21heat				
1	13	86.92	-3.52	9.29
2	13	87.43	-3.42	8.44
3	13	86.24	-3.51	10.74
4	13	86.37	-3.35	10.30
5	13	87.25	-3.37	9.14
m21swat				
1	14	75.28	2.13	35.32
2	14	76.11	1.61	35.55
3	14	76.45	1.21	35.48
4	14	75.48	2.00	34.44
5	14	75.84	1.69	34.41

Kiton Red unheated

IDsample	GROUP	LSTAR	ASTAR	BSTAR
1	00	64.06	31.50	9.84
2	00	65.55	29.55	10.12
3	00	64.02	31.46	9.65
4	00	64.29	31.02	9.73
5	00	63.57	30.71	9.62
6	00	66.79	29.20	9.56
7	00	66.45	29.73	9.37
8	00	65.30	30.96	9.72
9	00	65.82	30.14	9.62
10	00	65.71	30.79	9.38
11	00	63.88	30.71	9.47
12	00	63.84	30.84	9.42
13	00	63.80	30.82	9.51
14	00	65.05	30.62	9.39
15	00	64.91	30.63	9.64
7heat				
1	0	71.33	23.05	9.10

2	0	71.42	22.73	9.14
3	0	71.80	22.97	8.88
4	0	72.25	22.49	8.23
5	0	72.38	22.10	8.68
6	0	70.67	20.42	7.31
7	0	71.23	19.68	7.36
8	0	70.71	20.12	7.36
9	0	70.37	21.64	6.94
10	0	69.81	22.33	6.64
11	0	71.46	22.04	8.68
12	0	71.45	22.04	8.67
13	0	72.10	21.37	8.81
14	0	71.44	22.19	8.62
15	0	71.48	22.11	8.61
7swat				
1	1	66.39	28.34	10.25
2	1	66.66	28.27	10.30
3	1	66.74	28.14	10.23
4	1	66.49	29.16	9.79
5	1	66.54	29.05	9.68
6	1	65.00	30.00	8.95
7	1	68.35	25.81	7.37
8	1	65.28	29.79	9.16
9	1	68.43	26.40	7.61
10	1	68.05	25.51	7.49
11	1	67.60	26.27	9.46
12	1	66.57	27.89	9.58
13	1	68.16	26.41	8.28
14	1	66.38	27.59	9.79
15	1	66.69	27.01	9.84
21heat				
1	2	61.47	34.94	8.31
2	2	61.37	35.01	8.27
3	2	62.17	34.29	8.16
4	2	62.30	34.22	8.23
5	2	62.84	33.69	8.87
6	2	61.8	32.76	7.48
7	2	62.[illegible]	32.81	7.53
8	2	62.40	32.03	7.25
9	2	63.61	31.31	6.84
10	2	63.24	31.52	7.18
11	2	61.25	30.58	9.57
12	2	61.46	30.57	9.80
13	2	62.08	29.53	9.66
14	2	62.42	28.98	9.37
15	2	63.59	27.94	8.39
21swat				
1	3	70.76	23.23	10.83
2	3	70.76	23.72	11.00
3	3	71.44	23.65	10.37
4	3	71.21	23.42	10.72
5	3	70.80	23.86	10.88

6	3	71.	19.28	15.44
7	3	71.59	19.83	15.14
8	3	71.37	19.67	15.26
9	3	70.55	18.91	15.18
10	3	72.03	16.84	15.31
11	3	69.08	26.02	15.11
12	3	69.14	25.76	15.16
13	3	69.73	25.43	14.94
14	3	71.40	24.29	13.98
15	3	70.14	24.73	14.75
35heat				
1	4	68.82	23.26	5.93
2	4	69.71	22.32	5.83
3	4	70.34	21.67	6.03
4	4	70.09	22.02	6.13
5	4	67.98	24.26	6.38
6	4	68.56	23.47	6.34
7	4	68.63	24.43	6.16
8	4	68.08	25.01	6.58
9	4	68.52	24.68	6.15
10	4	68.25	25.35	6.55
11	4	68.20	25.35	6.91
12	4	69.37	22.97	5.79
13	4	69.20	22.78	5.77
14	4	68.00	23.78	6.52
15	4	67.29	24.87	5.81
35swat				
1	5	80.83	7.38	10.51
2	5	80.39	7.93	10.19
3	5	80.50	7.93	10.12
4	5	79.51	9.23	10.79
5	5	79.58	9.33	10.71
6	5	81.80	5.56	13.26
7	5	82.23	4.79	13.47
8	5	81.95	6.01	12.54
9	5	82.33	4.93	13.04
10	5	82.24	5.08	12.92
11	5	82.74	5.52	9.46
12	5	82.77	5.93	9.28
13	5	82.47	6.15	8.89
14	5	82.77	5.76	9.07
15	5	82.79	5.93	9.03
mheat				
1	6	75.11	18.63	11
2	6	75.61	18.31	8.94
3	6	76	19.12	7.96
4	6	75.67	19.04	8.13
5	6	76.15	18.75	8.16
6	6	75.76	20.58	7.84
7	6	75.46	21.04	7.96
8	6	75.91	20.29	8.01
9	6	75.26	20.57	8.32

10	6	75.25	20.55	8.46
11	6	74.86	19.64	8.35
12	6	.49	18.18	8.61
13	6	4.49	19.13	8.64
14	6	73.97	19.90	9.07
15	6	75.36	18.54	8.56
mswat				
1	7	64.32	23.54	18.26
2	7	64.11	23.10	18.28
3	7	63.87	23.49	18.59
4	7	64.44	24.39	19.39
5	7	65.03	24.18	19.31
6	7	63.00	25.61	17.62
7	7	63.54	25.42	17.81
8	7	63.36	25.45	18.79
9	7	63.36	25.14	18.74
10	7	63.40	25.47	18.85
11	7	60.95	29.26	16.92
12	7	61.03	29.25	16.98
13	7	61.64	28.92	16.65
14	7	60.57	29.30	16.62
15	7	61.44	29.21	16.99
mu				
1		77.15	10.78	8.75
2		76.82	11.67	8.55
3	8	77.70	10.87	8.50
5	8	77.27	9.99	8.87
6	8	78.02	8.67	9.03
7	8	77.49	9.86	8.66
8	8	77.30	10.68	8.52
9	8	77.74	10.33	8.57
10	8	79.12	8.77	8.37
11	8	77.22	11.01	8.68
12	8	77.37	11.02	8.66
13	8	77.66	10.86	8.72
14	8	77.76	9.05	8.75
15	8	78.11	8.93	8.62

Methylene Blue

7heat		L*	a*	b*
1	0	61.77	-11.20	-11.24
2	0	62.29	-11.27	-11.31
3	0	62.93	-11.55	-10.29
4	0	64.01	-12.27	-11.21
5	0	64.34	-11.85	-11.36
6	0	63.33	-11.79	-9.39
7	0	63.87	-11.86	-9.16
8	0	64.01	-12.25	-9.53
9	0	64.19	-11.35	-10.09
10	0	63.28	-11.89	-9.87
11	0	64.12	-12.28	-10.74
12	0	64.45	-11.91	-10.76

13	0	64.28	-11.89	-10.62
14	0	64.03	-12.61	-10.75
15	0	64.40	-12.65	-10.69
7swat				
1	1	67.06	-12.89	-8.30
2	1	68.75	-12.58	-7.34
3	1	67.05	-13.20	-8.48
4	1	66.41	-12.85	-8.66
5	1	66.06	-12.48	-8.77
6	1	63.96	-10.48	-5.38
7	1	63.92	-10.20	-5.56
8	1	63.10	-10.04	-5.56
9	1	60.06	-8.44	-5.54
10	1	59.00	-8.84	-6.12
11	1	64.12	-12.28	-10.74
12	1	64.45	-11.97	-10.76
13	1	64.28	-11.89	-10.62
14	1	64.03	-12.61	-10.75
15	1	64.40	-12.65	-10.69
21heat				
1	2	66.93	-13.26	-9.33
2	2	66.67	-13.20	-9.22
3	2	66.50	-13.12	-9.08
4	2	66.97	-13.33	-9.00
5	2	66.52	-12.78	-9.37
6	2	65.89	-13.55	-10.15
7	2	65.98	-13.29	-10.16
8	2	66.49	-13.10	-9.63
9	2	66.01	-13.26	-9.65
10	2	66.52	-13.18	-9.34
11	2	67.28	-13.80	-8.40
12	2	67.04	-13.24	-8.38
13	2	67.41	-13.47	-8.10
14	2	66.49	-13.80	-9.00
15	2	66.87	-14.21	-8.66
21swat				
1	3	71.78	-12.17	2.87
2	3	71.51	-11.73	2.71
3	3	70.23	-13.30	1.89
4	3	70.98	-13.63	2.24
5	3	70.44	-13.14	2.12
6	3	76.85	-13.56	3.56
7	3	76.54	-13.35	3.51
8	3	75.88	-13.60	3.09
9	3	75.82	-13.39	3.21
10	3	76.15	-13.30	3.33
11	3	74.17	-13.86	2.35
12	3	74.42	-13.04	2.48
13	3	74.41	-13.55	2.47
14	3	75.30	-14.09	3.28
15	3	75.07	-13.72	3.00
35heat				

1	4	68.90	-10.55	-8.96
2	4	68.99	-10.79	-8.66
3	4	68.06	-9.76	-9.00
4	4	68.69	-10.39	-9.22
5	4	69.32	-10.22	-8.75
6	4	69.11	-9.60	-7.82
7	4	69.76	-9.29	-7.86
8	4	69.41	-9.60	-7.89
9	4	68.99	-9.62	-7.73
10	4	68.99	-9.46	-7.71
11	4	66.65	-7.67	-5.37
12	4	66.98	-6.75	-4.56
13	4	66.82	-7.00	-4.78
14	4	65.42	-7.73	-5.92
15	4	66.77	-7.36	-4.93
35swat				
1	5	73.52	-11.36	4.05
2	5	74.07	-11.15	4.45
3	5	74.31	-11.16	4.62
4	5	73.84	-11.22	4.41
5	5	74.14	-10.72	4.48
6	5	77.98	-8.76	5.97
7	5	78.96	-8.67	6.52
8	5	78.25	-8.91	6.29
9	5	77.76	-8.56	5.58
10	5	79.76	-8.08	6.25
11	5	78.41	-8.64	3.73
12	5	78.47	-8.52	3.80
13	5	79.90	-8.02	4.56
14	5	78.58	-9.00	3.64
15	5	78.74	-8.35	4.24
mu				
1	6	74.41	-5.30	-1.14
2	6	74.60	-5.50	-.94
3	6	74.17	-5.45	-1.07
4	6	74.20	-5.48	-1.03
5	6	74.22	-5.44	-1.79
6	6	71.55	-4.92	-1.90
7	6	71.59	-5.11	-1.94
8	6	72.20	-4.97	-1.38
9	6	72.64	-4.81	-1.40
10	6	73.17	-5.80	-.89
11	6	72.92	-5.26	-1.38
12	6	72.12	-4.95	-1.72
13	6	72.79	-5.09	-1.56
14	6	72.47	-4.93	-1.88
15	6	72.70	-5.24	-1.54
m7heat				
1	7	63.24	-5.45	-14.[illegible]
2	7	63.16	-5.21	-14.20
3	7	63.65	-4.02	-14.39
4	7	64.21	-4.11	-14.05

5	7	64.86	-6.19	-13.42
6	7	63.53	-4.82	-11.93
7	7	63.24	-4.75	-12.84
8	7	64.01	-4.99	-12.71
9	7	64.60	-5.41	-10.77
10	7	65.54	-6.30	-11.03
11	7	62.35	-4.88	-12.55
12	7	63.09	-5.01	-12.03
13	7	62.23	-5.24	-13.58
14	7	61.69	-5.23	-13.75
15	7	61.61	-4.95	-14.00
m7swat				
1	8	70.59	-6.13	16.96
2	8	70.01	-6.08	16.30
3	8	70.73	-6.06	16.98
4	8	70.16	-5.84	16.69
5	8	70.70	-6.32	15.81
6	8	69.26	-6.15	16.54
7	8	69.08	-6.10	16.38
8	8	69.56	-6.12	16.59
9	8	69.09	-6.38	16.21
10	8	69.99	-6.00	15.97
11	8	67.69	-6.91	15.42
12	8	67.43	-6.54	15.14
13	8	67.43	-6.54	15.16
14	8	69.44	-6.38	15.52
15	8	70.29	-6.28	16.49
unheat				
1	9	67.42	-8.36	-4.72
2	9	67.72	-8.64	-4.72
3	9	66.80	-6.32	-5.45
4	9	66.68	-8.00	-5.21
5	9	66.09	-7.30	-5.17
6	9	66.61	-7.14	-5.05
7	9	63.67	-7.62	-6.71
8	9	63.78	-7.65	-6.64
9	9	63.43	-6.87	-6.54
10	9	64.30	-6.58	-6.23

ACRIDINE ORANGE

IDSAMP	GROUP	LSTAR	ASTAR	BSTAR
ur				
1	0	45.82	30.52	52.95
2	0	44.91	30.93	51.21
3	0	45.43	30.61	52.27
4	0	43.62	31.24	50.66
5	0	44.17	26.48	51.04
6	0	46.44	31.22	54.02
7	0	46.00	30.77	53.20
8	0	46.32	30.75	53.68
9	0	46.32	31.07	53.73
10	0	46.94	31.31	54.30

11	0	44.44	30.27	51.38
12	0	43.45	30.77	50 33
13	0	43.68	30.37	50.35
14	0	43.82	30.54	50.74
15	0	44.64	29.63	51.28
7h				
1	1	42.00	35.36	48.47
2	1	41.25	35.38	47.83
3	1	40.38	34.72	47.17
4	1	41.51	35.44	47.95
5	1	39.46	34.96	45.64
6	1	40.61	34.84	47.44
7	1	40.80	34.58	47.66
8	1	40.63	34.82	47.50
9	1	40.97	34.58	47.88
10	1	39.81	34.63	46.05
11	1	38.72	34.04	45.08
12	1	39.05	34.11	45.44
13	1	39.05	34.11	45.44
14	1	39.66	33.26	46.29
15	1	39.93	33.71	46.43
7swat				
1	2	49.44	28.92	54.60
2	2	49.44	28.95	54.72
3	2	50.20	29.61	55.25
4	2	49.60	29.03	54.83
5	2	49.80	29.03	55.19
6	2	51.04	29.32	56.54
7	2	50.87	29.65	56.54
8	2	50.68	29.69	56.11
9	2	52.25	29.40	57.58
10	2	51.73	29.54	57.37
11	2	47.65	28.06	52.57
12	2	47.67	28.30	52.27
13	2	48.22	28.36	53.10
14	2	49.80	29.12	54.83
15	2	49.21	29.11	54.19
21h				
1	3	47.15	29.52	51.05
2	3	46.20	30.16	51.60
3	3	46.70	30.40	51.40
4	3	47.58	30.81	53.98
5	3	47.75	30.38	53.94
6	3	45.66	30.68	52.19
7	3	45.11	30.52	51.52
8	3	45.49	30.56	51.98
9	3	45.60	30.65	52.25
10	3	45.57	30.52	52.09
11	3	44.20	28.22	49.10
12	3	44.81	28.31	49.65
13	3	44.54	28.24	49.29
14	3	43.92	29.21	49.37

15	3	44.57	28.32	49.29
21swat				
1	4	58.09	24.53	54.38
2	4	58.28	24.15	53.77
3	4	58.31	24.25	53.77
4	4	56.22	24.10	54.14
5	4	56.18	24.86	53.84
6	4	62.72	23.83	58.17
7	4	63.38	23.74	57.58
8	4	63.49	23.74	57.74
9	4	62.14	24.23	58.54
10	4	62.03	24.23	56.41
11	4	58.88	24.19	54.88
12	4	58.14	24.49	54.80
13	4	60.28	22.90	53.60
14	4	60.11	23.24	53.49
15	4	57.94	24.90	54.83
35heat				
1	5	49.31	31.80	57.27
2	5	49.85	31.87	57.89
3	5	49.35	31.59	56.31
4	5	49.67	32.47	57.19
5	5	53.29	32.16	59.97
6	5	47.91	31.33	55.34
7	5	49.13	31.54	56.66
8	5	51.49	30.00	49.37
9	5	48.38	31.38	55.85
10	5	48.42	31.68	56.02
11	5	48.36	31.60	55.94
12	5	47.32	29.84	53.79
13	5	46.93	30.02	53.45
14	5	47.07	30.00	53.63
15	5	47.18	30.31	53.79
35swat				
1	6	57.06	26.17	55.49
2	6	56.90	26.67	55.90
3	6	55.95	26.48	54.98
4	6	55.93	26.46	54.90
5	6	56.35	27.00	57.38
6	6	56.54	26.98	57.07
7	6	57.38	26.49	57.09
8	6	53.90	25.27	53.64
9	6	56.99	25.82	55.66
10	6	57.09	26.15	56.07
11	6	59.21	25.36	53.10
12	6	56.50	26.99	57.39
13	6	57.03	26.51	57.51
14	6	57.31	26.66	55.04
15	6	57.39	26.82	54.67
m				
1	7	49.31	31.50	56.55
2	7	48.13	31.46	55.44

3	7	47.54	31.34	55.01
4	7	47.83	31.64	55.62
5	7	49.97	31.46	56.37
6	7	46.26	30.45	52.75
7	7	47.14	30.39	53.82
8	7	47.15	30.60	53.55
9	7	47.22	31.06	54.30
10	7	46.92	30.66	53.99
11	7	46.80	30.75	53.88
12	7	45.67	30.16	52.14
13	7	45.82	30.71	52.69
14	7	45.79	30.55	51.96
15	7	46.87	30.73	53.21
m7				
1	8	49.95	32.13	56.41
2	8	51.08	30.87	51.93
3	8	49.12	31.55	55.68
4	8	48.98	31.69	55.14
5	8	49.23	31.47	55.60
6	8	49.73	31.39	56.79
7	8	49.63	31.61	56.68
8	8	50.50	31.61	57.42
9	8	46.81	30.67	52.13
10	8	46.78	30.28	52.77
11	8	46.32	29.99	52.39
12	8	49.31	32.16	57.10
13	8	50.03	32.19	58.06
14	8	50.61	32.26	58.50
15	8	50.49	32.11	58.80
mswat				
1	9	62.62	19.79	48.20
2	9	62.48	20.24	48.37
3	9	62.79	19.82	48.23
4	9	62.78	19.74	48.21
5	9	62.85	19.95	48.20
6	9	64.08	19.87	49.75
7	9	63.86	19.74	49.42
8	9	64.38	19.90	49.87
9	9	64.51	19.90	49.77
10	9	64.70	19.89	50.10
11	9	63.80	19.75	49.26
12	9	63.68	20.00	48.78
13	9	64.61	19.81	49.73
14	9	63.00	19.76	48.20
15	9	63.06	19.89	48.50

Lead Acetate

IDSAM	GROUP	LSTAR	ASTAR	BSTAR
unheat				
1	0	41.40	3.30	14.63
2	0	41.16	3.30	14.28
3	0	42.00	3.29	14.85

4	0	42.22	3.46	13.80
5	0	45.44	4.10	11.57
6	0	43.45	2.61	14.67
7	0	43.09	3.05	14.67
8	0	43.10	3.03	14.60
9	0	44.14	2.97	14.83
10	0	44.60	2.57	15.19
7heat				
1	1	44.70	2.84	13.66
2	1	56.09	1.74	8.15
3	1	54.40	2.04	9.58
4	1	44.09	2.59	13.75
5	1	45.10	4.25	11.87
6	1	55.18	1.65	15.05
7	1	55.44	1.50	15.14
8	1	54.62	1.86	15.03
9	1	54.71	1.65	15.39
10	1	55.12	2.10	16.10
7swat				
1	2	53.29	2.74	15.27
2	2	58.42	1.97	14.98
3	2	64.06	1.58	12.46
4	2	61.93	1.64	13.41
5	2	58.39	1.94	14.73
6	2	56.77	2.67	12.57
7	2	65.63	2.60	8.05
8	2	66.98	1.63	8.15
9	2	62.51	2.82	10.05
10	2	66.72	.55	11.60
11	2	55.24	1.72	14.96
12	2	55.50	1.77	14.94
13	2	56.51	1.68	14.83
14	2	54.60	1.87	15.19
15	2	54.47	1.76	15.31
21heat				
1	3	51.43	2.50	14.70
2	3	52.34	2.81	15.58
3	3	51.48	2.58	14.71
4	3	51.95	2.61	15.04
5	3	52.56	2.61	15.49
6	3	45.03	3.00	15.14
7	3	46.21	3.22	15.39
8	3	48.91	2.32	16.76
9	3	49.13	2.32	16.86
10	3	47.01	2.02	15.39
11	3	49.32	3.02	14.77
12	3	55.16	3.27	12.35
13	3	52.88	2.73	14.23
14	3	49.35	1.74	14.23
15	3	48.69	2.06	14.09
21swat				
1	4	58.74	2.45	18.23

2	4	59.35	2.60	17.91
3	4	59.05	2.54	18.25
4	4	57.34	3.07	17.49
5	4	59.40	2.99	15.22
6	4	49.34	1.44	15.28
7	4	49.06	1.46	15.18
8	4	49.02	1.58	14.87
9	4	49.67	1.51	15.73
10	4	49.45	1.54	15.92
11	4	53.49	2.84	16.67
12	4	53.45	2.77	16.65
13	4	53.91	2.91	16.56
14	4	54.1	3.17	16.46
15	4	53.49	3.23	17.08
35heat				
1	5	52.29	2.74	15.44
2	5	53.88	2.43	15.77
3	5	54.66	2.25	15.88
4	5	55.12	1.94	16.28
5	5	52.46	2.47	15.56
6	5	57.64	1.58	15.83
7	5	57.28	1.45	15.15
8	5	58.28	2.31	15.05
9	5	60.38	2.45	14.89
10	5	59.64	2.05	15.56
11	5	57.02	1.43	15.68
12	5	56.91	1.70	15.58
13	5	56.91	1.72	15.67
14	5	57.23	1.62	15.75
15	5	57.31	1.66	15.77
35swat				
1	6	51.41	2.83	15.83
2	6	51.61	2.74	15.93
3	6	52.38	2.28	16.15
4	6	51.47	2.35	16.37
5	6	51.74	2.59	16.31
6	6	49.51	2.46	15.93
7	6	50.47	2.26	16.29
8	6	48.89	2.61	15.82
9	6	49.36	2.38	15.89
10	6	48.75	2.61	15.75
11	6	49.37	2.16	15.59
12	6	49.20	2.14	15.62
13	6	49.29	2.19	15.69
14	6	47.46	2.86	15.13
15	6	47.78	2.84	15.26
m				
1	7	48.99	3.62	15.14
2	7	49.72	3.30	15.24
3	7	49.22	3.90	15.72
4	7	47.74	3.59	16.69
5	7	47.47	4.03	16.49

6	7	49.71	2.43	15.63
7	7	49.80	2.43	15.61
8	7	50.03	3.14	14.99
9	7	50.89	2.68	15.35
10	7	51.57	2.45	15.63
11	7	53.49	3.17	15.75
12	7	53.50	2.39	15.78
13	7	49.61	2.37	15.33
14	7	49.79	2.41	15.98
15	7	50.01	3.05	15.19
mheat				
1	8	51.53	2.43	14.70
2	8	51.68	2.29	14.68
3	8	51.87	2.16	14.70
4	8	50.81	2.19	15.29
5	8	51.11	2.44	14.92
6	8	55.38	2.38	16.49
7	8	55.61	2.27	16.59
8	8	55.61	2.27	16.58
9	8	57.80	3.47	16.58
10	8	57.65	2.22	16.27
7swat				
1	9	63.45	4.22	29.79
2	9	63.54	4.27	30.03
3	9	63.77	4.58	30.21
4	9	63.81	4.57	30.51
5	9	64.73	3.83	30.92
6	9	63.57	4.63	29.62
7	9	64.33	4.45	29.17
8	9	63.60	4.34	29.94
9	9	64.29	4.28	30.63
10	9	64.12	4.26	30.38
11	9	62.96	4.58	30.99
12	9	63.40	4.42	31.11
13	9	63.62	4.06	31.59
14	9	63.26	4.42	31.44
15	9	63.83	4.13	31.82

pH DATA FILE

IDSAMPLE	GROUP	PH
1	0	6.70
2	0	6.80
3	0	6.90
4	0	6.80
5	0	6.80
6	0	6.90
7	0	6.80
8	0	6.80
0	0	6.90
1	1	5.80
2	1	5.70
3	1	5.80
4	1	5.80
5	1	5.80
6	1	5.70
7	1	5.70
8	1	5.70
9	1	5.60
10	1	5.60
11	1	6.00
12	1	6.00
13	1	6.00
14	1	5.80
15	1	5.70
16	1	5.90
17	1	5.80
18	1	6.20
1	3	6.50
2	3	6.50
3	3	6.50
4	3	6.30
5	3	6.30
6	3	6.40
7	3	6.10
8	3	6.20
9	3	6.20
1	4	5.10
2	4	5.10
3	4	5.10
4	4	5.10
5	4	5.10
6	4	5.10
7	4	5.20
8	4	5.20
9	4	5.20
10	4	5.20
11	4	5.20
12	4	5.20
13	4	4.40
14	4	4.40
15	4	4.40
16	4	4.70
17	4	4.70
18	4	4.70
1	5	5.80
2	5	5.50
3	5	5.90
4	5	5.80
5	5	5.80
6	5	5.80
7	5	5.80
8	5	5.90
9	5	5.90
1	6	4.70
2	6	4.70
3	6	4.60
4	6	4.60
5	6	4.60
6	6	4.60
7	6	4.40
8	6	4.30
9	6	4.30
10	6	4.10
11	6	4.10
12	6	4.10
13	6	4.30
14	6	4.30
15	6	4.30
16	6	4.40
17	6	4.30
18	6	4.30
1	7	6.30
2	7	6.40
3	7	6.50
4	7	6.40
5	7	6.50
6	7	6.40
7	7	6.40
8	7	6.40
9	7	6.40
1	8	3.40
2	8	3.40
3	8	3.40
4	8	3.50
5	8	3.40
6	8	3.40
7	8	3.50
8	8	3.50
9	8	3.50
10	8	3.40
11	8	3.40
12	8	3.40
13	8	3.40
14	8	3.40
15	8	3.40
16	8	3.40
17	8	3.40
18	8	3.40

PHOSPHORUS AND CHLORINE DATA

	Sorption				Desorption	
1	1.3	1.3	1	1	2.2	1.3
2	1.9	1.4	2.9	3.4	2.9	1.4
5	1	1	5.8	2.7	6.6	1.8
7	3.7	1.5	1	1	7.2	1.9
14	3.7	1.6	9.8	4.4	9.2	2.5
21	3.8	2.6	14.6	7.6	9.7	3.0
Days	Cl	P	Cl	P	Cl	P
			Destilled Water Wash		Methanol Wash	

Data for graph Figure 6.11.1