Bulletin of the Rubber Research Institute of Sri Lanka, (2001) **43**, 57-61

LATEX PROTEINS - THE MAIN CAUSATIVE OF LATEX ALLERGY

Nadeeja Wickramarachchi

Natural Rubber (NR) Latex is an economically viable raw material obtained from the tree *Hevea brasiliensis*. It is important in many industrial applications such as in the manufacture of consumer products like toys, sports goods, latex foam, adhesives. Latex is also used for the manufacture of rubber items used in medical field, including catheters, condoms, examination and surgical gloves, bandages *etc. Hevea* latex constitutes rubber as well as non-rubbers like proteins, sugars and lipids. It is known that most of the non-rubbers in latex will be removed during processing and manufacturing. Residual water soluble proteins left in the finish product is considered to be the major source of latex allergy. Thus it is important to identify the types of proteins associated with the allergic conditions, symptoms observed under certain allergic situations and the ways to minimize such allergy causing proteins in the finished products.

Proteins in NR latex and method of analysis

Total protein content in NR latex accounts for 1-1.5 % by weight of rubber and they are distributed as water soluble and water insoluble fractions. For sometime attempts have been made by many workers to identify latex serum proteins. It is established that there are three types of protein fractions in NR latex. Electrophoresis analysis revealed that there are seven distinct proteins of widely varying isoelectric points. Paper electrophoresis technique had been used to identify the bottom fraction of proteins of *Hevea* latex. Recent studies on proteins have been conducted using Isoelectric Focusing and Sodium Dodecyl Sulphate Polyacrylamide Gel Electrophpresis (SDS-PAGE) based on the differences in Isoelectric Point (pI) and molecular weights of the proteins respectively.

However there isn't any established method to quantify the extractable protein levels in latex based products. Total water extractable protein associated with natural rubber latex and its products can be measured according to ASTMD 5712, which involves colorimetric determination by Modified Lowry Method. Other well-known methods used are BCA Protein Assay, Bradford method etc. Further evaluation of harmful protein levels in latex based products is done by two sophisticated test methods, RAST and LEAP to obtain more reliable informations compared to other test methods.

Proteins are linear high molecular weight polyamides derived from D-α-amino acids, which may also constitute with various prosthetic groups in the molecule. Presence of free acidic and basic units in each protein molecule act as the

contributing factor for the overall electric charge possessed by each molecule at different pH values, leading to proteins of wide range of isoelectric points (pI).

Proteins in fresh latex are distributed in the three main phases as follows;

Rubber phase	27%
Aqueous phase	25%
Lutoid phase	48%

Two main proteins predominantly dispersed in the above three phases of latex are;

α-globulin- this is a protein of molecular weight 20,000 and occurs exclusively as an adsorbed layer, surrounding the external surface of the rubber particle. This has been identified as the water insoluble protein in the latex serum and it is extractable only by using organic solvents.

Hevein - the second principal protein derived from latex bottom fraction, is a water soluble low molecular weight protein having isoelectric point of 4.5 and identified as the principle latex allergen.

Although the total amount of proteins present in the NR latex is relatively constant, the amount of extractable proteins in the finished products are highly variable due to removal of some of the proteins in various stages during processing and manufacture. Conversion of fresh NR latex to latex products cause inevitable changes in latex serum proteins quantitatively as well as qualitatively. In the process of ammoniation, protein content in serum phase and the rubber phase will drop down to 16-26 mg/g of rubber from 30-50mg/g of rubber. The reason behind is the loss of protein in skim latex and sludge and denaturation or degradation on ammoniation. Heating conditions employed in the prevulcanization (70^0C) and post vulcanization (110^0C) techniques converts the residual proteins to soluble forms. This is widely applicable in latex dipped product manufacture where the anionic proteins of pI 3.5-6.0 (ISO electric point) and molecular weight of 6-14KD form the major extractable proteins from the examination gloves.

Allergy caused by latex proteins

Hevea latex allergens include both soluble and particle bound proteins. Allergies concern in latex applications are two types.

Type I: This type of allergy is caused by water soluble proteins remaining in the finished latex goods.

Type IV: Allergy is initiated by residues from vulcanization ingredients such as accelerators like thiurams, dithiocarbamates and thiozoles.

Type I allergies seems to be more prominent due to its immediate hypersensitivity reactions on human skin. As there is no other successful competitor has been found yet to NR latex gloves, health care workers are keen to use them as effective barriers against bacteria and viruses. Evaluation done with regard to this respect reported that10-15% health care workers (Doctors, Dentists, Nurses) and up to 67%of Spina bifida patients have latex allergy.

Symptoms

Type I reactions occur within a very short time after exposure to the allergen. The body responds to the allergen by producing histamine and some other substances. Histamine encourages the formation of a weal or raised blister. Type I allergic reactions vary from mild itching to anaphylactic shock. These responses are a direct result of the production of histamine. Further theses types of reactions are classified as Ig E mediated hypersensitivity reactions. In addition to rashes developed on immediate vicinity on contact area on the hand when using latex gloves hay fever type reactions such as itchy swollen eyes, running nose and sneezing could be observed. Some patients may develop asthma, symptoms such as chest tightness, coughing and shortness of breath.

The most life threatening allergic reaction is found to be anaphylactic shock caused by direct tissue contact with latex products and characterized by breathing difficulties and low blood pressure. This type of allergic conditions encountered in medical field as auxillary hygienic articles such as surgical gloves, catheters get direct contact with body tissues. Moreover utricaria related to latex is possible after blowing up of toy balloons. Another possible way of causing discomfort in sensitized individuals will be created by wearing of powdered latex gloves. This is because proteins in latex become absorbed on the powder used to remove tack of the latex articles, through sweat.

Identification of latex allergy

This is primarily done by *skin prick* test which is the more sensitive and safer way of identification and more advanced *immunological* methods have been developed to identify the latex allergy reactions.

So far there is no other remedy to this problem. We should not forget the value of the great words ***"Prevention is better than cure"***. Thus the latex technologists have made concerted efforts to bring down the extractable protein levels in latex based products.

Methods of reducing extractable protein content

- **Leaching during production**

Leaching is an effective method of removing water soluble materials from latex based products such as gloves, balloons *etc.* This is considered to be the simplest and most practical method to reduce EP levels in gloves and other related items.

Leaching can be carried out in two ways, namely wet leaching and dry leaching. Though the leaching is efficient there will be trace amounts of proteins remaining on the surface of the final product which could be removed by a final wash on the dried article. Efficient leaching process is accompanied by continuous agitation of the medium.

- **Chlorination process**

This is another valuable method used to lower the extractable proteins in latex products. Basically the chlorination process is a surface reaction which results in certain changes to the surface. Chlorination process render the accumulation of insoluble surface proteins which would not be leached out on use. Chlorination reduces the tackiness of the glove surface thus eliminating the need of powdered gloves.

- **Radiation vulcanization**

Irradiation with gamma radiation disintergrate high molecular weight proteins (30,35,46 KD) bound to the rubber particle to soluble low molecular weight ones. Migration of these low molecular weight proteins to the serum phase facilitate easy removal during leaching. The best way of irradiating the latex is identified, in which initial irradiation stage is followed by centrifugation where the soluble proteins will get washed away and resulting in protein free latex.

Safety levels of extractable protein in examination gloves

Safety levels of glove proteins have been identified using Modified Lowry Method. Table drawn below specifies the appropriate levels of extractable protein and powder content in gloves.

Glove type	Protein content (µg/g)	Powder content (mg/glove)
Powdered	300	200
Powder free	50	20

Under the proposed labeling requirements labeling on all natural latex gloves would be required to include the level of water extractable proteins measured by the currently recognized ASTMD 5712 Modified Lowry Method.

The lowest acceptable level of water extractable proteins, that may be stated in the labeling will be limited by the sensitivity of the current ASTMD 5712 test method to 50 µg protein per g of natural rubber product to 300 µg per glove for a 6g glove. FDA believes that without a more sensitive standard method, lower claims would be misleading.

FDA recommends that manufacturers of powdered surgeon's and patients examination gloves limit the amount of powder to not more than 120 mg of powder

per glove, regardless of glove size and the residual powder of powder free Surgeon's and patients examination gloves to no more than 2mg per glove, regardless of glove size.

REFFERENCES

Kelly, J K *et al* (1994). The Diagnosis of Natural Rubber Latex Allergy. *The Journal of Allergy and Clinical Immunology* **93,** 813-816.

Nor, A and Aziz, A C (1993). Chlorination of Gloves. *Conference proceedings IRTC,* Kuala Lumpur, Malaysia. 59-63.

Rubber Developments. **46.** No.3/4

Susam, G *et al* (1995). Golden rules to prevent latex allergy. *Rubber Asia.* March-April 72-73.