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### Investigation of a method to reduce the levels of extractable latex proteins in dipped rubber products

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Natural Rubber Latex (NRL) allergy is an allergic condition caused by the extractable latex proteins in dipped rubber products. This allergic condition is a major concern for consumers, mainly for those who are sensitive to the allergenic extractable proteins in products such as NRL gloves. The objective of this research is to investigate the extractable protein levels in a range of dipped rubber goods and to develop an economical method to reduce the extractable protein quantities in dipped natural latex products. In our investigation of products, it was found that the NRL glove samples available in the Sri Lankan market belonging to different brands and batches contain about 500-4000  $\mu\text{g g}^{-1}$  of extractable proteins. For quantification of extractable latex proteins, modified Lowry assay (ASTM D5712-10) was used, as it was confirmed to be the most suitable through a literature survey and experimental data. Gloves were selected for further analysis on how this reduction of extractable latex proteins could be performed. In order to reduce extractable protein levels, two different natural proteases were extracted and purified; papain from papaya and bromelain from pineapple. Partial purification of bromelain and papain were carried out with 40% and 45% saturated  $(\text{NH}_4)_2\text{SO}_4$  solutions (optimum  $(\text{NH}_4)_2\text{SO}_4$  percentage according to literature) respectively from crude plant extracts. Activity of the isolated enzymes was confirmed prior to each trial through an egg white digestion test (2 hours), where the weight reduction of boiled egg white was used as an indicator to prove protease activity. With papain a weight reduction of 25 ( $\pm 5$ )% was recorded for this test and with bromelain it was 13.5 ( $\pm 2$ )%. Different glove samples of the same weight were treated with each enzyme, within an incubation time period of 2 hours at 60 °C temperature. Control experiments were carried out using distilled water in the place of enzyme. Triplicate of experiments carried out with bromelain, showed a reduction of 51 ( $\pm 11$ )% of the extractable proteins whereas with papain it was 58 ( $\pm 8$ )%. These results clearly indicate that the selected natural proteases – papain and bromelain contribute significantly towards the reduction of total aqueous extractable proteins in NRL products. This concept can be further reformed for application on an industrial scale to treat either the product or the natural rubber latex itself to decrease the health complications that occur due to allergic conditions in dipped rubber product consumers.

Keywords: Bradford assay, bromelain, modified Lowry assay, NRL allergy, papain

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