ORIGINAL ARTICLE

Estimating Allergenicity of Latex Gloves Using Hev b 1 and Hevamine

MF Lee,[1,2] NM Wang,[3] JL Han,[3] SJ Lin,[4] JJ Tsai,[1] YH Chen[5,6]

[1]Department of Medical Research, Taichung Veterans General Hospital, Taichung, Taiwan
[2]General Education Center, Overseas Chinese University, Taichung, Taiwan
[3]Institute of Biotechnology, National Changhua University of Education, Changhua, Taiwan
[4]School of Medical Laboratory and Biotechnology, Chung Shan Medical University, Taichung, Taiwan
[5]Division of Allergy, Immunology and Rheumatology, Taichung Veterans General Hospital, Taichung, Taiwan
[6]School of Medicine, National Yang-Ming University, Taipei, Taiwan

Abstract

Background: Latex allergy continues to be an increasingly serious occupational health problem in Taiwan, where it affects approximately 6.8% to 12% of health care workers. Contrasting with reports from western countries, Hev b 1 and hevamine, and not Hev b 3, 5 or 6.02, are the major latex allergens among health care workers in Taiwan. This study aimed at evaluating the allergenicity of 30 brands of commercially available medical latex gloves in Taiwan in 2007.
Methods: Residual Hev b 1 and hevamine from the gloves were measured by inhibition enzyme-linked immunosorbent assay using polyclonal antibodies against purified recombinant Hev b 1 and hevamine. The results were compared to those achieved with quantification of residual total extractable proteins and skin prick testing.
Results: The residual extractable protein levels in 30 medical gloves all conformed to United States Food and Drug Administration regulations. All the gloves except one yielded strong skin prick reactions in latex-allergic individuals. The only brand of gloves that consistently produced no skin prick reactions in latex-allergic individuals contained the lowest residual levels of Hev b 1 (0.60 μg/g) and hevamine (0.07 μg/g).
Conclusions: Our results suggest that the measurement of residual extractable total proteins is not sufficient to assess the allergenicity of latex gloves and that Hev b 1 and hevamine may be used as indicator allergens in areas where they are major latex allergens, such as Taiwan.

Key words: Latex allergy. Latex gloves. Allergen. Hev b 1. Hevamine.

Resumen

Antecedentes: La alergia al látex es un problema de salud laboral cada vez mayor en Taiwán, donde afecta aproximadamente al 6,8% 12% de los trabajadores sanitarios. A diferencia de los informes procedentes de países occidentales, los principales alérgenos del látex entre los trabajadores sanitarios de Taiwán son Hev b 1 y hevamina (y no Hev b 3, 5 ó 6.02). El objetivo de este estudio fue evaluar la alergenicidad de 30 marcas de guantes de látex de uso médico comercializados en Taiwán en 2007.
Métodos: Se midieron los residuos de Hev b 1 y hevamina de los guantes mediante enzimoinmunoanálisis de adsorción de inhibición con el uso de anticuerpos policlonales frente a Hev b 1 y hevamina recombinantes purificadas. Los resultados se compararon con los obtenidos con la cuantificación de proteínas extraíbles residuales totales y las pruebas de punción cutánea.
Resultados: Los niveles de proteínas extraíbles residuales de los 30 guantes de uso médico cumplieron las normativas de la Food and Drug Administration estadounidense. Todos los guantes salvo unos dieron lugar a fuertes reacciones en las pruebas de punción cutánea en pacientes alérgicos al látex. La única marca de guantes que de forma sistemática no produjo reacciones en las pruebas de punción cutánea en pacientes alérgicos al látex contenía los niveles residuales más bajos de Hev b 1 (0,60 μg/g) y hevamina (0,07 μg/g).
Conclusiones: Nuestros resultados indican que la determinación de las proteínas extraíbles residuales totales no es suficiente para evaluar la alergenicidad de los guantes de látex, y que Hev b 1 y hevamina pueden utilizarse como alérgenos indicadores en las zonas donde constituyen los principales alérgenos del látex, como en Taiwán.

Palabras clave: Alergia al látex. Guantes de látex. Alérgeno. Hev b 1. Hevamina. ELISA de inhibición.

Introduction

Natural rubber latex (NRL) is a milky fluid from the *Hevea brasiliensis* tree that functions as a protective sealant [1]. Because of its excellent elastic properties, it is widely used in the manufacture of medical devices and in a variety of everyday articles such as gloves, condoms, balloons, baby nipples, syringe plungers, and vial stoppers. According to a report by Perkin et al [2] in 2000, as many as 40 000 types of consumer products may contain NRL [2]. Immediate allergy to latex gloves was first reported in 1979 [3] and numerous cases of latex allergy have been reported since the 1980s due to the sharp increase in the use of latex gloves to reduce the risk of infection [4-6]. Latex hypersensitivity is observed in certain occupational and other high-risk groups with frequent exposure to NRL products, including health care workers, rubber industry workers [7], children with spina bifida [8,9], and atopic individuals [10]. Sensitization and development of latex allergy arise from exposure to products containing residual latex proteins. Clinical symptoms manifest as contact urticaria, rhinoconjunctivitis, asthma, and mucosal swelling. Systemic reactions consist of generalized urticaria and anaphylactic shock [6]. Reported prevalence of latex allergy ranges from 2.8% to 17% in Europe and the USA [11-13]. In health care workers in Taiwan, the rates range from 6.8% to 12% [14-16]. Prevalence in the general population is believed to be less than 1.5% [17].

NRL in its crude state contains more than 200 polypeptides, 56 of which have been identified as allergens with immunoglobulin (Ig) E–binding activities [18-21]. However, only a few studies have purified or cloned NRL allergens to date. The World Health Organization and International Union of Immunological Societies Allergen Nomenclature Committee (www.allergen.org) lists 13 NRL allergens characterized at the molecular level, designated from Hev b 1 to Hev b 13 [22]. It remains unclear which of these allergens are most resistant to the harsh rubber manufacturing processes and act as major sensitizing molecules. Information regarding the status of allergenic proteins in latex products is incomplete [23]. At present, skin prick testing with crude latex extracts is the most frequently used clinical test for the diagnosis of latex allergy [24]. Crude extracts are not an ideal source of standardized allergens due to their batch-to-batch variability and instability. Allergens produced by recombinant DNA technology, in comparison, are reported to be a safe and effective source of allergens for the diagnosis of allergy [25,26].

Although many nonlatex gloves have appeared on the market, NRL gloves have shown a lower rate of leakage compared to vinyl and nitrile gloves [27], which makes it unlikely that they will be completely replaced, despite the increase in latex allergy cases. Hunt et al [28] reported that replacing these gloves with hypoallergenic products that contain very low or undetectable levels of allergens has markedly reduced the incidence of latex allergies among health care workers [28]. Therefore a reliable method for evaluating the allergenicity of latex products is essential for the successful reduction of latex allergy. Previously, we identified that Hev b 1 and hevamine, reactive with 85% and 55% of patient sera, are major latex allergens in Taiwan [29]. In the present study, we report that both residual Hev b 1 and hevamine can serve as surrogate markers of allergenicity in latex gloves using antibodies against recombinant Hev b 1 and hevamine.

Methods

Serum Samples

Twelve latex-allergic health care workers and 5 healthy nonallergic individuals were enrolled in this study. The Institutional Review Board of Taichung Veterans General Hospital approved the study protocol.

Preparation of Latex Glove Extracts and Protein Quantitation

Proteins were extracted from 20 brands of examination gloves (E1-E20) and 10 brands of surgical gloves (S1-S10) available in Taiwan in 2007. Briefly, the gloves were cut into small pieces and mixed with 8 mL/g of phosphate-buffered saline (PBS, pH 7.4) for 16 hours at 4°C with shaking. Thereafter, the extracts were centrifuged to remove the glove powder and other particulates, and the clear supernatant was concentrated 80-fold using Amino Ultra centrifugal filter devices (Millipore, Bedford, Massachusetts, USA). The protein concentration was determined using the Bio-Rad Bradford assay (Bio-Rad, Hercules, California, USA).

Cloning and Purification of Recombinant Hev b 1 and Hevamine Proteins

Total RNA was extracted from fresh buds of *Hevea brasiliensis* with Concert Plant RNA Reagent (Invitrogen, Carlsbad, California, USA). First-strand complementary DNA (cDNA) synthesis was performed using the ThermoScript RT-PCR system (Invitrogen) according to the manufacturer's instructions. Oligodeoxynucleotide primers for Hev b 1 and hevamine cDNA amplification were designed according to previously reported sequences (GeneBank access No. GI:132270 and 234388, respectively). The cDNA coding regions of Hev b 1 and hevamine were cloned into vector pQE30 (Qiagen, Valencia, California, USA), and then transformed into *Escherichia coli* M15 [pREP4] for expression. The recombinant proteins were purified by rapid affinity column chromatography with the His-tag system under denaturing conditions (Novagen, Madison, Wisconsin, USA). The purified proteins were refolded using dialysis with the gradual removal of urea in 0.02 M PBS, pH 7.2. The reactivity of recombinant Hev b 1 and hevamine proteins was evaluated via direct binding enzyme-linked immunosorbent assay (ELISA), as previously described [29].

Antirecombinant Hev b 1 and Antihevamine Antibodies

Antibodies against recombinant Hev b 1 and hevamine were raised in rabbits. Young adult New Zealand white rabbits were injected subcutaneously at 10 to 20 sites on the dorsum with 150-μL aliquots containing 2.0 mg purified recombinant proteins with an equal volume of complete Freund's adjuvant (Sigma, St Louis, Missouri, USA). After a rest period, 2 booster injections were given using 1.0 mg antigen mixed with Freund's incomplete adjuvant (Sigma) on weeks 4 and 8.

Three weeks following the last injection, the rabbits were bled by heart puncture. Antisera were purified by Protein A-agarose (Bio-Rad) affinity chromatography.

Quantitation of Allergens in Latex Gloves by Inhibition ELISA With Anti-Hev b 1 and Anti-Hevamine Antibodies

Optimal concentrations of antigen and conjugate were determined by checkerboard titration. First, inhibition plates were prepared by blocking with 3% nonfat milk/PBS overnight at room temperature. After washing with PBST, 3 two-fold dilutions of each test extract (100 µL/well) and 5 two-fold dilutions of recombinant Hev b 1 and hevamine proteins beginning at 0.4 µg/mL were prepared in duplicate wells. Rabbit anti-Hev b 1 and antihevamine antibodies (1/16000 dilution) were added to each sample dilution (100 µL/well). The plates were incubated for 2 hours at 37°C.

Microtiter plates containing the solid-phase antigen were prepared by coating with 0.1 µg/well of recombinant Hev b 1 and hevamine in a carbonate buffer, pH 9.6. After incubation for 2 hours at 37°C, the nonreactive sites were blocked for 1 hour with 3% nonfat milk/PBS.

After incubation of the inhibition plates for 2 hours to allow antibody reaction with the test sample, the inhibited antiserum was transferred to 96-well assay plates containing the solid-phase antigen. The plates were then incubated for 2 hours at room temperature to allow the unbound antibodies to bind with the solid-phase antigen. After washing, a 1/5000 dilution of peroxidase-labeled goat antirabbit IgG was added and incubated for 1 hour at room temperature, and a colored reaction was developed by the addition of ABTS (ABTS, 55 µg/mL in 0.1 M sodium citrate buffer, pH 4.2, containing 0.03% of H_2O_2).

The results were read at 415 nm on a Sunrise Reader (TECAN, Grödig, Austria). The concentration of Hev b 1 and hevamine in the test samples were determined by comparing the reference standard range between 0.025 and 0.4 µg/mL.

Skin Prick Test

Protein samples at concentrations of 100 µg/mL in PBS containing 50% glycerol were used for skin testing. Allergens were applied to the testheads with epicutaneous sterile disposable Sharp Test applicators (Greer Laboratories, Lenoir, North Carolina, USA). Histamine (1 mg/mL) and 50% PBS-glycerol were used as positive and negative controls, respectively. All the skin test results were read 20 minutes after placement for immediate wheal-and-flare reactions. A response with a wheal 3 mm larger than that produced by the negative control was considered positive. Results of the skin prick test were expressed as wheal area in mm^2.

Results

Characteristics of Participants

Table 1 shows the demographic, clinical, and serologic characteristics of the 12 latex-allergic and 5 nonallergic individuals. The prevalence of IgE reactivity to recombinant Hev b 1 (rHev b 1) and hevamine (rHevamine) determined by

Table 1. Characteristics of Latex-Allergic Patients and Nonallergic Individuals

Participants	Age, y/Sex	Occupation	Clinical Symptoms[a]	LatexCAP, kU/L	Glove Extract ELISA, OD	rHev b 1 ELISA, OD	rHevamine ELISA, OD
P1	53/M	Surgeon	AR, AC	4.77	0.24	0.33	0.09
P2	28/F	Technician	AS, AR	0.39	0.64	0.22	0.32
P3	30/F	Technician	AS, AR, AC	37.7	1.06	1.43	0.84
P4	33/F	Technician	AS, AR, AC, U	6.14	0.36	0.51	1.34
P5	30/F	Nurse	AR	0.61	0.28	0.25	0.05
P6	41/M	Physician	AR, AC, U	4.89	0.90	0.08	0.07
P7	36/F	Dentist	AS, AR	1.84	0.24	0.27	0.08
P8	31/F	Nurse	AS	3.36	0.73	0.23	0.29
P9	44/F	Technician	AS, AR, U	40.4	0.69	0.26	0.39
P10	26/M	Technician	AS, AD, U	7.16	0.66	0.58	0.54
P11	26/M	Medical student	AR, U	4.68	0.63	0.46	0.38
P12	45/F	Nurse	AR, U	4.13	0.58	0.39	0.28
NA1	42/F	Physician	None	<0.35	0.08	0.09	0.08
NA2	28/M	Technician	None	<0.35	0.09	0.07	0.08
NA3	26/F	Technician	None	<0.35	0.10	0.11	0.10
NA4	34/M	Technician	None	<0.35	0.06	0.08	0.06
NA5	23/F	Technician	None	<0.35	0.09	0.09	0.07

Abbreviations: AR, allergic rhinitis; AC, allergic conjunctivitis; AS, asthma; AD, allergic dermatitis; ELISA; enzyme-linked immunosorbent assay; F, female; M, male; OD, optical density; rHev b 1, recombinant Hev b 1; rHevamine, recombinant hevamine; U, urticaria.
[a]Symptoms on exposure to latex product.

Table 2. Summary of Allergen and Protein Levels and SPT Results for 30 Latex Gloves

No.	Brand/lot no.	Quantity, µg/g Glove[a]			SPT, mm[2b]				
		Total protein	Hev b 1	Hevamine	P3[b]	P4[b]	P9[b]	NAI[c]	NA2[c]
EE1(pf)[1]	Anderson	4.646	1.806	0.298	342	1190	960	–	–
E2(pf)	CSD	7.894	0.804	0.216	625	1089	1190	–	–
E3(pf)	DECROWN	7.246	1.354	0.136	-	600	1120	–	–
E4(p)	DECROWN	23.18	7.762	0.668	992	1116	756	–	–
E5(p)	ENCHS	9.148	0.692	0.21	1254	918	1056	–	–
E6(pf)	FLOWER/ 3051205	7.416	0.6	0.07	–	–	–	–	–
E7(pf)	FLOWER/ HAW0040506	27.8	2.322	0.164	180	980	700	–	–
E8(pf)	FLOWER/ HAW0050101	6.816	3.69	0.438	49	960	750	–	–
E9(pf)	FORCARE	24.58	7.122	2.938	176	650	992	–	–
E10(p)	GE	7.742	3.71	0.206	-	90	306	–	–
E11(pf)	KH	7.15	2.782	0.45	182	169	784	–	–
E12(pf)	MODERN	20.792	4.446	0.394	-	48	1155	–	–
E13(pf)	PINSIN	11.91	6.718	0.186	750	729	1152	–	–
E14(p)	PROTOS/ BL09/1007U	29.386	7.386	2.506	986	806	960	–	–
E15(pf)	PROTOS/ ITH18/0904	18.362	5.964	0.224	132	1296	1089	–	–
E16(p)	PROTOS/ 60001044	29.924	2.928	0.45	400	1122	1080	–	–
E17(p)	PROTOS/ YF02/0806	53.338	6.486	0.284	500	1020	1023	–	–
E18(pf)	PROTOS/ YF02/0806	7.056	1.176	0.162	928	675	–	–	–
E19(p)	TOP/ 1200004125	4.654	2.602	0.238	1292	504	400	–	–
E20(p)	TOP/ 300005116	7.988	0.766	0.186	528	598	1020	–	–
S1(pf)	ENCHS	6.044	2.24	0.388	nd	nd	nd	nd	nd
S2(pf)	MODERN/ 40577	8.27	6.024	1.73	nd	nd	nd	nd	nd
S3(pf)	MEDI/ 16.97/2.8DH	8.364	1.872	0.176	nd	nd	nd	nd	nd
S4(pf)	MEDI/ 18.97/4.8DH	21.562	1.016	0.112	nd	nd	nd	nd	nd
S5(pf)	MEDI/ 21.97/7.2DH	8.34	1.07	0.148	nd	nd	nd	nd	nd
S6(pf)	TRIFLEX/ 07TS	30.956	1.406	0.256	nd	nd	nd	nd	nd
S7(pf)	TRIFLEX/ 06TS	10.354	3.044	0.222	nd	nd	nd	nd	nd
S8(pf)	TRIFLEX/ 08TS	6.388	1.616	0.204	nd	nd	nd	nd	nd
S9(pf)	TRIFLEX/ 0621TS	5.512	0.896	0.164	nd	nd	nd	nd	nd
S10(pf)	MODERN/ 709081	8.016	0.814	0.126	nd	nd	nd	nd	nd

Abbreviation: E1-20, examination gloves 1-20; nd, not done; p, powdered; pf, powder-free; SPT, skin prick test; S1-10, surgical gloves 1-10.
[a]Values represent mean of data from 3 independent experiments.
[b]SPT results for latex-allergic individuals. Patient numbers (P) are the same as those used in Table 1.
[c]SPT results for nonallergic individuals. Nonallergic individual numbers (NA) are the same as those used in Table 1.

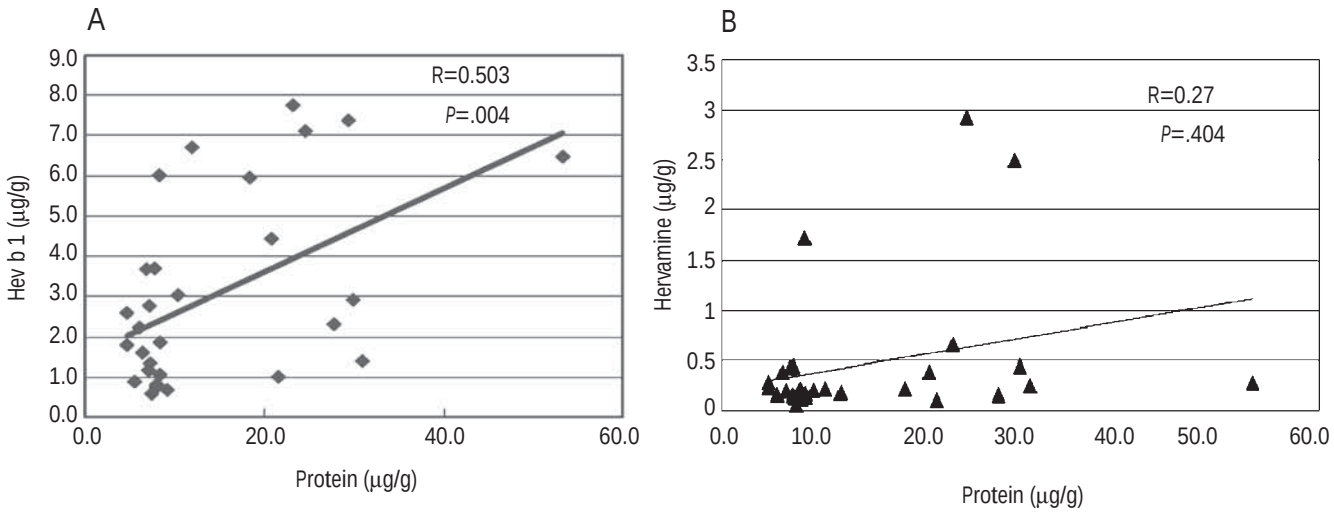

Figure 1. Relationship between protein content (μg/g glove) and allergens (μg/g glove) in 30 glove eluates. A, Hev b 1. B, hevamine.

ELISA was 92% (11/12) and 67% (8/12), respectively, which was consistent with a previous report [29].

Detection of Residual Total Protein, Hev b 1 and Hevamine From Latex Gloves

Total extractable protein and allergen levels were investigated in 30 brands of latex gloves including 20 examination gloves and 10 surgical gloves. The amount of protein extracted from each latex glove was evaluated by the Bradford method, using bovine serum albumin as a protein standard. The levels of total extractable protein varied considerably between surgical and examination gloves, ranging from 4.65 μg to 53.34 μg per gram of glove. The quantity of Hev b 1 and hevamine was measured in extracts from the 30 glove samples by inhibition ELISA using rHev b 1 and rHevamine as standards. Firstly, we confirmed that in-house made rabbit anti-rHev b 1 and anti-rHevamine IgG were able to detect each recombinant protein specifically (data not shown). The levels of Hev b 1 and hevamine were 0.60 to 7.76 μg and 0.07 to 2.94 μg per gram of glove, respectively. The results are summarized in Table 2.

Identifying the Marker Allergen and Estimating the Allergenic Potential of the Gloves

Regression analysis was performed to examine the correlation of residual Hev b 1 and hevamine with total extractable protein. The correlation between total extractable protein and allergen levels (μg/g glove) from 30 batches of gloves is shown in Figure 1. The level of Hev b 1 was only marginally significantly correlated with the total extractable protein levels by analysis of variance (ANOVA) (R=0.503, $P<.01$, Figure 1A). Hevamine levels did not correlate with total extractable protein levels (R=0.27, $P=.404$), as shown in Figure 1B.

In order to investigate whether gloves with high- and low-allergen contents exhibit a different capacity to elicit biologically relevant reactions, skin prick tests were performed in 3 latex-allergic patients and 2 nonallergic individuals. The forearms of 1 latex-allergic patient and 1 nonallergic individual after SPT are shown in Figure 2. Nineteen of the 20 latex gloves tested elicited wheal and flare reactions in the 3 latex-allergic patients. The glove that did not elicit any skin reactions (E6) contained the lowest amounts of Hev b 1 and hevamine, as shown in Table 2.

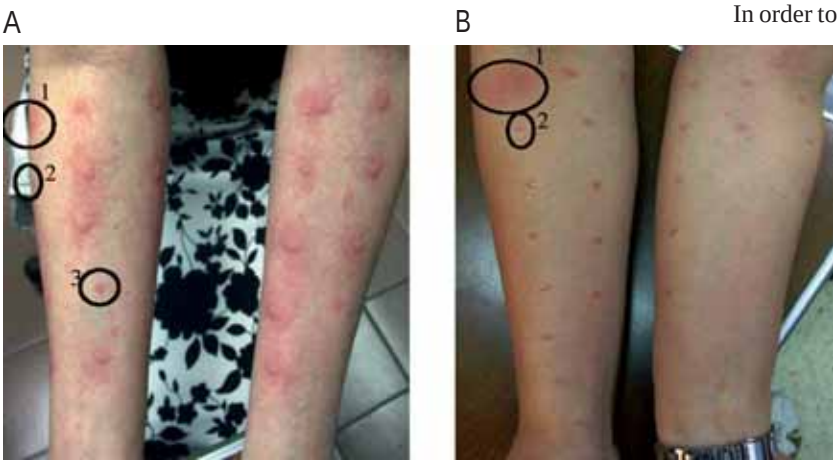

Figure 2. Two representative pictures taken from the forearms of a latex-allergic subject (A) and a nonallergic individual (B) after a skin prick test with protein extracts from 20 brands of examination gloves. Encircled (1) histamine 1 mg/mL as positive control, (2) phosphate-buffered saline as negative control, (3) crude extract of glove E6.

Discussion

In recent years, the prevalence of latex allergies has increased steadily, particularly among health care workers in Taiwan [16,30].

Several studies have documented considerable differences between the allergen content of latex gloves made by different manufacturers, and even between gloves of different batches from a single manufacturer [31,32]. In 1999, the United States Food and Drug Administration (FDA) issued a proposed regulation regarding medical gloves that recommended a maximum allowable extractable protein level of 1.2 mg per glove [33]. With latex glove manufacturers actively taking steps to reduce extractable proteins in their products, current gloves tend to have lower total protein levels [23]. Our data revealed that the residual extractable total protein content in the 30 medical gloves we examined were all conformable to the 1999 FDA limit of 1.2 mg per glove. Nevertheless, 19 of the 20 latex gloves elicited strong skin prick test reactivity in the 3 patients with latex allergy tested. Although protein and allergen content used to be assumed to be in parallel, it has been seen that many extracted proteins do not exhibit IgE-binding capacity [31,34]. Our data further indicate that the determination of total extractable protein content is not sufficient to assess the true allergenicity of latex gloves.

We have previously reported that, in contrast to the situation in western countries, where Hev b 5 and Hev b 6 are major latex allergens, more than half of latex-allergic health care workers in Taiwan are sensitive to Hev b 1 and hevamine [29]. These are the major latex allergens among health care workers in Taiwan. In the present study, Hev b 1 and hevamine concentrations were found to be in the range of 0.60 to 7.76 μg/g and 0.07 to 2.94 μ/g, corresponding to 5% to 73% and 0.5% to 8.5% of the total extractable protein content in NRL glove extracts, respectively. In theory, an ideal method for measuring the allergenicity of NRL products would be skin prick testing in voluntary latex-allergic subjects, the gold standard for diagnosing latex allergy. For obvious ethical reasons, however, such tests cannot be routinely used for monitoring allergen content in latex gloves. Only 3 latex-allergic subjects allowed us to perform skin prick tests using 20 laboratory-prepared glove extracts on their forearms. To our surprise, nineteen of the gloves elicited wheal and flare reactions in all 3 patients. The exception was glove number E6, which contained the lowest level of Hev b 1 (0.6 μg/g glove) and hevamine (0.07 μg/g glove) and yielded no SPT reactivity in any of the 3 patients tested. The data suggest that Hev b 1 and hevamine are better indicators of in vivo allergenicity in Taiwan than total extractable protein content. However, in areas where Hev b 3, 5 and 6.02 are major latex allergens, in which they should also be measured the sum quantity of major latex allergens, as reported by Reinikka-Railo et al [35]. They suggested that medical gloves with sum values of below 0.15 μg/g for Hev b 1, 3, 5 and 6.02 can be considered to have low allergenic potential in Finland. Future studies enrolling a higher number of patients to establish acceptable cutoff levels for Hev b 1 and hevamine in Taiwan are required.

Minimizing allergen concentration in latex goods to prevent sensitization to NRL is an important issue for the regulatory health authorities. The FDA has recognized the measurement of total protein as a simple option for glove manufacturers to monitor their products. However, measuring total protein rather than specific allergens cannot be deemed a satisfactory regulatory activity to control the allergen content of NRL products. Early methods based on human IgE-containing reagents are not readily available and suffer from a lack of standardization. In this study, we developed specific polyclonal antibodies to measure major allergen levels in medical gloves. We were able to successfully measure Hev b 1 and hevamine by inhibition ELISA using polyclonal antibodies and purified recombinant allergens as standards.

In conclusion, our results suggest that determination of residual extractable total protein content is not sufficient to estimate the allergenicity of latex gloves and that Hev b 1 and hevamine may be used as indicator allergens in areas where they are major latex allergens, such as Taiwan.

Acknowledgments

This study was supported by grant TCVGH-977306B from Taichung Veterans General Hospital, Taiwan. We thank Dr Chi-Huei Wu for reviewing the manuscript and offering valuable suggestions. We also thank Miss Hsin-Ju Wu for assistance with skin prick testing and blood collection.

References

1. Ownby DR. A history of latex allergy. J Allergy Clin Immunol. 2002: 110:S27-32.
2. Perkin JE. The latex and food allergy connection. J Am Diet Assoc. 2000: 100:1381-4.
3. Nutter AF. Contact urticaria to rubber. Br J Dermatol. 1979:101:597-8.
4. Carrillo T, Cuevas M, Munoz T, Hinojosa M, Moneo I. Contact urticaria and rhinitis from latex surgical gloves. Contact Dermatitis. 1986:15:69-72.
5. March PJ. An allergic reaction to latex rubber gloves. J Am Dent Assoc. 1988:117:590-1.
6. Ownby DR, Tomlanovich M, Sammons N, McCullough J. Anaphylaxis associated with latex allergy during barium enema examinations. Am J Roentgenol. 1991:156:903-8.
7. Piskin G, Akyol A, Uzar H, Tulek N, Boyvat A, Gurgey E. Comparative evaluation of Type 1 latex hypersensitivity in patients with chronic urticaria, rubber factory workers and healthy control subjects. Contact Dermatitis. 2003:48:266-71.
8. Bernardini R, Novembre E, Lombardi E, Mezzetti P, Cianferoni A, Danti DA, Mercurella A, Vierucci A. Risk factors for latex allergy in patients with spina bifida and latex sensitization. Clin Exp Allergy. 1999:29:681-6.
9. Cremer R, Lorbacher M, Hering F, Engelskirchen R. Natural rubber latex sensitisation and allergy in patients with spina bifida, urogenital disorders and oesophageal atresia compared with a normal paediatric population. Eur J Pediatr Surg. 2007:17:194-8.
10. Holme SA, Lever RS. Latex allergy in atopic children. Br J Dermatol. 1999:140:919-21.
11. Bollinger ME, Mudd K, Keible LA, Hess BL, Bascom R, Hamilton RG. A hospital-based screening program for natural rubber latex allergy. Ann Allergy Asthma Immunol. 2002:88:560-7.
12. Bousquet J, Flahault A, Vandenplas O, Ameille J, Duron JJ,

Pecquet C, Chevrie K, nnesi-Maesano I. Natural rubber latex allergy among health care workers: a systematic review of the evidence. J Allergy Clin Immunol. 2006:118:447-54.
13. Arellano R, Bradley J, Sussman G. Prevalence of latex sensitization among hospital physicians occupationally exposed to latex gloves. Anesthesiology. 1992:77:905-8.
14. Chen YH, Lan JL. Latex allergy and latex-fruit syndrome among medical workers in Taiwan. J Formos Med Assoc. 2002:101:622-6.
15. Lai CC, Yan DC, Yu J, Chou CC, Chiang BL, Hsieh KH. Latex allergy in hospital employees. J Formos Med Assoc. 1997:96:266-71.
16. Lin CT, Hung DZ, Chen DY, Wu HJ, Lan JL, Chen YH. A hospital-based screening study of latex allergy and latex sensitization among medical workers in Taiwan. J Microbiol Immunol Infect. 2008:41:499-506.
17. Liss GM, Sussman GL. Latex sensitization: occupational versus general population prevalence rates. Am J Ind Med 1999; 35:196-200.
18. Nel A, Gujuluva C. Latex antigens: identification and use in clinical and experimental studies, including crossreactivity with food and pollen allergens. Ann Allergy Asthma Immunol. 1998:81:388-96.
19. Kurup VP, Alenius H, Kelly KJ, Castillo L, Fink JN. A two-dimensional electrophoretic analysis of latex peptides reacting with IgE and IgG antibodies from patients with latex allergy. Int Arch Allergy Immunol. 1996:109:58-67.
20. Czuppon AB, Chen Z, Rennert S, Engelke T, Meyer HE, Heber M, Baur X. The rubber elongation factor of rubber trees (Hevea brasiliensis) is the major allergen in latex. J Allergy Clin Immunol. 1993:92:690-7.
21. Arif SA, Hamilton RG, Yusof F, Chew NP, Loke YH, Nimkar S, Beintema JJ, Yeang HY. Isolation and characterization of the early nodule-specific protein homologue (Hev b 13), an allergenic lipolytic esterase from Hevea brasiliensis latex. J Biol Chem. 2004:279:23933-41.
22. Posch A, Chen Z, Dunn MJ, Wheeler CH, Petersen A, Leubner-Metzger G, Baur X. Latex allergen database. Electrophoresis. 1997:18:2803-10.
23. Yip E, Cacioli P. The manufacture of gloves from natural rubber latex. J Allergy Clin Immunol. 2002:110:S3-14.
24. Hamilton RG, Peterson EL, Ownby DR. Clinical and laboratory-based methods in the diagnosis of natural rubber latex allergy. J Allergy Clin Immunol. 2002:110:S47-S56.
25. Pittner G, Vrtala S, Thomas WR, Weghofer M, Kundi M, Horak F, Kraft D, Valenta R. Component-resolved diagnosis of house-dust mite allergy with purified natural and recombinant mite allergens. Clin Exp Allergy. 2004:34:597-603.
26. Astier C, Morisset M, Roitel O, Codreanu F, Jacquenet S, Franck P, Ogier V, Petit N, Proust B, Moneret-Vautrin DA, Burks AW, Bihain B, Sampson HA, Kanny G. Predictive value of skin prick tests using recombinant allergens for diagnosis of peanut allergy. J Allergy Clin Immunol. 2006:118:250-6.
27. Baumann MA, Rath B, Fischer JH, Iffland R. The permeability of dental procedure and examination gloves by an alcohol based disinfectant. Dent Mater. 2000:16:139-44.
28. Hunt LW, Kelkar P, Reed CE, Yunginger JW. Management of occupational allergy to natural rubber latex in a medical center: the importance of quantitative latex allergen measurement and objective follow-up. J Allergy Clin Immunol. 2002:110:S96-106.
29. Lee MF, Chen YH, Lin HC, Wang HL, Hwang GY, Wu CH. Identification of hevamine and Hev b 1 as major latex allergens in Taiwan. Int Arch Allergy Immunol. 2006:139:38-44.
30. Lee MF, Tsai JJ, Hwang GY, Lin SJ, Chen YH. Identification of immunoglobulin E (IgE)-binding epitopes and recombinant IgE reactivities of a latex cross-reacting Indian jujube Ziz m 1 allergen. Clin Exp Immunol. 2008:152:464-71.
31. Alenius H, Makinen-Kiljunen S, Turjanmaa K, Palosuo T, Reunala T. Allergen and protein content of latex gloves. Ann Allergy. 1994:73:315-20.
32. Palosuo T, Alenius H, Turjanmaa K. Quantitation of latex allergens. Methods. 2002:27:52-8.
33. Surgeon's and patient examination gloves: Reclassification and medication glove guidance manual availability; proposed rule and notice. Fed Regist. 1999:64:41709-43.
34. Baur X, Chen Z, Raulf-Heimsoth M, Degens P. Protein and allergen content of various natural latex articles. Allergy. 1997:52:661-4.
35. Reinikka-Railo H, Kautiainen H, Alenius H, Kalkkinen N, Kulomaa M, Reunala T, Turjanmaa K. Latex allergy: the sum quantity of four major allergens shows the allergenic potential of medical gloves. Allergy. 2007:62:781-6.

Manuscript received January 4, 2010; accepted for publication February 26, 2010.

Yi-Hsing Chen

Division of Allergy, Immunology and Rheumatology
Taichung Veterans General Hospital
160 Taichung Harbor Road, Section 3,
Taichung 40705, Taiwan
E-mail: ysanne@vghtc.gov.tw, mflee@vghtc.gov.tw