

Specific IgE and IgG4 antibodies to Japanese cedar pollen and total IgE antibody in lumbermen

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Abstract

Serum levels of specific IgE and IgG4 antibodies to Japanese cedar (*Cryptomeria japonica*) pollen and total IgE antibody in 75 lumbermen and in 53 male office workers at an urban establishment were measured by means of an enzyme linked immunosorbent assay (ELISA) and compared. No significant differences of specific IgE and IgG4 to cedar pollen and total IgE were found between the lumbermen and the office workers. There were no significant differences of incidence of cedar pollinosis and positive (greater than 100 FU/ml) rate of serum specific IgE between the two groups, though the lumbermen were exposed to dense concentrations of cedar pollen in their working area. In the lumbermen who showed positive values of specific IgE, the mean value of the specific antibody in Japanese cedar pollinosis lumbermen was significantly higher than that in symptom-free lumbermen, while no significant differences of serum level of specific IgG4 were found between the two groups. © 1998 Published by Elsevier Science Ireland Ltd. All rights reserved.

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1. Introduction

In the recent decades, the number of Japanese cedar (*Cryptomeria japonica*) pollinosis patients has increased in Japan for two suggestive reasons. Increase in Japanese cedar pollen counts was pointed out as a principal cause, while environmental pollution and our life style including foods also may be another possibility. If exposure to a great amount of Japanese cedar pollen is a sole cause, lumbermen working in Japanese cedar forests throughout pollinating season might be expected as the most affected in Japan.

To determine the incidence of Japanese cedar pollinosis and the extent of serum IgE antibody to Japanese cedar pollen in lumbermen, we assessed and compared serum level of specific IgE and IgG4 antibodies to

Japanese cedar pollen and total IgE antibody in lumbermen and in office workers at an urban establishment in this study.

2. Materials and methods

A total of 75 volunteer lumbermen employed by the Aichi Forestry Association (aged 21–67 years, with a mean age of 47.7 years) who were working in the woodland near Shinshiro city and 53 male volunteer office workers at an urban establishment located in an industrial large city as a contrast (aged 24–53 years, with a mean age of 43.9 years) were assessed. The lumbermens' working period ranged from 1 to 50 years with a mean of 20.9 years and the office workers had no occupational history of lumbering.

Cry j I protein, one of major allergens of Japanese cedar pollen, was prepared according to the methods of

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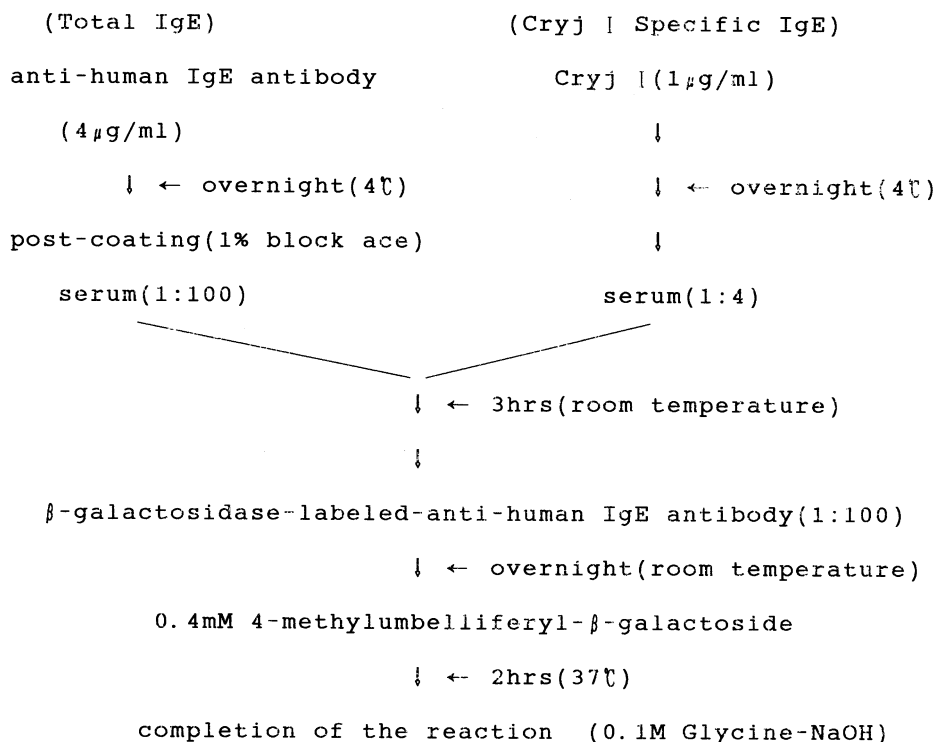


Fig. 1. Method of measurement of serum total IgE antibody and Cry j I specific IgE antibody.

Yasueda et al. (1983). Serum level of specific IgE and IgG4 antibodies to Cry j I antigen and total IgE antibody (Tago, USA) were measured by means of an indirect enzyme-linked immunosorbent assay (ELISA) in the lumbermen and the office workers immediately after the pollinating season of cedar in 1996. β -Galactosidase-conjugated anti-human IgE antibody (Pharmacia, Sweden) was employed as the second antibody and 4-methylumbelliferyl- β -galactoside (Sigma, USA) as a fluorogenic enzyme substrate respectively (Fig. 1) according to methods introduced by Sakaguchi et al. (1990). Peroxidase-conjugated anti-human IgG4 antibody (Yamasa, Japan) was employed as the second antibody in this ELISA (Fig. 2) introduced by Nerurkar et al. (1984) and Miyazawa et al. (1988).

The unpaired *t*-test and χ^2 test were applied for statistical significance.

3. Results

Japanese cedar pollen including Japanese cypress (*Chamaecyparis obtuse*) pollen was collected with a rotary type pollen sampler and counted within the area $1 \times 1 \text{ cm}^2$ in Shinshiro (woodland) and Nagoya (industrial large city) during 1990–1996 as shown in Fig. 3. The pollen counts in the woodland were 3–5 times higher than those in the urban industrial area.

No significant differences in age between the lumbermen and the office workers were found.

The mean value of serum total IgE was $233.13 \pm 348.08 \text{ U/ml}$ in the lumbermen and $238.04 \pm 294.99 \text{ U/ml}$ in the urban office workers. Mean value of serum specific IgE to Cry j I was $313.91 \pm 463.44 \text{ FU/ml}$ in the lumbermen and $246.30 \pm 389.54 \text{ FU/ml}$ in the office workers. The mean value of serum specific IgG4 to Cry j I was $67.40 \pm 199.15 \text{ U/ml}$ in the lumbermen and $80.13 \pm 232.59 \text{ U/ml}$ in the office workers. There were no significant differences in total IgE and specific IgE and IgG4 between the two groups.

The positive value of serum specific IgE to Cry j I was greater than 100 FU/ml in this method. A total of 29 lumbermen (38.7%) of the 75, and 20 office workers (37.7%) of the 53 showed positive values of specific IgE to Cry j I respectively. Thirteen lumbermen (17.3%) of the 75, and 10 office workers (18.9%) of the 53 suffered from Japanese cedar pollinosis respectively. No significant differences in the positive rate of serum specific IgE to Cry j I and the incidence of cedar pollinosis were found between the two groups.

In the 29 lumbermen who showed positive values of ELISA titer of specific IgE antibody to Cry j I, the mean value of the antibody in 16 symptom-free subjects was $585.38 \pm 365.86 \text{ FU/ml}$ and in 13 pollinosis sufferers $1051.31 \pm 341.88 \text{ FU/ml}$ respectively, and a significant difference between the two groups was found ($P < 0.01$).

In the same subjects, the mean value of specific IgG4 to Japanese cedar pollen in symptom-free lumbermen was $175.12 \pm 327.02 \text{ U/ml}$ and in pollinosis sufferers

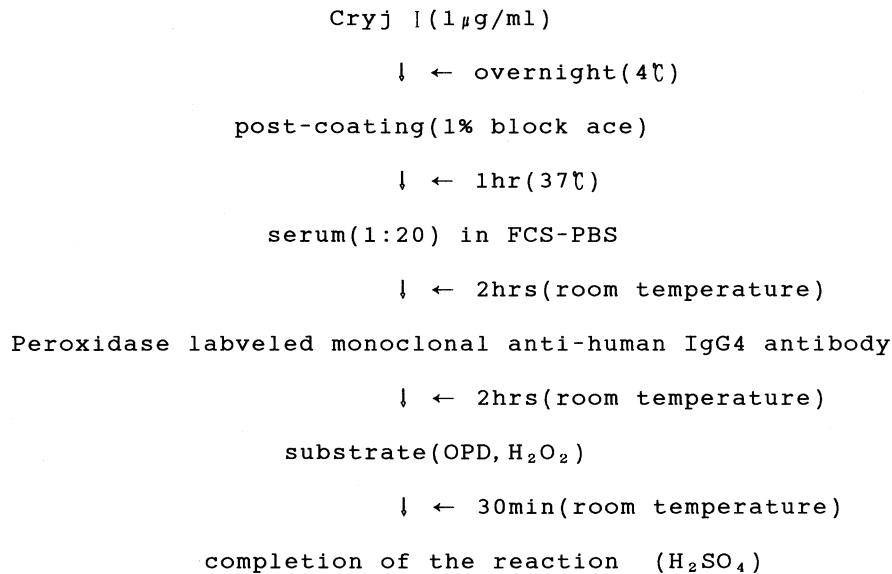


Fig. 2. Method of measurement of serum IgG4 antibody to Cryj I.

70.03 $\pm$ 92.83 U/ml, and no significant differences between the two groups were found.

4. Discussion

Japanese cedar and cypress trees have been planted under the direction of the Japanese government since the second world war for industrial prompt use in Japan. As a result, cedar and cypress comprise more than 80% of Japanese forests. Cedar trees older than 30 years acquire the ability to pollinate. Thus, in re-

cent decades, the small islands have become covered with fully pollinating cedar trees. Unfortunately, changes of recent industrial system of Japan has promoted importation of cheaper timber from foreign countries and it has caused Japanese cedar forestry to decay. Cedar plantations without suitable management increase the pollen amount for preservation of their species. These are probable reasons for the increasing airborne pollen counts of Japanese cedar in Japan (Saito, 1996).

Increase in ambient antigen is one of the most likely reasons for the increase in allergic disease. Imaoka et al. (1996) reported that higher cedar pollen counts tended to increase in population of cedar pollinosis patients. If the supposition is true, cedar pollinosis would be the most common in lumbermen working in the Japanese cedar forests and exposed to dense concentrations of cedar pollen. In our present study, however, no significant differences of incidence of Japanese cedar pollinosis and positive rate of serum specific IgE to Japanese cedar pollen between lumbermen and urban office workers were found. Furthermore, no significant differences of serum level of specific and total IgE between the two groups were found. According to these results, only increase in exposure to cedar pollen does not seem to be the major reason of increase in Japanese cedar pollinosis.

Environmental pollutants, for instance NO_x and DEP (diesel exhausted particle), are considered to be a cause of increase in pollinosis (Saxon et al., 1996), as well a cause of exacerbation symptoms of pollinosis to cedar (Yokoyama et al., 1992). High concentrations of indoor NO₂ from heating also exacerbate the pollinosis symptoms (Yokoyama et al., 1996).

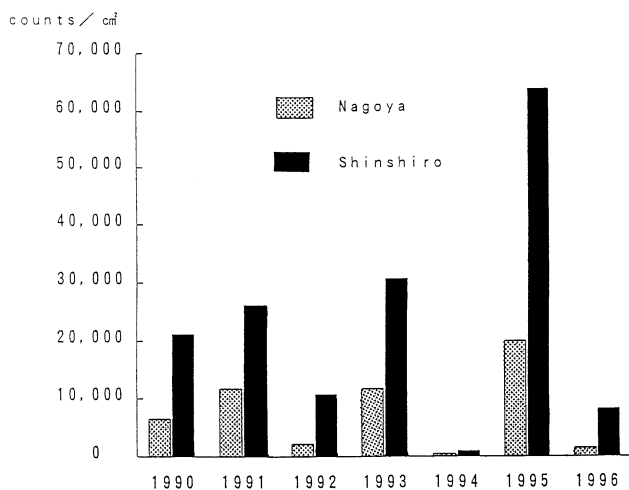


Fig. 3. Pollen counts of Japanese cedar (*Cryptomeria japonica*) including Japanese cypress (*Chamaecyparis obtuse*) in Nagoya (industrial large city) and Shinshiro (woodland) during the period between 1990 and 1996.

Clean mountain air may protect lumbermen from promotion of sensitization with cedar pollen and, when they are sensitized, it may diminish pollinosis symptoms despite exposure to dense concentrations of cedar pollen.

In the lumbermen who showed positive values of sepecific IgE to Japanese cedar pollen antigen, the mean value of the specific antibody in Japanese cedar pollinosis sufferers was significantly higher than that in symptom-free lumbermen, while no significant differences of the mean value of specific IgG4 to cedar pollen between the two groups were found. The findings suggest that the subjects with higher values of serum specific IgE to Japanese cedar pollen tend to suffer from cedar pollinosis.

Specific IgG4 is considered to be a blocking antibody against the anaphylactic type allergy. Hyposensitization to antigen that helps allergic patients and increase in antigen specific IgG4 is regarded as a appropriate indicator of the response to immunotherapy (Loveless, 1940; Nakagawa et al., 1983; Gracia et al., 1988; Nakagawa, 1991). On the contrary, Gwyman et al. (1982) reported the value of serum specific IgG4 to mite in severe atopic dermatitis to be high. Takagi (1992) also mentioned that a high value of specific IgG4 to Japanese cedar pollen in children tended to exacerbate pollinosis symptoms. Evidence of natural hyposensitization in the lumbermen exposed to dense concentrations of cedar pollen was not strong according to our study, and further investigations are required to determine role of specific IgG4 antibody in allergic patients.

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References

- Gracia BE, Sanz ML, Oehling A. Effect of immunotherapy on antigen-specific sensitive to *D. ptenonyssinus*. *Allergol Immunopathol* 1988;16:379–83.
- Gwyman CM, Almosawi T, Stanwrth DR. Clinical association with serum allergen-specific IgG4 antibodies. *Clin Allergy* 1982;12:459–64.
- Imaoka K, Miyazawa H, Nishihara S, Sakaguchi M, Inouye S. Effect of pollen exposure on serum IgE and IgG antibody responses in Japanese cedar pollinosis patients. *Allergol Int* 1996;45:159–62.
- Loveless MH. Immunological studies of pollinosis. *J Immunol* 1940;38:25.
- Miyazawa H, Inouye S, Sakaguchi M, Koizumi K. A reverse-sandwich ELISA for IgG antibody to a pollen allergen. *J Allergy Clin Immunol* 1988;82:407–13.
- Nakagawa T, Takaishi T, Sakamoto Y, Ito K, Miyamoto T, Skvaril F. IgG4 antibodies in patients with house-dustmite sensitive bronchial asthma: relationship with antigenspecific immunotherapy. *Int Arch Allergy Appl Immunol* 1983;71:122–5.
- Nakagawa T. The role of IgG subclass antibodies in the clinical response to immunotherapy in allergic disease. *Clin Exp Allergy* 1991;21:289–96.
- Nerurkar LS, Namba M, Brasheare G, Jacob AJ, Lee YJ, Sever JL. Rapid detection of herpes simplex virus in clinical specimens by use of a capture bition-streptavidin enzyme-linked immunosorbent assay. *J Clin Microbiol* 1984;20:109–14.
- Saito Y. Airborne pollen survey of *Cryptomeria japonica* and cupressaceae in Yushima, Bunkyo-ku, Tokyo in 1996 and past 20 years (1977–1996) (In Japanese with English abstract). *Jpn J Palynol* 1996;42:149–53.
- Sakaguchi M, Inouye S, Taniai M, Ando S, Matuhasi T. Identification of the second major allergen of Japanese cedar pollen. *Jpn J Allergol* 1990;45:309–12.
- Saxon A, Zhang K, Takenaka H, Casillas A, DaizSanches D. The role of diesel exhaust in the allergic antibody response of the human airway. *J Jpn Immunol Allergol Otolaryngol* 1996;14(2):9–11.
- Takagi M. Environmental and immunological survey of sensitization with Japanese cedar (*Cryptomeria japonica*) pollen in children (In Japanese with English abstract). *Jpn J Allergol* 1992;41:1540–6.
- Yasueda H, Yui Y, Shimizu T, Shida T. Isolation and partial characterization of the major allergen from Japanese cedar (*Cryptomeria japonica*) pollen. *J Allergy Clin Immunol* 1983;71:77–86.
- Yokoyama N, Naito K, Ibata K, Ishihara M, Takeda N, Senoh Y, Iwata S. A clinical comparison of Japanese cedar pollinosis patients at two different areas in Aichi prefecture (In Japanese). *J Jpn Immunol Allergol Otolaryngol* 1992;10(2):120–1.
- Yokoyama N, Naito K, Senoh Y, Ishihara M, Ibata K, Iwata S, Ishii J. The influence of indoor air pollution on symptoms in Japanese cedar pollinosis (In Japanese). *J Jpn Immunol Allergol Otolaryngol* 1996;14(2):168–9.