STUDIES ON TREATMENTS OF EXAMINATION GLOVES FOR REDUCING THE EXTRACTABLE PROTEIN CONTENT

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BY

ELIZABETH K. I.

DEPARTMENT OF POLYMER SCIENCE AND RUBBER TECHNOLOGY
COCHIN UNIVERSITY OF SCIENCE AND TECHNOLOGY
COCHIN - 682 022

JULY 1995

PHONE: 578311 (7 Lines)

GRAMS: RUBRBOARD
TELEX: 888 285 RRII IN
FAX: 91-481-578317

रबड बोर्ड

(वाणिज्य मन्त्रालय, भारत सरकार)

THE RUBBER BOARD

(Ministry of Commerce, Government of India)

DEPARTMENT OF TECHNICAL CONSULTANCY

कोट्टयम - ९, केरल
KOTTAYAM - 686 009,
KERALA STATE.

Ref. No. TC/95/Uni

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CERTIFICATE

Certified that the thesis entitled **"Studies on treatment of Examination Gloves for reducing the Extractable Protein Content"** being submitted by Smt.ELIZABETH K.I. in partial fulfilment for the award of the degree of Master of Technology in Polymer Technology of Cochin University of Science & Technology, Cochin-22 is a record of bonafide work done by her under my guidance and supervision.

JOINT DIRECTOR (TC)

pv.

Phone : 8-5572

DEPARTMENT OF POLYMER SCIENCE AND RUBBER TECHNOLOGY
COCHIN UNIVERSITY OF SCIENCE AND TECHNOLOGY
COCHIN - 682 022

No.

Date July 15, 1995.

Prof.(Dr.)D.JOSEPH FRANCIS,
Head of the Dept.

C E R T I F I C A T E.

This is to certify that the thesis entitled **STUDIES ON TREATMENTS OF EXAMINATION GLOVES FOR REDUCING THE EXTRACTABLE PROTEIN CONTENT** submitted by Smt. **ELIZABETH K.I.** for the award of the degree of Master of Technology in **POLYMER TECHNOLOGY** of the Cochin University of Science & Technology, is a record of bonafide work carried out by her under the guidance of Mr.K.S.Gopalakrishnan, Joint Director, Technical Consultancy Department, Rubber Board, Kottayam - 9.

Prof.(Dr.)D.JOSEPH FRANCIS,
(Head of the Dept.)

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Elizabeth K.I.

ELIZABETH K.I

GLOSSARY

NR latex	- Natural rubber latex
EP content	- Extractable protein content
RP membrane	- Rubber particle membrane
HA latex	- High ammonia latex
DCL	- Doubly centrifuged latex
PV latex	- Pre vulcanised latex
PFL	- Preserved field latex
SLS	- Sodium lauryl sulphate
EBC	- Estate brown crepe
PTA	- Phosphotungstic acid
TCA	- Trichloro acetic acid

C O N T E N T S

CHAPTER 1

INTRODUCTION

Natural rubber (NR) products play an important role in our day to day life. They are useful in the fields such as transportation, communication, agriculture, sports and game, defence, health and family planning etc . It is estimated that there are about 35,ØØØ different rubber products are manufactured in India.

The bulk of the crop collected from rubber plantations is as liquid latex.[1] It is a hydrosol of dispersed rubber particles protected by an adsorbed complex film of protein, neutral lipids and phospholipids. The crop is processed into different forms which facilitate easy storage, transportation and utilization by the manufacturing industries.

The different steps in the processing of the crop to the different marketable forms of NR are shown in fig (1).

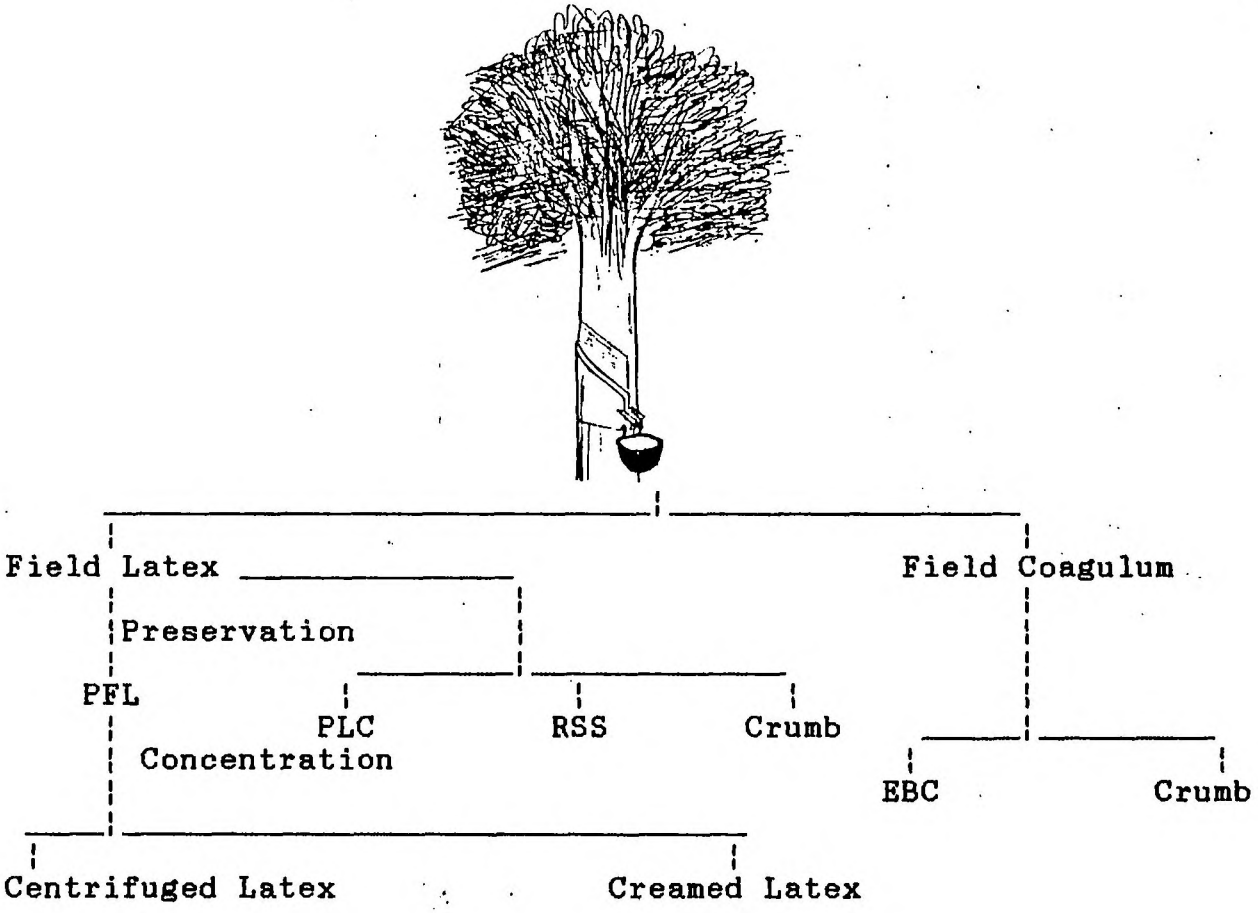
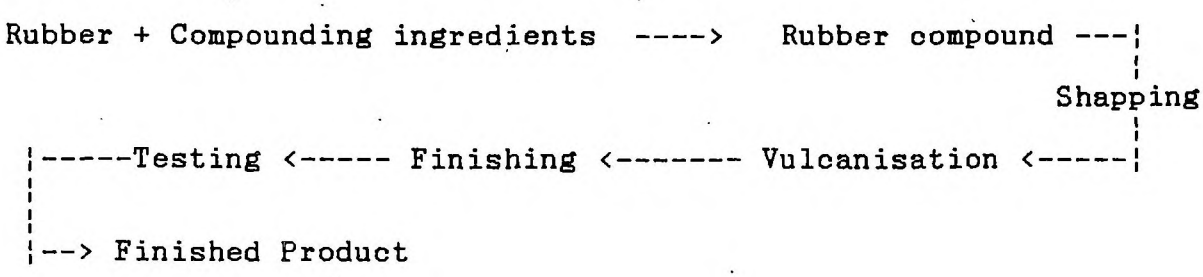


Fig. 1

The preserved field latex is processed into 60% concentrated latex by methods like creaming and centrifugation. It finds use in the manufacture of a variety of latex goods like gloves, catheters, toy balloons, condoms, nipples, foam rubber etc. The latex processed into dry forms like sheet, crepe, crumb rubber etc are used for the manufacture of automobile tyres, belting, hoses, footwears etc. The field coagulum obtained in the form of

the cup lump and tree laces are processed into crepe or crumb rubber and used for the manufacture of various dry rubber articles

The operations involved in the manufacture of rubber goods are the following-



During the process of manufacture of rubber products, rubber gets intimately mixed with a number of chemicals in definite proportion (compounding) for getting the required properties to the product. In the case of latex goods, the ingredients usually admixed with latex are stabilizers like soap, casein, Potassium hydroxide, Zinc hydroxide, accelerators like ZDC, DMBT, TMT, ZDBC etc, anti oxidants and sulphur.

Gloves, Condoms, Catheters, Balloons etc which come under the latex products are made by the process called dipping. The process essentially consists of dipping clean formers in the

latex coagulant, drying and then dipping in the suitably compounded latex. The thin film of the latex compound deposited on the surface of the former (the thickness of which can be adjusted by controlling the number of dips.) is leached in hot water to remove non-rubber constituents to the maximum possible level and then vulcanised at about 11Ø-13ØºC for a definite time depending on the compound formulation. The vulcanised film is then removed from the former, subjected to finishing operation, tested for ascertaining conformity to quality standards, packed and marketed.

I.2 THE PROTEIN RELATED ALLERGY PROBLEM

The usage of NR latex products has been reported to cause two class of reactions.[2]

Type I response - Contact urticaria and possibly anaphylactic shock.

Type IV response - Allergic contact eczema.

Type IV response is attributed to the presence of certain class of residual compounding ingredients in latex products. Thiuram, dithiocarbamates, thiazoles etc may cause dermatic reactions in hypersensitive individuals. The response is characterised by the appearance of itching, redness, swelling etc, a few

hours after contact with the rubber products and this response has been known for several years. It is difficult to avoid the use of at least one of these accelerators in sulphur vulcanised articles and those allergic to these materials should avoid close contact with latex articles. An alternative to sulphur vulcanisation is prevulcanisation with peroxides, radiation etc, but this will result in low modulus products.

Type I response appears to be relatively recent occurence (first reported in 1979) and has the following characteristics.

a) It occurs quickly on contact with rubber products.
b) It produces redness, wheals,flares (contact urticaria) at the contact site or else where and may cause an asthma attack.
c) It can produce anaphylactic reaction.
d) It is caused by the naturally occurring proteins and not by the compounding ingredients.

Much speculations are being made about the sudden occurence of the protein allergy problem, when latex surgical gloves, catheters, condoms etc have been in use for more than thirty years. The correct answer to this question will be difficult to establish. But some of the speculations that have been made need to be considered.

Firstly, the idea that some drastic changes in the protein composition have been taken place within the rubber tree due to new clones,changing plantation practices etc.It is to be noted here that the development of new clones and their acceptance by the plantation sector can not taken place all on a sudden. As such even if some minor changes have occurred in the composition, it may not be that significant. Clonal variations in latex protein have been studied by Yeang[4] and more recently by Kekwick using SDS- PAGE. Some clonel differences can be detected but the similarities in serum protein compositions are more striking than their differences. So the present results do not really support the idea that clonal variations cause the current problem.

Secondly there is the suggestion that the cause of the problem is in some way related to the use of yield stimulants on the plantations.This also seems to be unlikely because the yield stimulants of various types have been in use since 1955 so that this problem might have been encountered many years ago had the use of stimulants been involved.

More probably, the problem has been resulted from the recent very high usage of latex goods like examination gloves and condoms following the AIDS scare. It is also to be observed that the latex sensitivity problems are reported from U S A and Eu-

rope, though latex and latex products are mainly produced in Asian countries. Improved diagnostic facilities available in USA and European countries and the fact that Western populations appear to be increasingly prone to allergies of almost every type are the factors to be reckoned with.

I.3 ORIGIN OF PROTEINS IN LATEX

Fresh NR latex from Hevea Brasiliensis is a complex colloidal dispersion of rubber particles in an aqueous medium.[3] The colloidal system is stabilized by the non rubber materials present in it. The non rubber constituents include proteins, lipids, phospholipids, carbohydrates, amino acids and inorganic ions. The total solids content of NR latex is normally 30 — 40%.[4] The non rubber content is 3 - 4 % of which 0.95 % is proteins.

The most important group of non rubber material is protein of which a large number is present exhibiting a wide range of molecular weight. Fresh latex, when Ultracentrifuged at higher speed can be separated into three major fractions,[5] a top rubber fraction, a particulate C serum and a dense bottom fraction. An yellow orange layer containing the Frey_Wyssling particles in latex is separated immediately above the C serum. Fig (2).

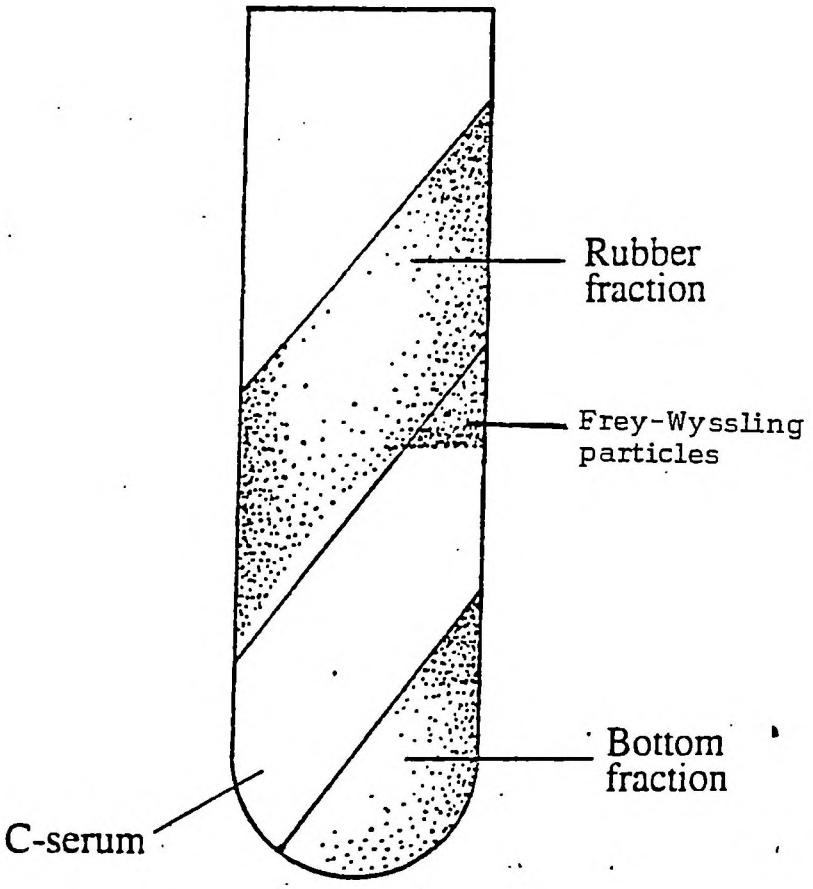


Figure 2. Centrifuged fresh Hevea brasiliensis *latex.*

Table. 1.

Composition of NR Latex

Rubber content	--	30 - 40 %
Proteinous substances	--	1 - 1.5 %
Resineous substances	--	1 - 2.5 %
Sugars	--	Upto 1%
Ash	--	Less than 1%
Water	--	55 - 60 %

Of the total protein content in fresh Hevea latex,[5,6] 27.2 % was strongly adsorbed on to the rubber fraction, 47.5 % in the serum fraction and 25.3 % in the bottom fraction. The rubber adsorbed proteins have a major influence on the colloidal stability, but the serum phase proteins are thought to have little technological significance.

Attempts to characterise the latex proteins was reported from 1930 onwards. Bondy and Freundlich[7] have separated two proteins from centrifugally separated serum of ammonia preserved NR latex which are protein A & B having isoelectric points of 4.55 and 3.9 respectively. The former is insoluble in water and alcohol while the latter is soluble in water and 70 % alcohol.

Kemp and Straitiff[8] isolated three proteins from ammoniated latex serum by ammonium sulphate precipitation method - protein A, B & C.

Archer and Sekhar[9] detected seven electophoretically distinct protein compounds from aqueous serum of unpreserved NR latex obtained by high speed centrifuging and by freezing at $-25^{\circ}C$

The different proteins that had been studied in detail are ∝ globulin, Hevin, fibrillar protein and basic protein. From freeze dried serum solids, Archer and Cockbain[10] separated ∝ globulin by an isoelectric ammonium sulphate precipitation method. It has an isoelectric point 4.8 and molecular weight of the order of 200 kDa. The similarity in the electrophoretic and colloidal behavior between dissolved ∝ globulin and the particles of Hevea latex suggest that this protein is an important component of the protein layer which is absorbed on the rubber particles. The nature of ∝ globulin corresponds to 'Protein A' reported earlier[7,8]

Hevin is the second important protein, which is dissolved in latex serum and isolated by ammonium sulphate fractionation of cold aqueous extract of the freeze-dried solids derived from the so called bottom fraction[11] It has a low molecular weight of

10kDa and isoelectric point of 4.5. It is water soluble at all pH.

Leutoid particles[5,12,16] in young latex vessels contain a protein deposited in the form of bundles of microfibrils, each having a double helical structure and have a lower isoelectric point than Hevin.

The major soluble basic protein of B serum is Hevamine which remain in solution after dialysis and there are two types of Hevamine, Hevamine A & B.

Paper electrophoresis studies[13], ion-exchange chromatographic and starch gel electrophoretic studies[14] of Bottom fraction, Starch gel electrophoretic studies of C serum proteins[15] have detected several protein components. Recently it is shown that proteolipids are associated with the rubber particles in latex.

Ammoniation of field latex prior to concentration starts hydrolysis process particularly of glycolipids, phospholipids and probably some proteins.[16,17] Lipid hydrolysis is completed after one month, but other changes may continue for a longer period. During the process of centrifugation of preserved field latex into centrifuged latex, a portion of the protein along with the non rubbers get removed in the skim latex. Those proteins remaining in the latex have been associated with some of the properties

of latex and latex products.

Eventhough a lot of studies have been carried out on proteins in NR latex, the volume of work on proteins in concentrated latex is much less despite the fact that concentrated latex is the main raw material for almost all latex products. Recently the studies on proteins present in concentrated latex has been intensified [18] and such studies are very relevant in the present circumstances because some proteins in NR latex gloves have been identified as source of some allergic problems in sensitive human beings.

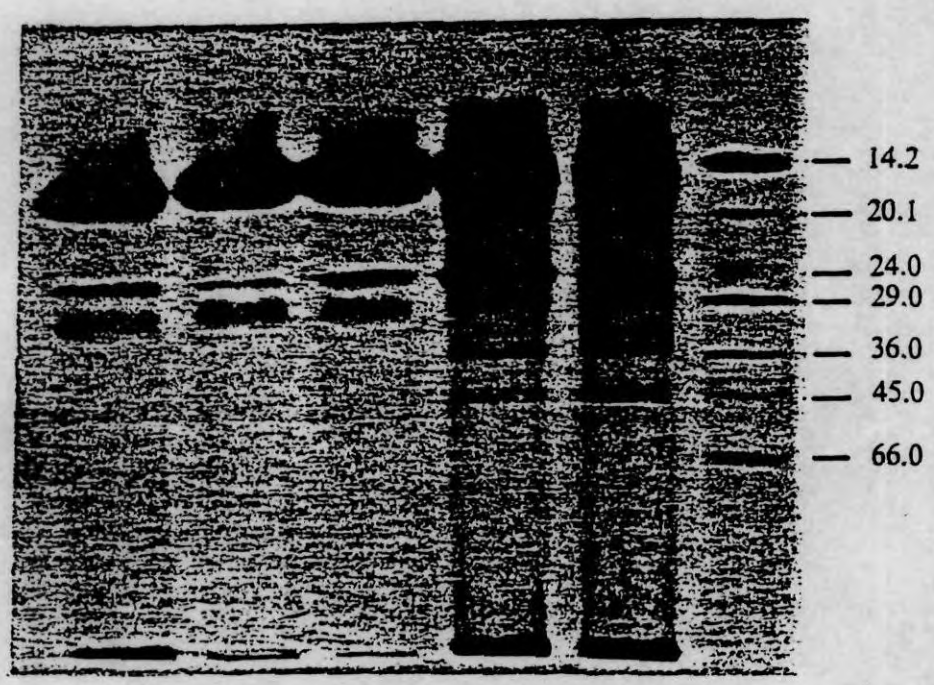
Proteins in HA latex concentrate [18] are distributed between two main fractions, serum fraction and rubber fraction. The serum fraction contain six proteins and had molecular weights 14,24,29,36 & 45 kDa and another protein of molecular weight greater than 1ØØ kDa. (Fig.3). They form a group of proteins which are most likely to be present in aqueous extracts of latex products. They are most readily leached out during processing.

Proteins associated with the rubber particles (RP) are mainly the 14 kDa with a minor 24 kDa protein. they are presumed to be loosely bound to the RP membrane and were partly extractable by ammonia solution. A greater portion was strongly bound to

RP membrane and require a detergent solution (SDS) or organic solvent (Chloroform- methanol). The ammonia extracts on analysis (SDS_PAGE) showed a 14 kDa and a 24 kDa proteins. A 2 % SDS solution extract on analysis showed one prominent protein band at 14 kDa which may again be the 14.6 kDa protein, and two less prominent bands at 24 kDa and 29 kDa. (Fig.3). A similar extraction of serum free RP of Hevea Brasiliensis latex by Dennis and Light [19] showed a minor 24 kDa protein and a major 14.6 kDa protein which they termed "Rubber Elongation Factor" (REF). A more selective extraction of RP proteins can be obtained with Chloroform-Methanol. [19] These proteins associated with RP would be expected to remain in the leached latex products. Thus any influence of proteins on properties of latex products would be largely attributed to these proteins. Therefore studies are more centered on the proteins associated with the RP, since these have been reported to be responsible for the anaphylactic reactions in sensitised people.

I.4 EXTRACTABLE PROTEINS (EP) FROM LATEX FILM

On processing fresh latex into HA concentrated latex, protein content reduces from 30 - 50 mg/g rubber to 20 -26 mg/g



A. SDS-extracted proteins of RP from 30-day-old RRIM 729
B. Proteolipid fraction of RP from 35-day-old RRIM 600
C. Glycoprotein fraction of RP from 44-day-old PR 255
D. Ammonia-extracted proteins of RP from 48-day-old RRIM 701
E. Serum proteins of 48-day-old RRIM 701 concentrate
F. Molecular weight markers (kDa)

Figure 3 SDS-PAGE of proteins from the rubber and serum fractions of HA latex concentrates.

rubber with the proteins getting distributed between the rubber and serum phase.[16,17] Qualitatively the proteins retained on the rubber particles in the concentrate are more or less the same as those in fresh latex, but quantitatively they differ owing to extraction of some of the proteins by ammonia. However, serum proteins differ greatly.

Of the total proteins in a latex, only a small portion is water soluble. Whereas the total protein content of HA latex concentrate is 20 - 26 mg/g rubber, the EP is only about 0.04 mg/g rubber. Compounding increases the level of EP to 0.14 mg/g rubber which on heating get further increased to about 0.34 mg/g rubber.[21] The observations indicate that the presence of soap, Potassium hydroxide and ionic species in the latex compound plays a role in the extractability of proteins. The nature of the proteins also contribute to the extractability.

I.5 WHY PROTEINS ARE BELIEVED TO BE THE CAUSATIVE

It was A.F Nutter[22] who first reported in 1979 that contact urticaria could be caused by latex itself, contrary to the belief that latex compounding ingredients present in gloves were respon-

sible for it. The main evidence for the above findings were standard dermatological tests.[23,24] (eg.: Skin prick tests, patch test etc.) Use of in vitro RAST (Radioallergosorbent test)[57] procedure also showed that latex proteins were allergens.[25]

The first report[26] of an anaphylactic shock caused by the use of surgical gloves come from Finland in 1984, The syndrome of it can be described as follows:-[27] exposure to protein allergens in certain individuals leads to response in their immune system which brings about a condition of hypersensitivity known as anaphylaxis. (Greek ana = against, phylaxis = protection) On subsequent exposure, the interaction of the allergens with antibody formed in the sensitizing phase culminates through a complex mechanism in a systemic release of histamine and other amines. The main effect of hystamine in the circulation is to dilate the peripheral vessels, with a consequent severe fall in blood pressure and anaphylactic shock.[27] Among the effects seen are speeding of heart rate, breathing difficulty, urtricaria, fluid release in the tissues and unconsciousness. Severity and danger of anaphylaxis have also been reported in a number of publications.[23,28,29,30]

However it is pertinent to note here that though the effect

of anaphylaxis may be severe to the individuals suffering from it, considering the very large number of the users of latex products world wide, persons getting affected are only a minority.

Between 1979 and 1986 Morales et al[29] studied six patients with anaphylactic shock caused by gloves and balloon contact and published strong evidence that proteins in these articles were responsible.Since 1986 more than 40 papers have been appeared and the number continues to increase. From these papers, dipped products causing Type I allergic reactions can be said to include gloves,(house hold, surgical, examination) condoms, toy balloons, cuffs on enema administrating devises, dental coffer dams etc.

All works done in this area show that there are a number of allergenic proteins of varying molecular weights. "Electrophoresis (SDS-PAGE) of B- serum, C- serum and rubber particle membrane extracts by E Sunderasen et al.[31] revealed the difference in the protein composition of the various extracts. Fig. 4. C serum showed the most numerous bands ranging from 5 kDa up to 100 kDa. B serum has fewer in number but discernible bands appeared at 14, 20, 25, 30, 35 kDa and less prominent bands at 43 and 57 kDa. Two major components of rubber particle membrane extract were detect-

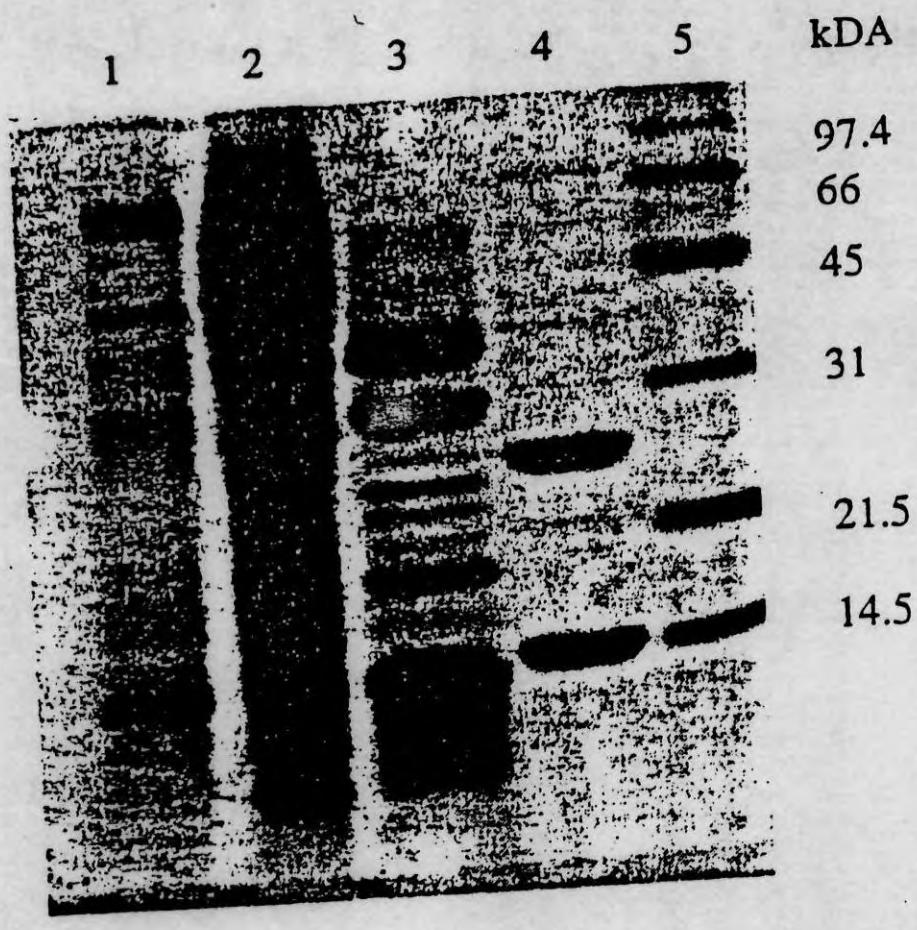


Figure 4 SDS-PAGE of proteins from various fractions of latex (15% gel). Lane 1: leaf extract, 2: C-serum, 3: B-serum, 4: rubber particle extract, 5: molecular weight markers.

ed at 14 kDa and 24 kDa respectively.

Immunobloting of the SDS - PAGE with eleven individuals and two pool plasma IgE positive to latex protein demonstrated the presence of allergens in all the Hevea extracts examined although there were notable differences in each individuals response to the extracts.

Nine out of the eleven patients were found to be positive to B serum while positive bands were also observed in C serum from eight patients. Rubber particle membrane proteins have the least allerginity , with only three patients found to be positive. Both the pool plasma tested were positive to B serum proteins while only one of them reacted to a C serum protein.

The immunoblots also revealed the presence of some allergens that were recognised by IgE in many of the plasma samples tested. The most frequent bands observed in B serum blots were in the region of 43 kDa to 65 kDa and with a thick smear at 58 kDa. The majority of the C serum positive patients had a IgE binding to a thick band in the region of 14 kDa. Slater and Chabra[32] described a 14 kDa protein to be a major allergen among U.S spina bifida patients. Anti bodies against a 14 kDa protein were also found frequently among the Finnish patients with congential anomalies and also among adult latex allergic patients. In view of the

numerous molecular weight (14 kDa and below) polypeptides that have been reported to be allergenic,[33, 35] the possibility exists that some of these could be the break down fragments of larger proteins." The above observations also point out why difficulties are experienced in comparing the results on what is apparently the same material (eg. in extracts of examination gloves from a manufacturer) let alone from different sources. It should be surprising for three reasons. Firstly, individuals may be allergic to different extents (including not allergic at all) to different proteins. Secondly, the protein composition of commercial concentrate can not be considered strictly invariable. Thirdly, the kinds proteins to be found in dipped goods might depend critically on the manufacturing process[39] and production of safe products is not impossible.

I.6 EXTRACTABLE PROTEIN FROM LATEX DIPPED PRODUCTS

NR latex dipped products contain two kinds of proteins _ that tightly bound to the rubber particles and the soluble serum derived proteins remaining after the leaching / washing during manufacture. The latter give rise to the most of the EP.

I.7 EXTRACTABLE PROTEIN FROM NR LATEX GLOVES

A major demand for NR latex concentrate is in the manufacture of latex gloves for examination, surgical and domestic use. The global demand for NR latex examination gloves in particular has increased significantly in recent years with the rise in the incidence of the acquired immuno deficiency syndrome (AIDS) infection.

A lot of work had been done relating the EP from latex gloves after the report of the anaphylactic shock occurring in sensitised persons by the proteins present in the gloves and a number of research papers had been published.[34,35,36,37,38] E Sunderasen et al[35] conducted a two dimensional (2D) immunoelectrophoresis and Western-immunoblot to investigate whether the water soluble antigens from latex examination gloves originated from the RP membrane protein, the C serum or the bottom fraction.Fig 5(a-f). They found that latex B serum proteins featured strongly as antigens in the proteins extracted from latex gloves.

Recently, A R Shamsul Bahri et al[36] investigated that the cause of soluble proteins is eluting mainly from the inner surface of latex gloves as compared with the outer surface. It was

Figure 5a. 2D-Immunoelectrophoresis of glove eluate.

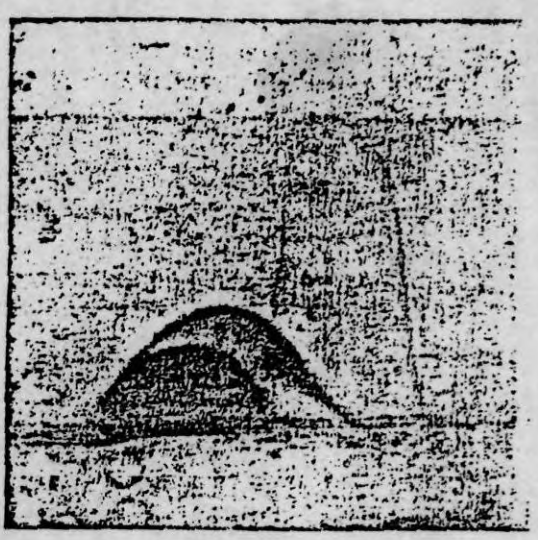


Figure 5b. 2D-Immunoelectrophoresis of high ammoniated (HA) latex serum.

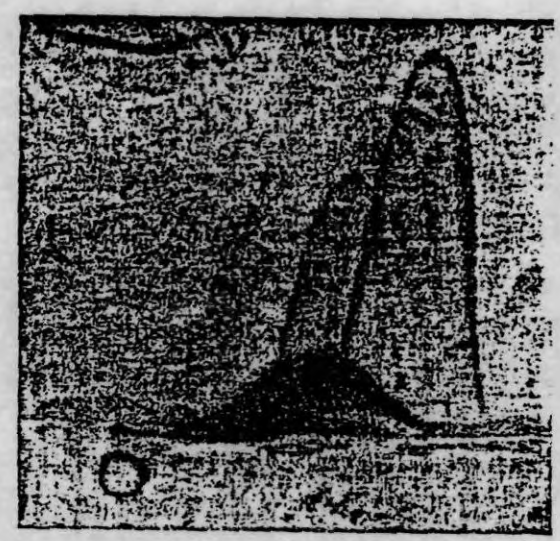


Figure 5c. 2D-Immunoelectrophoresis of pre-vulcanised latex serum.

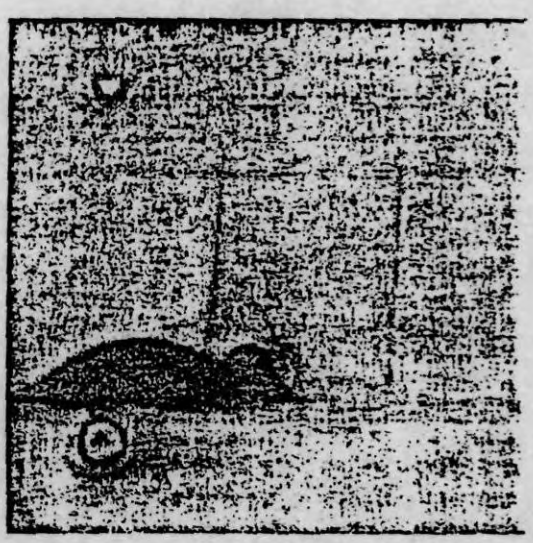


Figure 5e. 2D-Immunoelectrophoresis of C-serum.

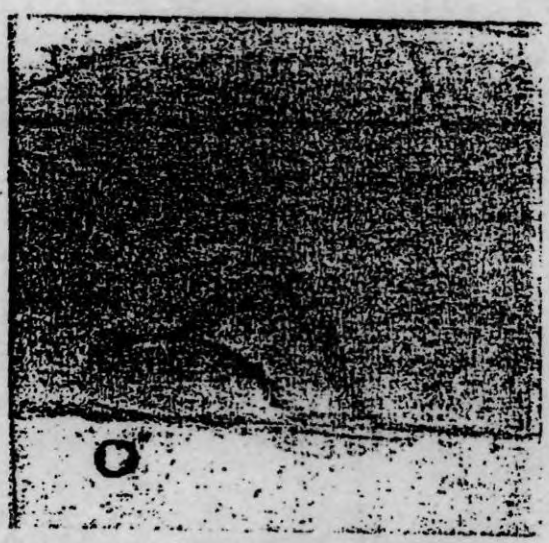


Figure 5d. 2D-Immunoelectrophoresis of B-serum.

Figure 5f. 2D-Immunoelectrophoresis of rubber particle membrane proteins.

suggested that differential leaching of proteins from the two glove surfaces was not due to the existence of a barrier to protein diffusion. It was best explained by the migration of proteins to the inner glove surface during manufacture.

Studies by Cardosa et al[39] introduces to rubber manufacturers the concept of enzyme immuno assay and describes various immunoassay formats by which antigens can be detected. The paper describes a number of examples which show that glove produced by different manufacturers contain different amounts of antigens and monoclonal antibodies are used to show that different antigens are found in different gloves.

I.8 WHY PROTEIN REMOVAL IS ESSENTIAL

As it has been fairly well established now that the cause of the immediate type allergic reaction to latex products is the presence of EP, It must be theoretically possible to prevent allergenity by extracting the protein from the products during manufacturing. However no extraction process is 100 % effective and even the minute traces of the allergens may cause allergic reaction to already sensitised persons. What is essentially required now is to reduce the level of extractability of proteins

from latex products to the lowest possible level so that no new person become sensitised by contact with such products.Studies show that [34] higher EP contents are always associated with positive allergic responses, while very low EP levels tend to exhibit weak or no allergic reaction. No threshold levels have been established.

THe Food and Drugs Administration (FDA),U.S.A issued an advisory letter in May 1991 to manufacturers of latex devises suggesting different steps to minimise the EP level from latex goods. Steps which can help the production of low EP latex goods include the use of low protein latex concentrate for the manufacture of latex products, proper choice of compounding ingredients,proper leaching of the products, imparting special treatments like chlorination, surface coating etc.

Firms wishing to obtain permission from F D A to market surgical gloves in the United States must supply F D A with data from a Primary Skin Irritation Study and Dermal Sensitization Study. Before Feb. 1993 , these studies were required only for surgical gloves. Because all medical devices must be demonstrated to be biocompatible, these studies are now required for patient examination gloves also.

I.9 REMOVAL OF PROTEINS FROM LATEX PRODUCTS

The amount of proteins present in a latex concentrate appears to be constant; but the amount of EP remaining in a dipped latex product is highly variable.[39] This variability exists due to the differences in the manufacturing process in various factories.

H Hasma[6] has studied the changes in the composition of proteins from fresh latex to HA latex to compounded latex and pre vulcanised latex. Proteins are analysed by iso electric focussing and SDS - PAGE electrophoresis. The proteins undergo some changes on processing the latex products leaving mainly the anionic proteins of PI between pH 3.5 to 6.Ø and molecular weight 6.Ø to less than 14.Ø kDa from the major extractable proteins from the examination gloves.

Centrifuged latex concentrate has a total protein content of about 16 to 2Ø mg/g rubber.[21] Doubly centrifugation reduces the total protein content by another 25 - 3Ø % when freshly prepared. The EP content of singly centrifuged latex concentrate is only about Ø.5 mg/g rubber. This is considerably lower than in fresh latex film. When stored before filming, the EP value increases and can reach 1 mg/g or slightly higher. Double centrifuged latex

shows a decrease in EP content. The very low values of EP content for double centrifuged latex is believed to be due to the removal of ions from the system thus making the proteins less soluble.

On compounding the latex concentrate, the EP content increases, probably due to the presence of soap,KOH, and other ionic species in the system. Freshly compounded latex can have EP values twice that of uncompounded latex. On maturation the EP values can decrease or even increase depending on experimental conditions.

Vulcanisation can be carried out either by heating the latex to give pre vulcanised latex or by heating the dipped film from the compounded latex (post vulcanisation). Heating compounded latex to produce pre vulcanised latex increases the EP content. The extent of increase depends on the formulation, the time and the temperature of heating.

Leaching of latex film reduces the EP content. [40,41,42,43,44] But the extent of reduction is dependent on the conditions used in leaching. Leaching can be carried out on the wet film (wet gel leaching) or on dry film (Dry gel leaching). Wet gel leaching is quiet effective for pre vulcanised film where the latex already has high EP content.

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There are three approaches to decrease the EP content in latex products:- pre processing,processing and post processing.

I.9.1. Pre processing

Ammoniation and preservation of latex degrades the NR latex proteins. A minimum time is essential before the maximum degradation process. There is evidences that ammoniated hold time is most effective for protein degradation before any compounding has occurred. Single, double and triple centrifugation process decreases the total protein content. Enzyme treatment with alcase, papain or endogeneous proteotype enzymes also decreases the protein content. The resulting increased cure time and potential loss of some of the physical properties limits the use of this approach.Use of the surfactants during centrifugation helps to dis__place hydrophobic proteins from the poly isoprene particles and mobilize proteins facilitating migration during processing.

Use of de-protenised latex (DPNR) by enzyme treatment and centrifugation or by pre vulcanisation and centrifugation for product manufacturing can reduce the EP content.

A substantial amount of water soluble protein can be generated during the compounding of latex and upon heating of the

latex compound (pre vulcanisation). By a process of re-centrifugation of diluted prevulcanised latex, it is possible to remove a large amount of water soluble proteins from the prevulcanised latex.[45]

Thus the use of a low protein latex[46,47] will effectively reduce the level of the EP content from latex products.

I.9.2. Processing

Leaching process is critical. Leaching is the process of removal of hydrophylic materials from latex dipped products by washing them in water. It is an essential process for the manufacture of dipped latex products. The removal of excess of the coagulant and the added compounding ingredients, water soluble non- rubbers including proteins etc results in the improvement of physical properties such as Tensile strength, film clarity, prevention of surface blooms and reduction in water absorption of dipped products.The effectiveness of leaching process is critical in the determination of the overall quality of the product.

It is interesting to note, that a non leached oven dried film is lower in EP than a slightly leached oven dried film. It is apparently essential to hydrate the film to provide both swelling

within the matrix and transport medium for migration during the heat curing process when the matrix contracts. Utilization of hotter, longer, fresher leach water [44] addition of ammonium sulphate, surfactant etc affect the level of proteins in the product.

There are basically two methods of leaching, wet gel leaching [41,43] and dry film leaching. The former involves the washing of the wet gel, ie the gelled deposit on the former,prior to drying and vulcanisation.It is usually carried out on-line.Dry film washing is an off-line process and consists of washing of the dried vulcanised latex product after removal from the former.

In the production of latex examination gloves, [42] wet gel leaching is often carried out for a period of several minutes,1 to 5 minutes in a continuous chain dipping line. the actual leaching time is very much dependent upon the design of the dipping unit.

The various leaching parameters like temperature and flow rate of leaching water, time of leaching, TDS (total dissolved solids) and hardness of leaching water etc can affect the residual protein content in latex gloves. [49] The thickness of the film also affect the residual protein content in the products.

The extent of cure after leaching may affect migration over time. Oven temperature do affect the EP content despite the optimal cure.

I.9.3. Post processing

Leaching and chlorination are both very effective. Enzyme treatment has some promise,however one must also be alert to the allergic reactions to the enzymes over time. Coating of NR latex products with non permeable substances is possible.Lubricants like silicone help to reduce the migration of proteins. Petroleum products (petroleum gel, mineral oil etc) utilized as lubricants or as skin protectants should be avoided when used in contact with gloves, condoms or other protective devices as they break down the latex compromising barrier integrity. Autoclaving degrades the allergens but also decreases the stability of the gloves.

I.9.3 a. Chlorination

It is an off-line process and is a very effective approach for reducing soluble proteins. It is believed that chlorine renders the proteins insoluble (denaturing) or it forms an impermeable barrier that prevents proteins migrating to the sur-

face. It is also used where the usage of power is not suitable, as it enhances the smoothness of the surface. However the process changes the properties and appearances of the products.

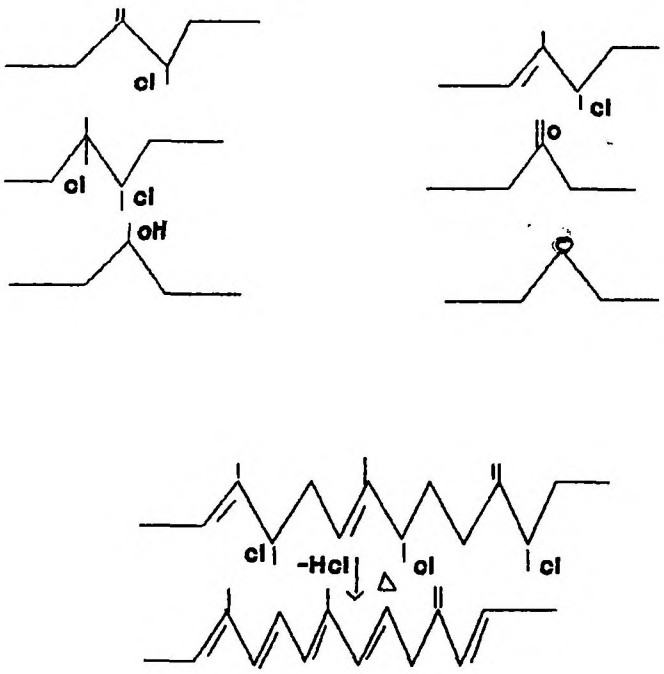
Chlorination of surgical and examination gloves [47,48,49] is now being increasingly carried out due to the demand for power free and low protein containing gloves. As these products are extremely thin, careful control of the process is required.

The chemistry of aqueous chlorination of NR surfaces [20] has been discussed extensively. Various reactions are occurring during chlorination. When Hydrochloric acid is added into a solution of hypochlorite, chlorine is released. In an aqueous system, chlorine react with NR via an ionic mechanism to form carbonium ions. This carbonium ions can react by addition of chlorine, cross linking with each other, cyclisation and oxidation depending on pH of the solution.

Basically the possible products that are formed after chlo rination can be shown in a simplified form as shown in Fig 6.

Chlorination of gloves and other latex products involves only a surface reaction in the initial stage. Depending on the thickness of the glove, the time of chlorination and the concentration of the chlorine solution, the reaction can proceed to the inside.

Fig.6 - The possible products that are formed after chlorination



The colour change is due to the loss of HCl and formation of conjugated vinylidene unsaturation as above.

After chlorination, neutralisation and washing,the products would have to be heated or tumble dried. Extreme care should be taken to ensure that no over heating occurs. Over heating of chlorinated gloves will result in colour changes and poor properties.

I.8.3 b. Dry gel leaching

Since heating increases the EP content, latex films leached after drying have lower EP content than wet gel leached films for short leaching times.However, on prolonged leaching, the difference between wet gel leaching and dry gel leaching become narrower.

For short leaching times,usually prevalent in gloves industry, dry leaching is more effective. The dry leached gloves should be further dried at lower temperature. However a combination of wet gel leaching and dry leaching is more preferable method.[42]

During leaching, the extraction of soluble proteins is rapid initially, but the rate of extraction decreases quickly and levels off after some time. Slow leaching of proteins can continue for several hours ,but further reduction in EP content is small.

I.1Ø. ESTIMATION OF EP FROM LATEX GLOVES

Many methods are used for the quantitation of proteins and they include 1) use of UV absorbence at 28Ø nm, 2) colorimetric analysis (Lowery,[52] Bradford[53] or Biuret assay), 3) High Performance Liquid Chromatography (HPLC)[54] The Lowery and Bradford assays are comparable in their sensitivity where as the Biuret assay and UV absorbence are respectively about 5 and 25 times less sensitive than the Lowery assay and hence are less suitable for routine assays in view of the very low glove protein normally encountered (of the order of µg of protein per gram glove. HPLC method eventhough gives highly reproducible results,is time consuming and involves the use of costly instruments. The Lowery and Bradford methods involves the extraction of proteins from the product, purification by acid precipitation and colorimetric assay. Extraction of soluble proteins is effected by eluting two pieces of glove sample of 7cm X 7cm in 5Ø ml distilled water.

Recently, the latex ELISA test for antigenic proteins (LEAP)[55] have proven to be very useful for the quantitation of the level of antigenic proteins in latex extracts.[56] The extraction of proteins for this type of testing is normally carried out in buffered solutions in order to increase protein stability.

I.11. SCOPE OF THE PRESENT WORK

HA centrifuged latex is being used for the commercial manufacture of gloves in our country. A wet gel leaching of 1 to 2 minutes at a temperature of 5Ø - 8Ø C is commonly employed. This will not effectively remove the proteins to lower levels of EP content. The level of EP content in commercial glove samples manufactured in our country is found to be ranging between 4ØØ - 9ØØ mg/Kg. Table 2. So additional treatments should be given to reduce the EP content.

On line treatments or treatments during processing like changing the flow rate of leaching water, hardness and total dissolved solids (TDS) of water etc can decrease the EP content to a certain extent,[44] but is not sufficient. Other treatments like increasing the wet gel leaching time or using chemicals like chlorine solution etc is effective, but is not easy, as it requires alteration in the design of the plant and it is cost prohibitive.

Another alternative is the use of low protein lattices like deprotenised latex (DPNR), doubly or multiple centrifuged latex or recentrifuged pre vulcanised latex. This is very effective, but these type of low protein lattices are not being commercially available in our country.

So the easiest way to reduce the EP content in gloves and other latex dipped articles is the off- the line treatments or post process treatments.

As already discussed, some of the proteins remaining in the products are causing immediate type of allergy reactions to sensitised persons, [57] and since the effect of such hypersensitivity is very severe, the demand for gloves having low levels of EP is increasing. Therefore it is essential for the latex glove manufacturers to take all possible steps to reduce the level of EP content to the lowest level as possible.

Eventhough a lot of study had been done in reducing the EP levels and about the nature, activity and quantitation of the proteins in latex and products, only limited studies have concentrated in the off-the line treatments on products.

In view of the above, it was considered appropriate to take up a systematic study on the off the line treatments of gloves for reducing the EP content.

The present study includes,

i) the level of EP content in commercial gloves manufactured in our country,

ii) treatments on latex gloves for reducing the EP content. This include both leaching studies and surface treatments and

iii) treatments on latex films for reducing the EP content.

CHAPTER 2

EXPERIMENTAL

The materials used and the experimental procedure adopted in the present work are given in this chapter.

II.1. MATERIALS USED

II.1.1. Glove samples

Glove samples used in this study were supplied by M/s Anusham Rubber Industries Pvt Ltd, Shaga Nagar, K.K Nagar, Tamil Nadu.

II.1.2. NR Lattices for film preparation

Centrifuged NR latex (LA-TZ type) and double centrifuged HA type were obtained from M/s Pilot Latex Processing Centre, Chethakkal.

II.1.3. Chemicals

a) Calcium chloride - Used as the coagulant and was obtained from M/s Qualigens Fine Chemicals, Bombay.

b) Ammonia solution - Commercial grade (25 %) obtained from M/s

Laboratory and Industrial Chemicals, Madras.

c) Methanol - Commercial grade obtained from M/s Indian Drugs and Pharmaceuticals Ltd, Hyderabad.

d) Trichloro acetic acid (TCA) and Phosphotungstic acid (PTA) Both were used for the purification of proteins and was chemically pure grade obtained from M/s Qualigens fine chemicals, Bombay.

e) Folins Ciocalts Phenol Reagent - Obtained from M/s SISCO Chemicals, Bombay and was used for the colorimetric estimation of proteins.

f) Sodium carbonate - obtained from M/s Qualigens Fine Chemi cals, Bombay.

g) Copper sulphate - GR grade obtained from M/s Sarabhai M Chemicals, Baroda.

h) Sodium citrate - AR grade obtained from M/s E Merk (India) Ltd, Bombay.

i) Sodium hydroxide - obtained from M/s Qualigens Fine Chemicals, Bombay.

j) Sodium Lauryl Sulphate - A detergent and was obtained from M/s New India Chemical Enterprisers, Kochi.

k) Sodium hypochlorite - A 1Ø % solution, used for the preparation of Chlorine solution and was obtained from M/s Laboratory and Industrial Chemicals, Kochi.

l) Hydrochloric acid - AR grade obtained from M/s E Merk (India) Ltd, Bombay.

m) Rubber chemicals - Sulphur, Zinc diethyl dithio carbamate, Zinc oxide etc were commercial grade obtained from local sources. They were made into a 50 % dispersion in a ball mill. Potassium hydroxide and Potassium Laurate were used as 10 % solutions.

II.2. TREATMENTS ON GLOVES FOR REDUCING THE EP CONTENT

Off- the line treatments on glove samples were conducted in three ways. 1) Leaching technique, 2) Autoclaving or steam treatment under pressure and 3) Surface treatments like chlorination and siliconisation.

II.2.1. Leaching of gloves

Off the line leaching on gloves were done i) under alkaline conditions using 0.5 % ammonia, ii) using a detergent like sodium lauryl sulphate (SLS) and iii)using a solvent like Methanol.

II.2.1 a. Using Ø.5 % ammonia solution

The effect of varying time and temperature of leaching by ammonia solution on EP content from glove samples were studied. The ratio of glove sample : leaching solution used was 1 : 4Ø and the solution was gently stirred occasionally during the leaching process. The temperatures selected were room temperature, 5Ø $^{\circ}$C and 6Ø $^{\circ}$C. After the leaching, the glove samples were properly washed using water and dried at room temperature and protein estimation was done within days.

II.2.1.b. Using Sodium Lauryl Sulphate solution

The leaching studies was done at two concentrations, Ø.5% and 2 %. With each solution, the leaching was done at different temperatures for different time. The ratio of glove to water was

1 : 4Ø. After leaching, the samples were properly washed with water to remove any detergent if present and dried at room temperature.

II.2.1c. Using 50 % Methanol solution

Glove samples were leached in 50 % Methanol solution at different temperatures and time. The ratio of glove : leaching solution was 1 : 40 occasional stirring was given during the process. Proper washing was given and dried at room temperature.

II.2.2. Autoclaving

The effect of steam treatment under pressure on EP content and physical properties of glove samples were studied. The glove samples were autoclaved under 1 Kg pressure for different time intervals in an autoclave. After steam treatment, the samples were dried at room temperature.

II.2.3. Surface treatments

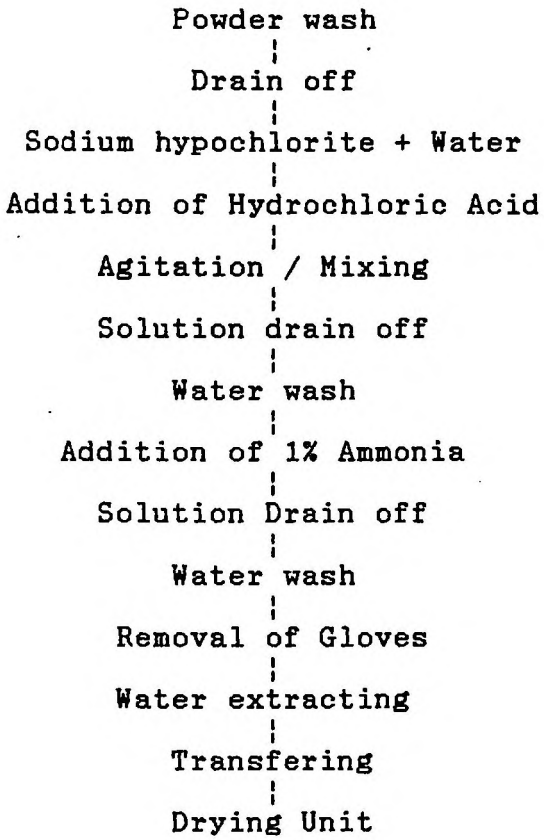
The surface treatments employed were chlorination and siliconisation.

II.2.3a. Chlorination

To study the effect of chlorination on the EP content, glove

Fig. 7.

Chlorination of gloves - Process flow chart



samples were subjected to chlorination to different levels. Chlorine solution was used for this purpose. It was prepared by

acidifying Sodium hypochlorite (1Ø %) solution with concentrated Hydrochloric acid. Chlorine solutions of different concentrations were prepared. The ratio of glove sample : chlorine solution was 1 : 5Ø. The experiment was done in a closed glass vessel.

The glove samples were first washed with water to remove the dusting powder present on it. Then it is immersed into the hypochlorite - water mixer in the glass vessel and then acidified with Hydrochloric acid. Proper agitation was given through out the process.The chlorination was done at different dosages for different time intervals. After chlorination, the glove samples were washed with water and then washed with a 1.Ø % ammonia solution to neutralise any acid present and again immediately washed with water to remove excess ammonia. The process can be diagramatically shown as shown in Fig 7. The gloves were then dried at a low temperature between 4Ø - 5Ø$^{\circ}$C. The remaining EP content on the glove samples were determined within one week.

II.2.3b. Siliconisation

Glove samples were treated with a 1.Ø % and a 1Ø % solutions

of silicone emulsion for 30 minutes at 60°C. It was then dried at 60°C in an air oven and the remaining EP content on the gloves was determined within days.

II.3. TREATMENTS ON LATEX FILMS FOR REDUCING THE EP CONTENT

II.3.1. Preparation of Latex compound

Centrifuged latex (LA-TZ Type) compound was prepared as per the formulation given in Table 2. Pre vulcanised latex compound was prepared by heating this latex compound at 60°C for four hours.

Compounding of the doubly centrifuged latex was also done as per the formulation given in Table 2.

Table 2

Formulation of Latex compound

Ingredients	LATZ Latex	DC Latex
	(Parts by weight)	
60% Latex	167	167
10% Pot. Hydroxide	1	1
50% Sulphur	2.5	2.5
50% 2DC	2.0	2.0
50% ZnO	1	1
50% Anti oxident	2	2

II.3.2. Preparation of latex films

Latex films (Ø.15 - Ø.2Ø mm thickness) were prepared by the coagulant dipping of glass plates in the latex compounds and vulcanising at 1ØØ°C for 1 hour.The coagulant used was a 15 % solution of calcium chloride. The films were lightly dusted with talc,stripped off from the formers, stored in polythene bags and analysed for EP content within few days.

II.3.3. Treatments

II.3.3a Wet gel leaching

The effects of wet gel leaching with varying periods of time and temperature on EP content were studied for pre vulcanised and post vulcanised films of LA-TZ latex and post vulcanised films of doubly centrifuged latex. Wet gel leaching was carried out on films dried for 5 minutes at 7Ø°C and the ratio of rubber : leaching water was 1 : 4ØØ.The water was gently stirred occasionally during the leaching process.

II.3.3b. Dry film leaching

Dry film leaching was carried out for LA-TZ and doubly centrifuged latex films by immersing the dry vulcanised films in distilled water at different temperatures for various periods of time. The proportion of rubber : leaching water selected was 1 : 400 and the water was occasionally stirred gently during the process.

II.3.3c. Wet gel leaching and dry film washing using 50% Methanol

Wet gel leaching and dry film leaching was carried out for both LA_TZ and doubly centrifuged latex films as described earlier using a 1 : 1 water _ methanol mixture instead of distilled water.

II.3.3d. Chlorination

Latex films prepared from centrifuged latex and doubly centrifuged latex were subjected to wet gel leaching at 60^{o}C for 5 minutes were used for chlorination. Chlorine solution was

prepared by acidifying Sodium hypochlorite solution with Hydrochloric acid as described earlier. The ratio of latex film : chlorine solution used was 1 : 5Ø. The films were immersed in chlorine solutions of different concentrations for three minutes at room temperature with agitation. The experiment was done in a closed glass vessel. After chlorination, proper washing and neutralisation was given as shown in Fig 7.The films were dried at 4Ø _ 5Ø°C in an air oven and the remaining EP content was estimated within one week.

II.4. ESTIMATION OF EP CONTENT

II.4.1. Extraction of total soluble proteins

Samples of two pieces measuring 7 cm X 7 cm and weighing about 1.5 to 2.Ø g each were cut from the latex gloves / films. Each piece was again cut into 16 pieces of equal size. The pieces were weighed accurately and extracted with 5Ø ml distilled water in a 1Ø cm diameter glass container at room temperature for 24 hours with occasional agitation. The extract was then centrifuged in a REMI Laboratory centrifuge R 8C at 3,5ØØ rpm for 3Ø minutes to remove any insoluble matter.

II.4.2. Protein purification and concentration from the extract

Proteins in the extract were purified and concentrated by precipitation with Trichloro acetic acid (TCA) and phosphotungstic acid (PTA).

To 6 ml of the protein extract, 1 ml of 35% (W/V) TCA was added and mixed. Then 1 ml of 40% (W/V) PTA was added, mixed well and allowed to stand for 20 minutes. The precipitated protein was then recovered by centrifuging the mixture in a REMI Laboratory centrifuge R 8C at 3,500 rpm for 30 minutes and decanting away all the supernatent. The very thin film of the precipitate was then redissolved in 2.6 ml of 0.1 M Sodium hydroxide for at least 20 minutes.

II.4.3. Protein estimation

Protein samples were assayed by the Modified micro Lowery Assay method.[51] The reagents were prepared as follows:-

Reagent A _ 6 % Sodium carbonate in 0.2 M Sodium hydroxide.

Reagent B _ 1.5 % Copper sulphate in 3.0 % Sodium citrate.

Reagent C _ (Working reagent prepared on the day of estimation) 5Ø ml of reagent A mixed with 1 ml of Reagent C.

Reagent D _ Folins - Ciocaults Reagent. Diluted 3 parts Folins with 1 part water.

The test reaction was carried out directly in the centrifuge tube. Added Ø.3 ml of Reagent C to the 2.6 ml of the test sample already present in the centrifuge tube. Mixed well and allowed to stand for 1Ø minutes.

Then added Ø.1 ml of Reagent D and mixed well in a cyclo mixer. The mixer was then allowed to stand for 3Ø minutes and the absorbance was read on the spectrophotometer,Shimadzu U V 24Ø at 75Ø nm. The concentration of proteins was read off against the standard protein used for calibration.(Bovine serum albumine, Ø -12Ø mg/ml.)

CHAPTER 3

RESULTS AND DISCUSSIONS

III.1. EXTRACTABLE PROTEIN CONTENT IN COMMERCIAL GLOVES

To study the level of EP content in commercial gloves, manufactured in India, glove samples were procured, tested for soluble or extractable protein content and the results are given in Table 3.

TABLE 3

Extractable protein content in commercial gloves

Extractable protein content (mg / Kg gloves)	Number of samples
Ø - 49	1
5Ø - 99	2
1ØØ - 399	19
4ØØ - 699	22
Above 7ØØ	13
Total	57

Out of the 57 samples tested, only three samples were having EP content below 1ØØ mg/Kg. Majority of samples were having EP content in the range of 1ØØ - 699 mg/Kg. This result shows that our manufacturers have to take special care to reduce the EP content in latex products.

III.2. EFFECTS OF TREATMENTS ON LATEX GLOVES

III.2.1. Effect of leaching

The effect of leaching on latex glove samples by Ø.5 % ammonia solution at different temperature and for various periods of time are as shown in Fig 8. FIg 9 and Fig 1Ø respectively shows the effect of leaching on glove samples by sodium lauryl sulphate (SLS) Ø.5 % and by 5Ø % Methanol _ water mixture. In all these three cases, the EP content is considerably reduced by the treatment. The extraction of EP content is rapid initially and levels off after some time. Slow leaching can continue. The reduction in EP content can be improved by leaching at higher temperature.

The effect of Ø.5 % ammonia solution and SLS solution (both Ø.5 % and 2.Ø % solutions) are almost comparable. But the Methanol - water (5Ø %) treatment is less effective even at elevated temperatures.

Also it was noted that the increased concentration of SLS solution will not improve the extraction of EP content.

The effect of leaching by the different solutions on the physical properties were also studied and given in Table 4. It was seen that the physical properties are not affected much.

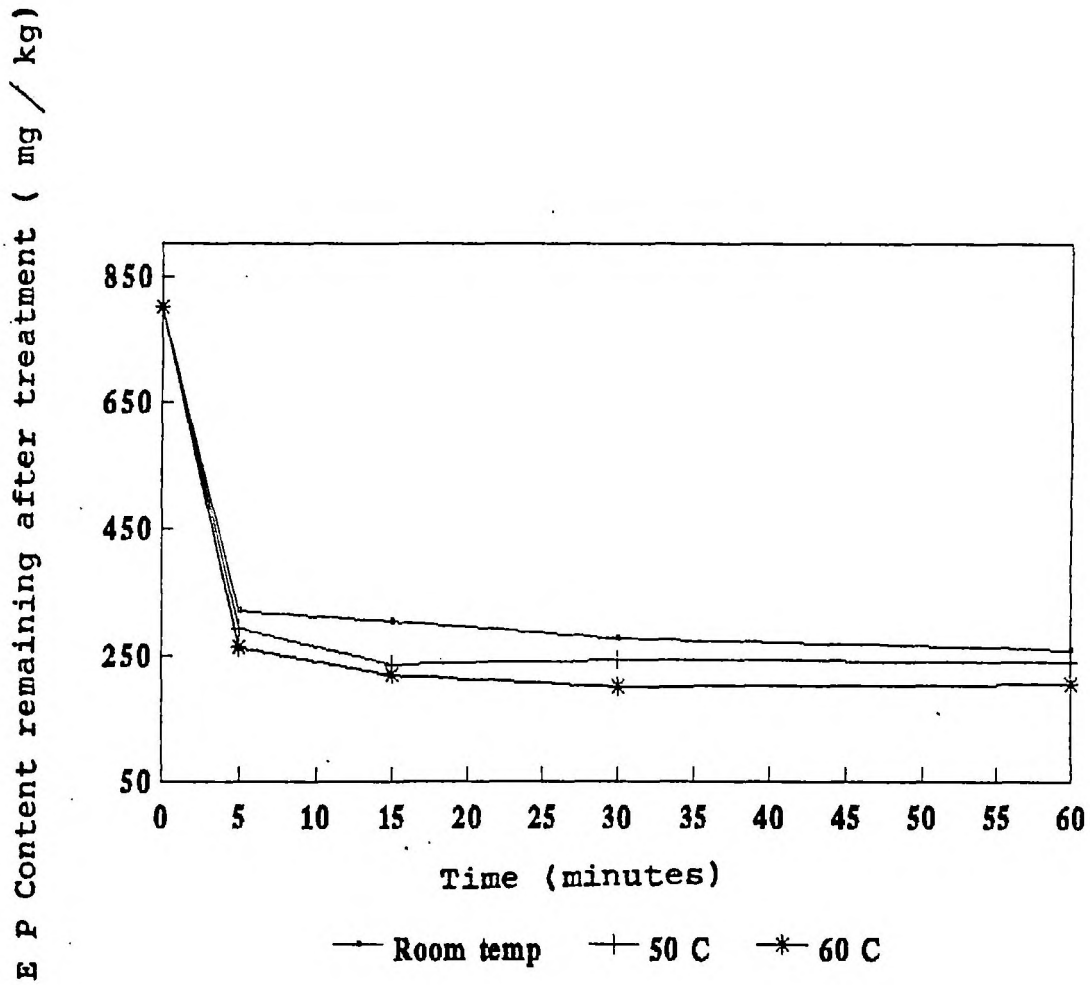


Fig. 8

Effect of leaching by 0.5% ammonia solution on E P content in Examination Gloves.

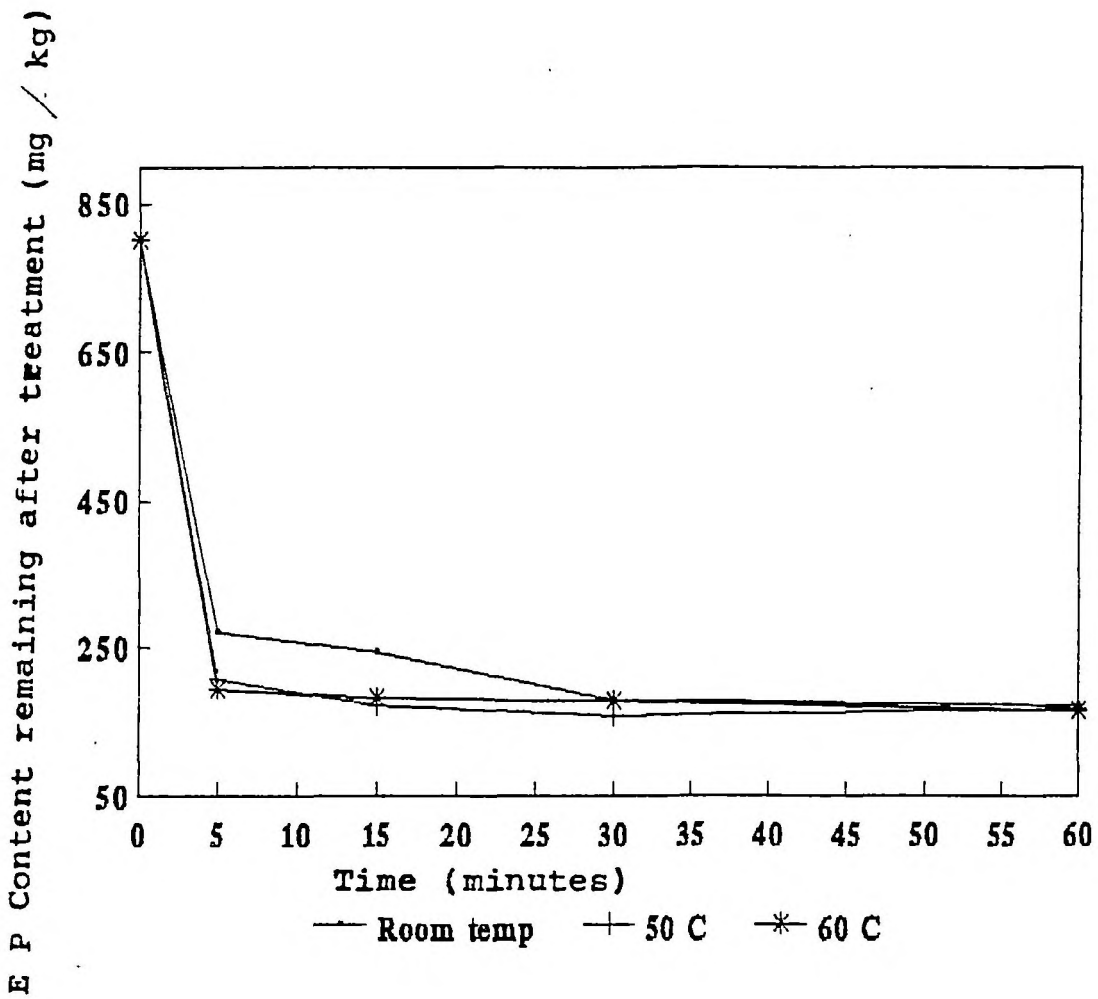


Fig. 9

Effect of leaching by 0.5% SLS solutiono on E P content in Examination Gloves

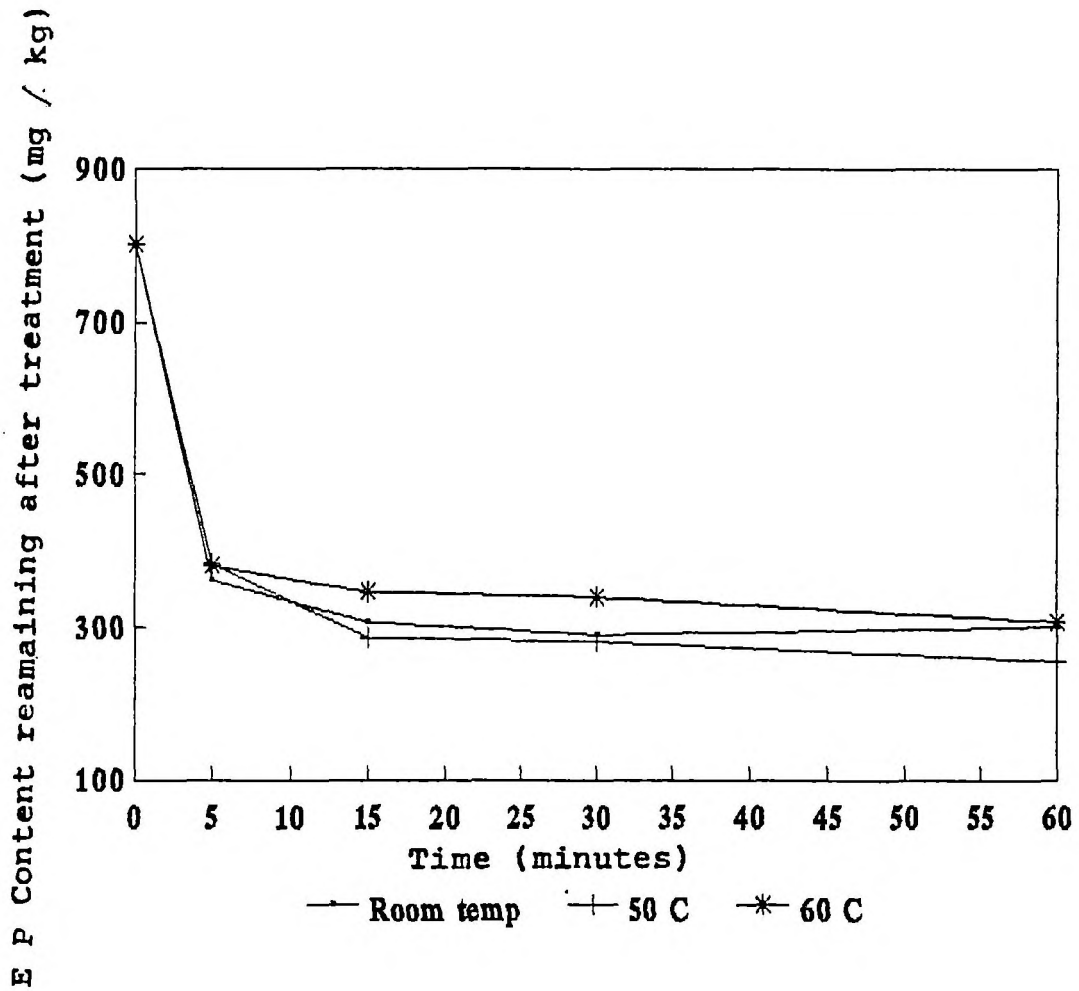


Fig. 10

Effect of leaching by 50% Methanol- Water mixer on E P content in Examination Gloves

TABLE 4

Effect of leaching on the physical properties of latex gloves

Sample details	Physical properties					
	Before ageing			After ageing*		
	Tensile strength (MPa)	Modulus 300 % (MPa)	E.B (%)	Tensile strength (MPa)	Modulus 300 % (MPa)	E.B (%)
Control	21.5	1.87	1290	18.6	2.01	1142
1hr leaching with 0.5% SLS	20.67	2.013	1294	17.4	2.01	1020
1hrleaching with 0.5% ammonia	20.31	1.866	1126	17.8	2.05	1045

* Ageing at 100°C/24 hrs.

III 2.2. Effect of autoclaving

The effect of steam treatment / autoclaving on latex gloves for removing EP content under a pressure of 1 Kg for different durations of time is given in Fig 11.

It was observed that the steam treatment is only very little effective in removing EP content from gloves. Variation in physical properties due to autoclaving was also studied. Table 5. The physical properties are only very little affected.

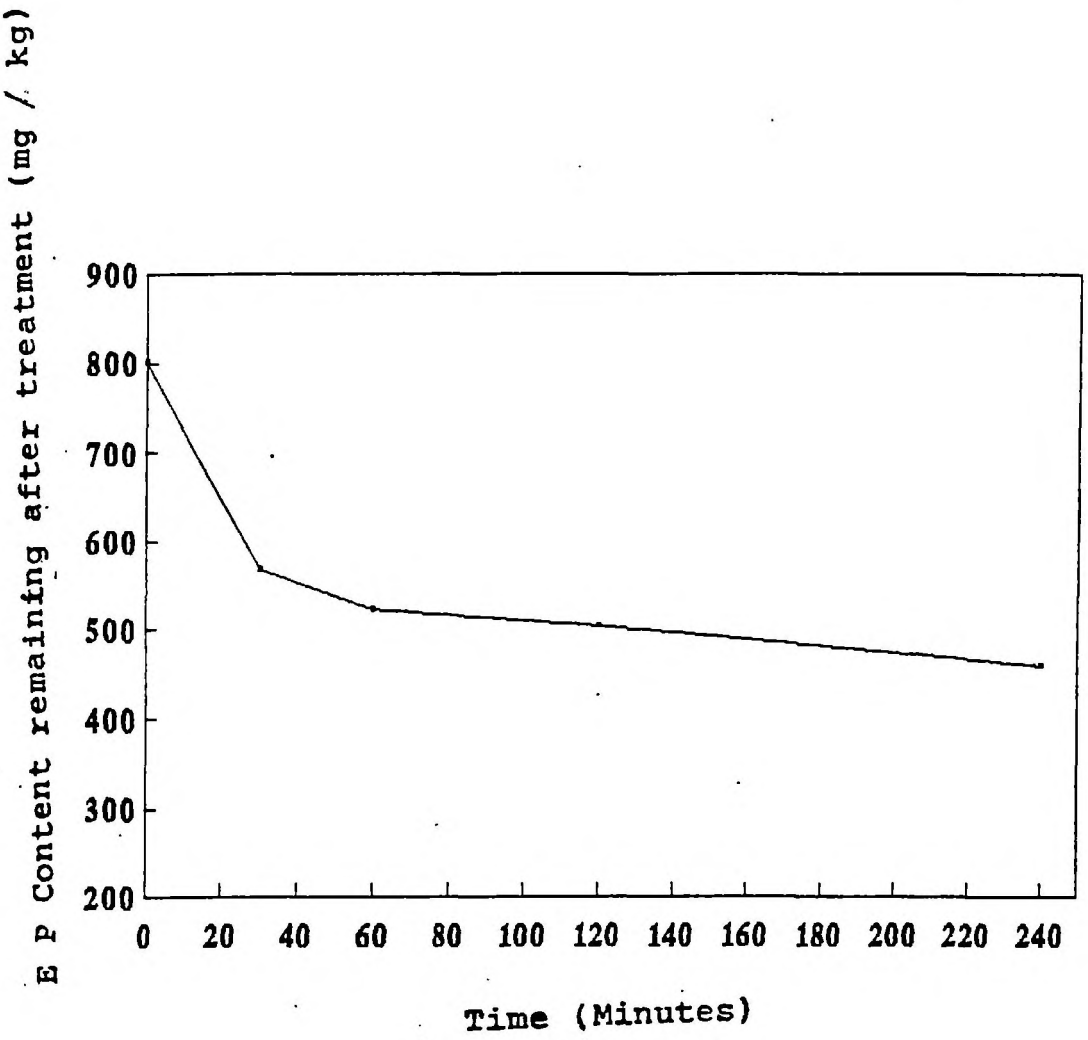


Fig. 11

Effect of steam treatment under pressure on E P content in Examination Gloves

TABLE 5

Effect of steam treatment on physical properties of latex gloves

(Autoclaving at 1 Kg pressure)

Sample details	Physical properties		
	Tensile strength (M Pa)	Modulus 300% (M Pa)	E.B (%)
Control	21.5	1.87	1290
30mts autoclaving	21.4	1.84	1210
60 mts autoclaving	20.6	1.71	1200
120 mts autoclaving	20.1	1.60	1190

III.2.3. Surface treatments

III.2.3.i) Effect of chlorination

The effect of chlorination on latex gloves using different dosages of chlorine solution are given in Fig. 12. Glove samples were treated with chlorine solution for different durations of time and its effect on EP content is as shown in Fig 13.

It was found that chlorination is very effective in reducing the EP content. Also the removal of EP is found to be less af-

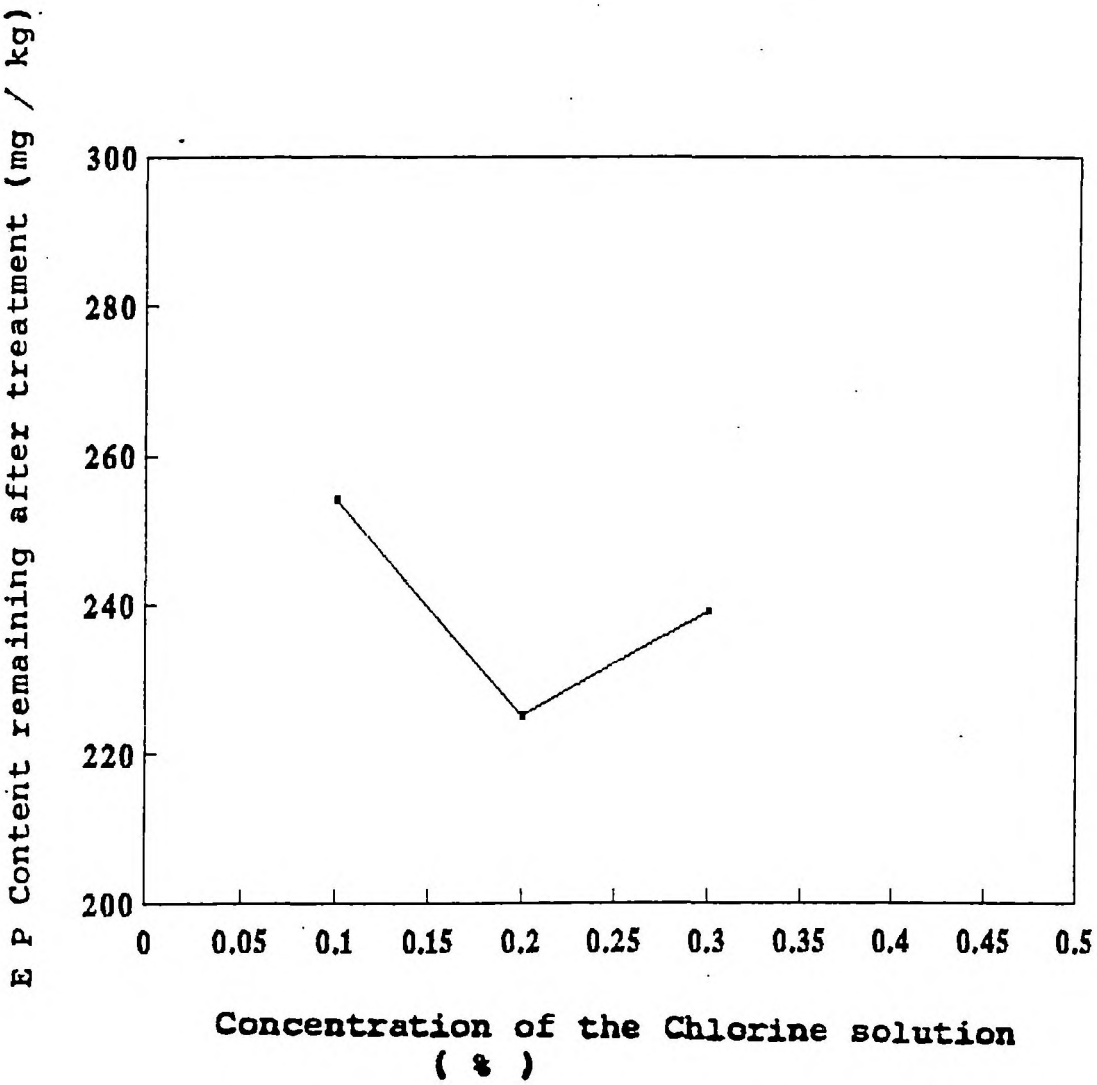


Fig. 12

Effect of Chlorination on E P content in Examination Gloves

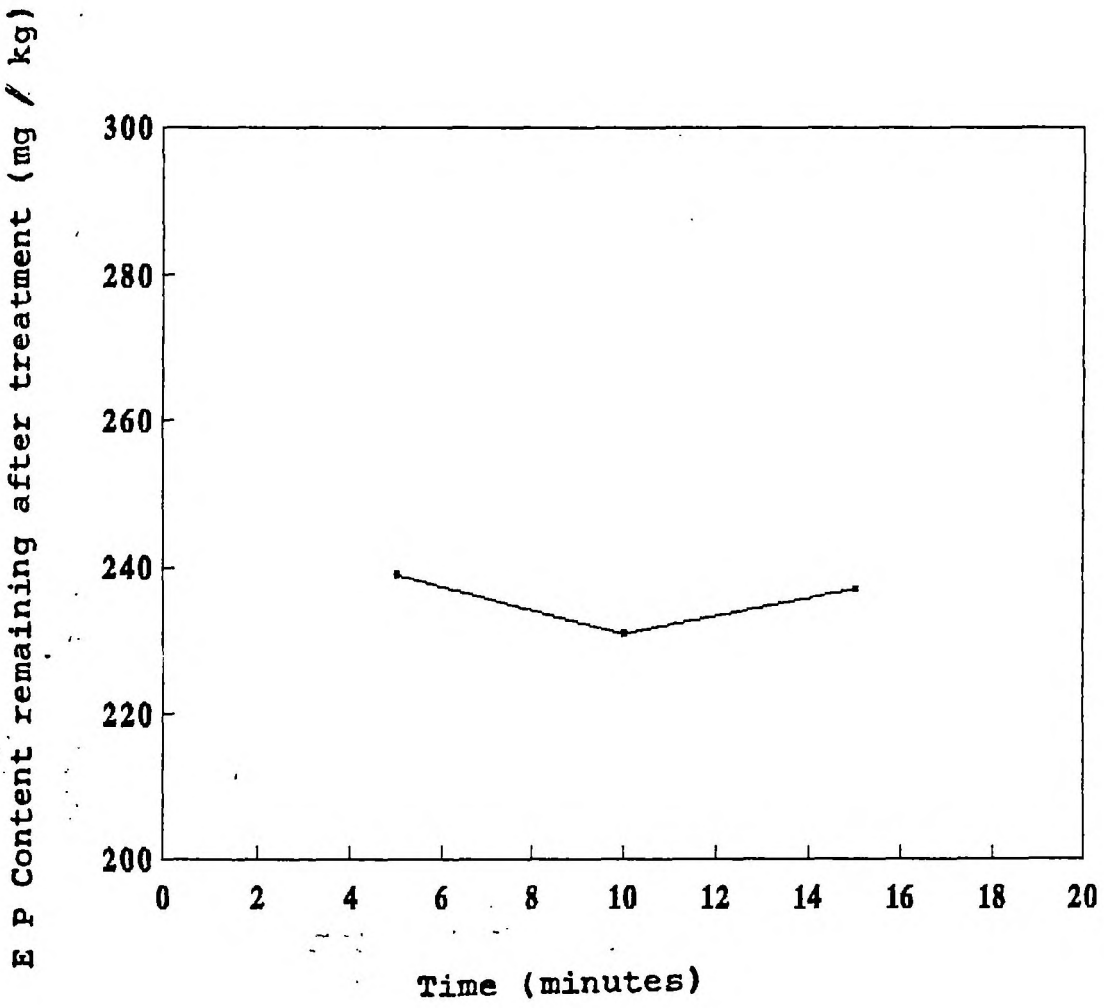


Fig. 13

Effect of Chlorinating time on E P content in Examina tion Gloves

fected by the concentration of the chlorine solution, or by the time of chlorination.A Ø.3 % solution for 5 -1Ø minutes is observed to be sufficient.

The effect of chlorination on physical properties of glove samples was also studied and the results are given in Table 6 and Table 7.

TABLE 6

Effects of chlorination on physical properties of latex gloves.

Sample details	Physical properties					
	Before Ageing			After ageing (100°c/24hrs)		
	T.S (MPa)	Modulus (MPa)	E.B (%)	T.S (MPa)	Modulus (MPa)	E.B (%)
Control	21.5	1.87	129Ø	18.6	2.Ø1	1143
Treated with Ø.1% chlorine soln, 3 mts.	21.14	1.84	1275	17.6	2.Ø1	1142
Treated with Ø.2% chlorine soln,3 mts.	2Ø.Ø3	2.Ø1	1Ø62	16.19	2.Ø2	1Ø22
Treated with Ø.3% chlorine soln, 3mts.	2Ø.Ø1	1.98	998	15.4	2.Ø4	681

TABLE 7.

Effect of the time of chlorination on physical properties of latex gloves.

Sample details	Physical properties					
	Before ageing			After ageing (100°C/24 hrs)		
	T.S (MPa)	Modulus (MPa)	E.B (%)	T.S (MPa)	Modulus (MPa)	E.B (%)
Treated with Ø.3% chlorine soln. 3mts.	2Ø.Ø1	1.98	998	15.4	2.Ø4	681
Treated with Ø.3% chlorine soln. 5mts.	19.86	1.89	875	13.8	1.9	62Ø
Treated with Ø.3% chlorine soln. 1Ø mts.	19.8	1.84	84Ø	11.4	1.86	58Ø
Control	21.5	1.87	129Ø	18.6	2.Ø1	1143

III.2.3.ii) Effect of siliconisation

The effect of siliconisation using a 1 % and a 1Ø % solutions of silicone emulsion are given in Table 8. It is observed that siliconisation by this method is not effective in removing EP content.

Table 8

Effect of siliconisation on EP content of latex gloves

Time of leaching	EP content remaining after treatment (mg/Kg)
Control	801
5 minutes	351
30 minutes	358
60 minutes	349

III.3. EFFECT OF TREATMENTS ON LATEX FILMS

III.3.1.Effect of leaching

The effect of wet gel leaching at different temperature for various periods of time for pre vulcanised and post vulcanised films are given in Fig 14 and Fig 15 respectively. Fig 16 represents the effect of dry film washing at different temperature and for various periods of time for post vulcanised film.

It is seen that the EP content can be greatly reduced by treatment with water even at room temperature. The extraction of soluble protein is rapid initially, but the rate of extraction decreases quickly and levels off after 5-10 minutes. Slow leach-

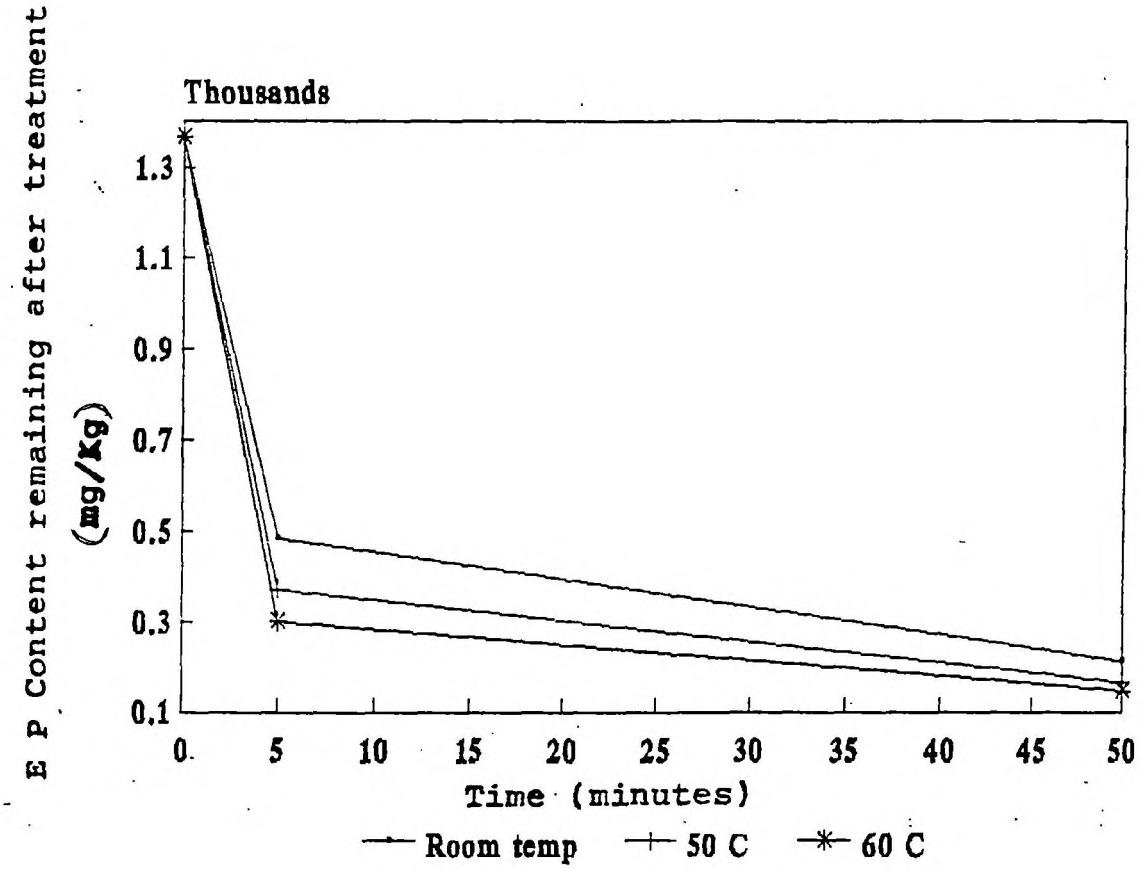


Fig. 14

Effect of Wet Gel leaching on E P content of Pre-Vulanised Latex film

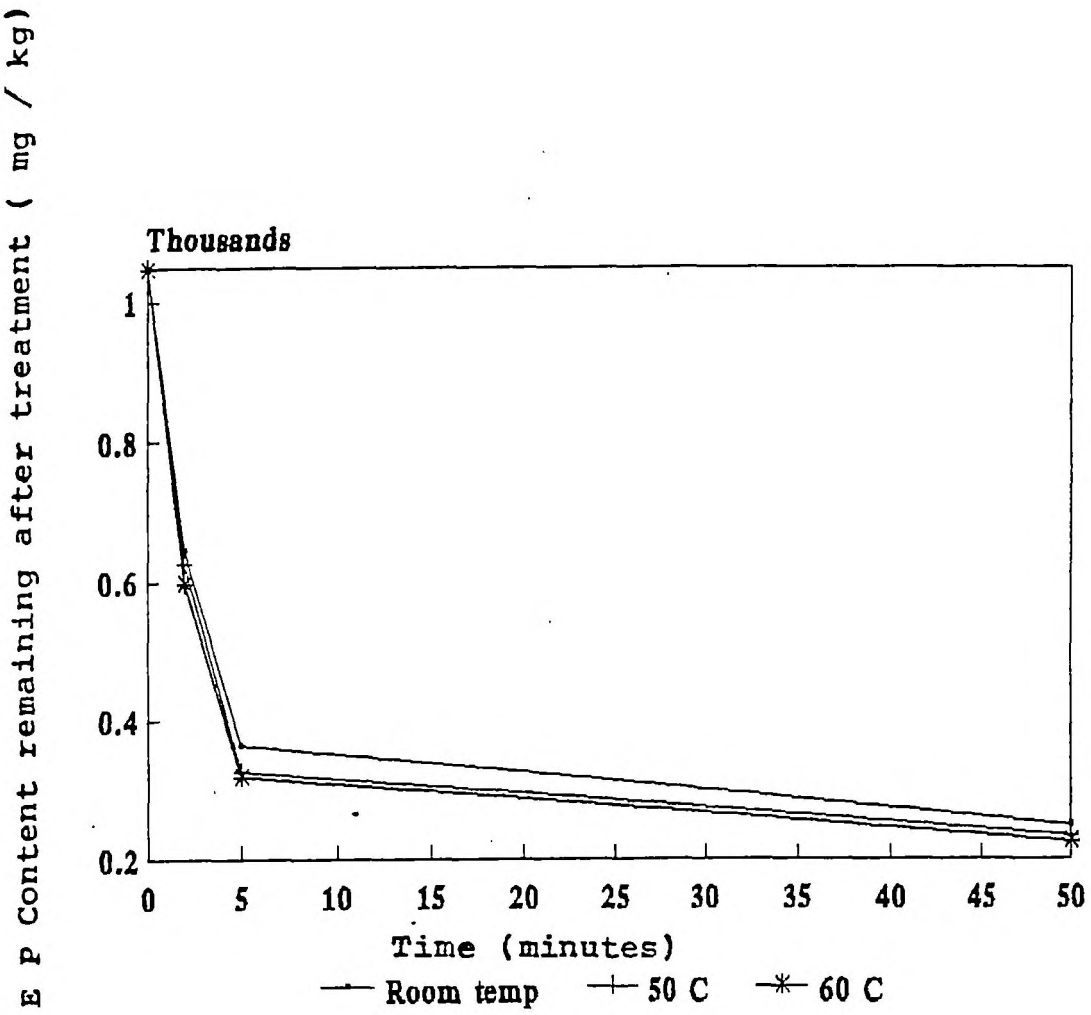


Fig. 15

Effect of Wet Gel Leaching on E.P content of Post vulcanised Latex film

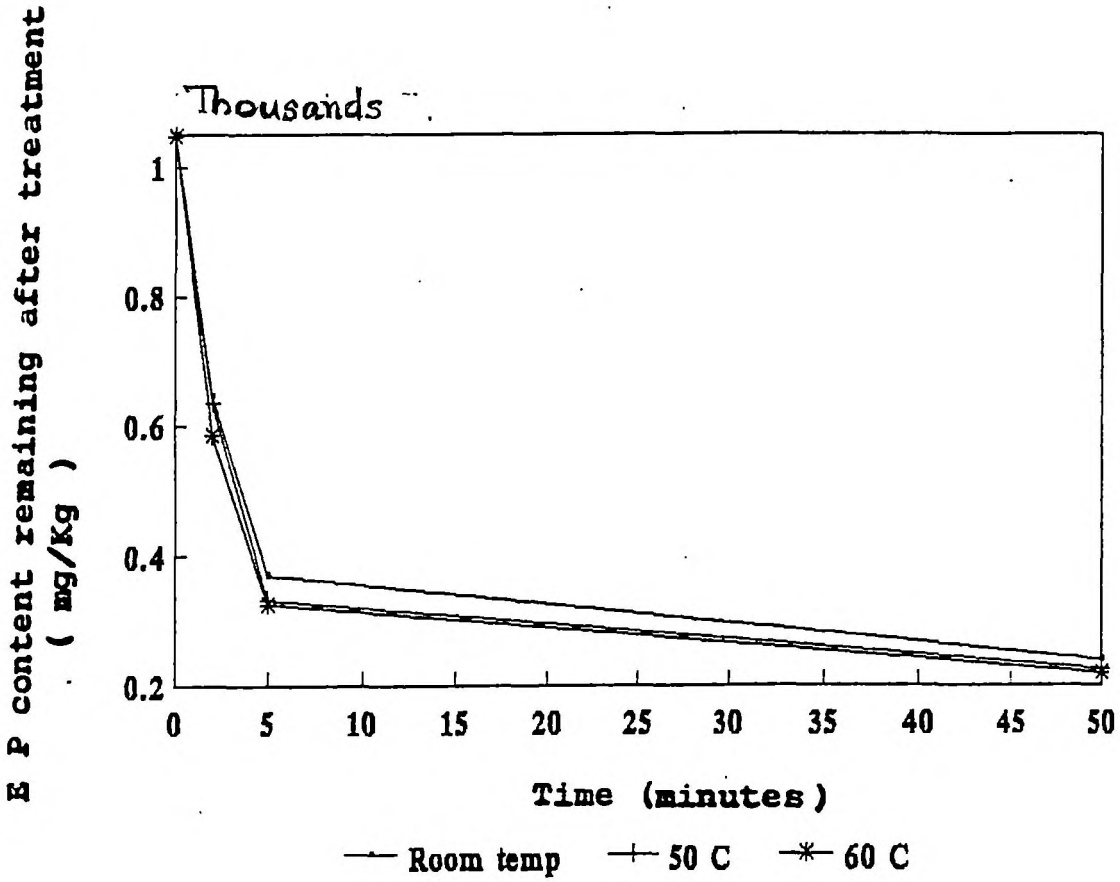


Fig. 16

Effect of dry film washing on E P content of Post vulcanised Latex film

ing of EP can continue for several hours but further reduction in EP content is small.

The reduction in EP content can be improved by leaching at elevated temperature as in the case of glove samples.

The effect of wet gel leaching and dry film washing were found to be comparable in reducing the EP content of post vulcanised latex films.

Fig 17&18 shows the effect of wet gel leaching and dry gel leaching at 60°C on post vulcanised films from doubly centrifuged latex compound.It was found that both wet gel leaching and dry gel leaching are very effective in removing the EP and the remaining EP content after treatment on doubly centrifuged latex films are very low.

The effect of leaching using water - Methanol mixture instead of water was found to be comparable with that of leaching with water.

III.2.2.Effect of chlorination

Chlorination was found to be effective in reducing the EP content for both centrifuged and doubly centrifuged latex films. Table 9. It was seen that EP content is reduced by 50 - 60 % by this process.

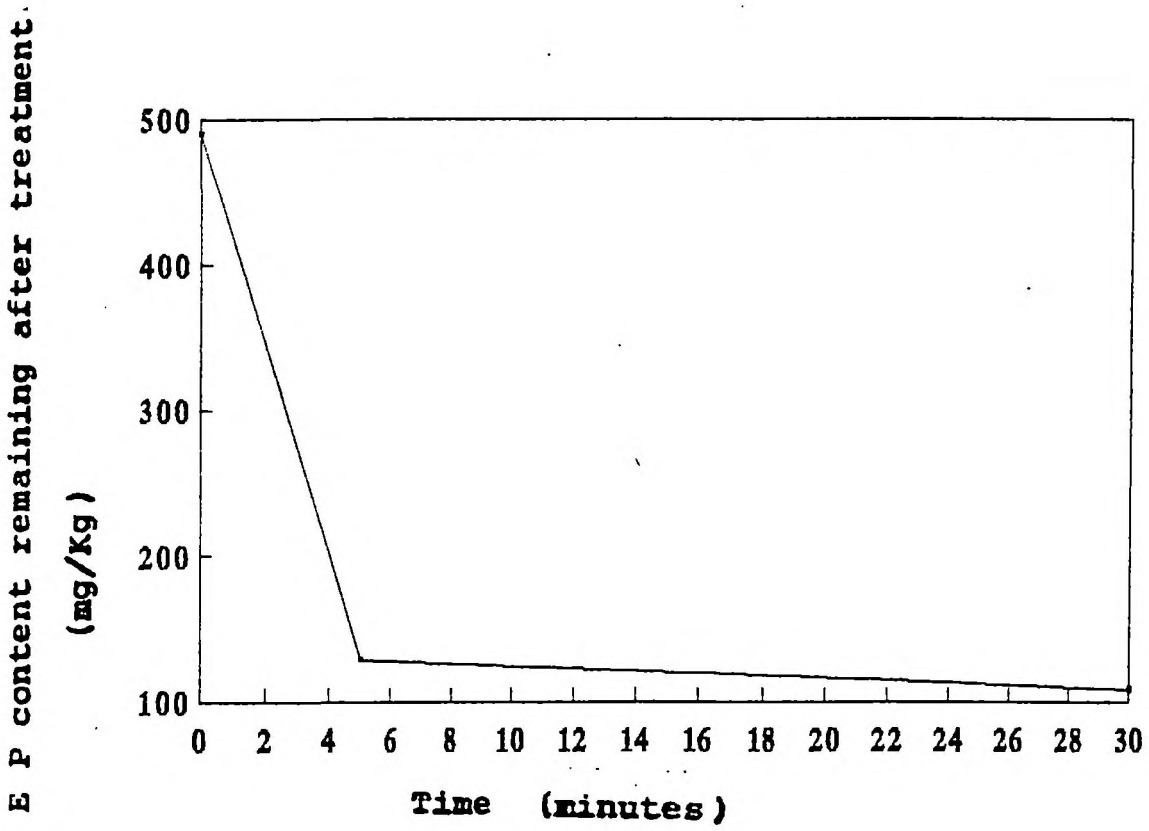


Fig . 17

Effect of wet gel leaching on E P content of D C L film (Leaching temp- 60°C)

TABLE 9

Effect of chlorination on EP content of latex films

Sample details	Concentration of chlorine solution	Extractable protein content (mg/Kg)
Film I Centrifuged	Control	310
latex	Ø.1 %	257
	Ø.3 %	160
Film II		
Doubly centrifuged	Control	104
latex	Ø.1%	52
	Ø.3%	45

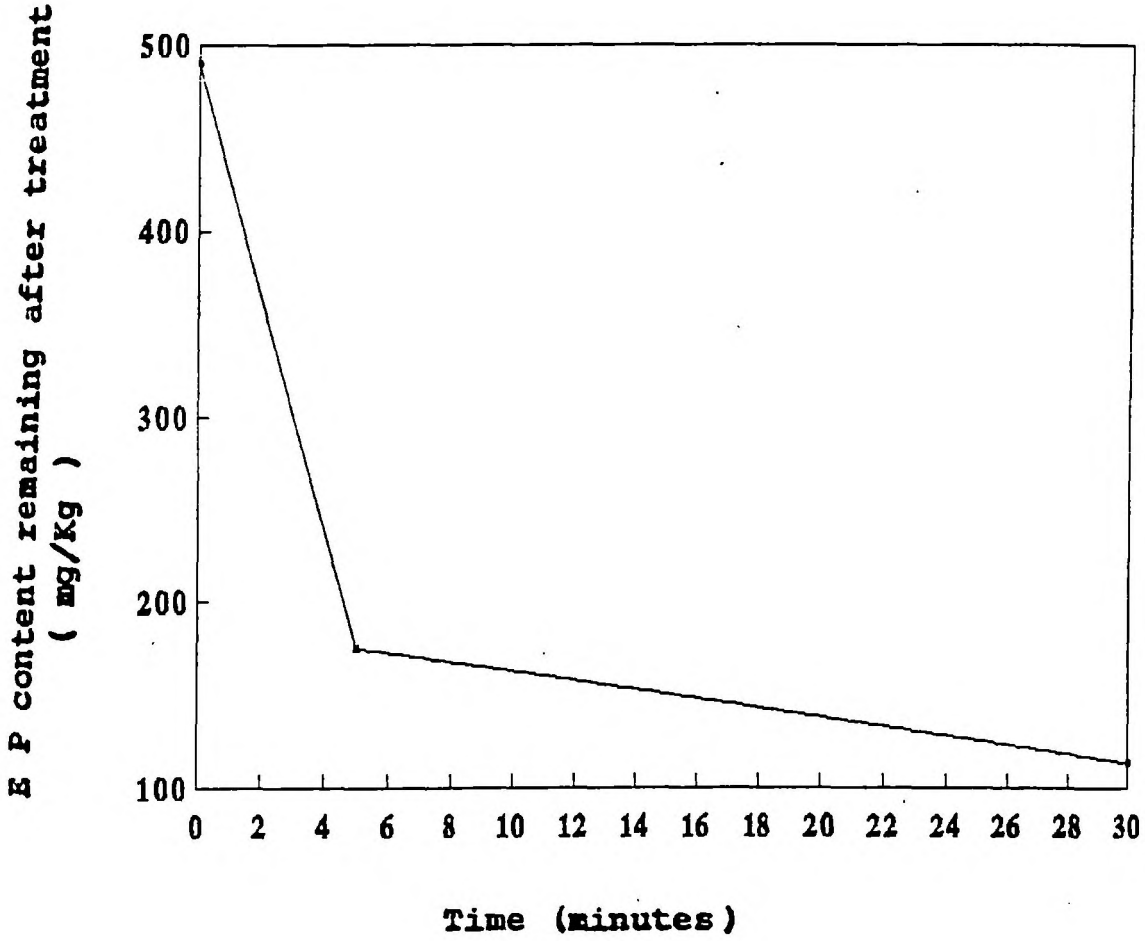


Fig. 18

Effect of dry gel leaching on E P content of D C L film (Leaching temp.- 60°C)

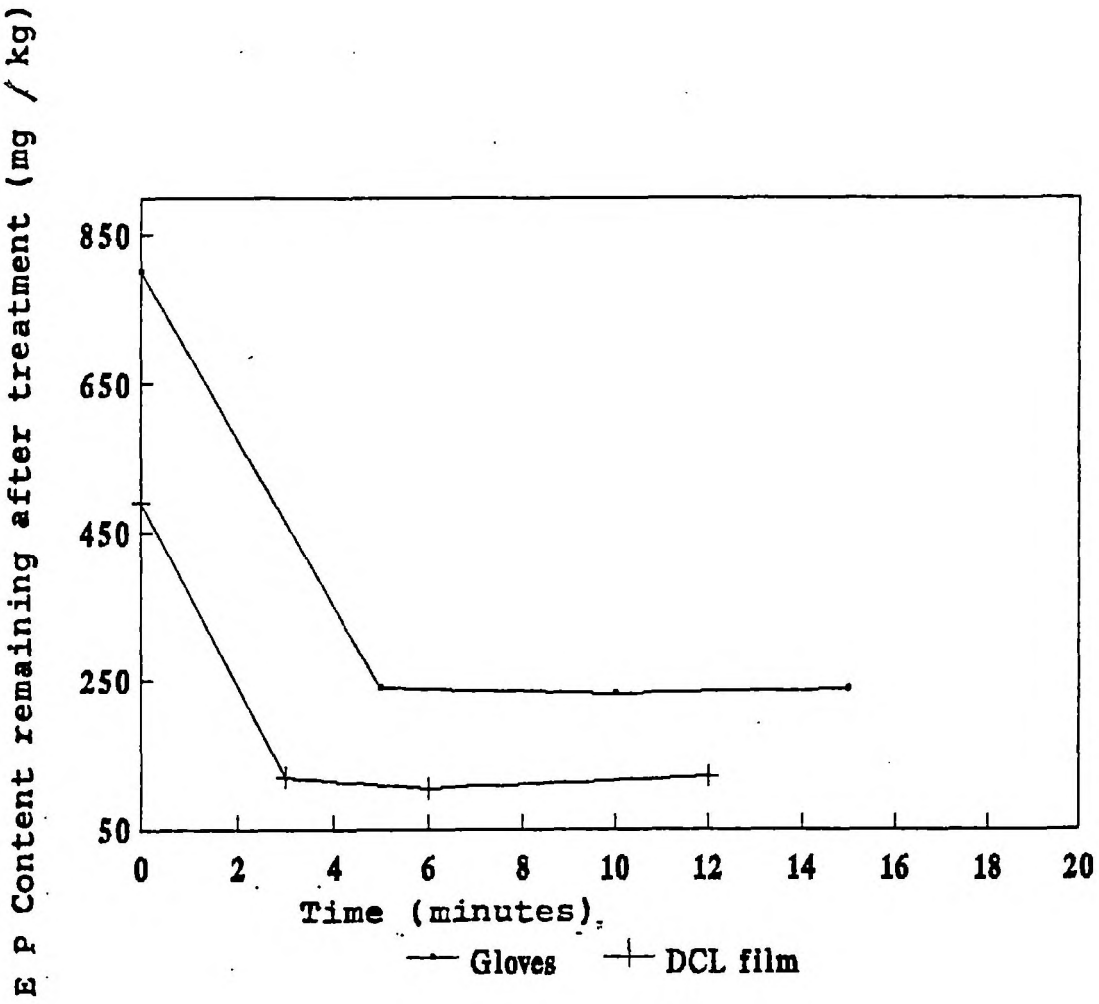


Fig. 19

Effect of Chlorination on E P content on Examination Gloves and D C L films*

* D C L films prepared by coagulant dipping, 5 minutes leaching was given and dried at 70°C

CHAPTER 4

SUMMARY AND CONCLUSIONS

In the present work, different off- the line treatment methods to remove the extractable protein content from latex products like gloves and latex films from centrifuged, pre vulcanised and doubly centrifuged lattices were studied.The methods studied were leaching studies and surface treatments.The products were also tested for their physical properties.

From the results obtained, the following conclusions could be drawn :-

(1) Off the line treatments like leaching around 60°C using 0.5 % ammonia solution or 0.5 % SLS solution and / chlorination can effectively reduce the EP content from latex products.

(2) The chlorination should be done under controlled conditions , otherwise physical properties will be affected.

(3) Use of double centrifuged / low protein latex and a suitable combination of on -the line and off -the line treatments can reduce EP content in latex products to very low levels.

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