

Decrease of Specific and Total IgE Levels in Allergic Patients After BCG Vaccination

Preliminary Report

Giovanni P. Cavallo, MD; Mariateresa Elia, MD; Daniela Giordano, MD; Cristina Baldi, MD; Raffaella Cammarota, MD

Background: A systemic reaction to mycobacteria biases the balance of T helper cell types 1 and 2 toward T helper cell type 1. BCG vaccination mimics some characteristics of mycobacterial infection. Children who have undergone tuberculin conversion after BCG vaccination seem to be more likely to lose their atopic symptoms. Inhibition of both allergic response and airway hyperreactivity after vaccination for mycobacteria has been observed in animal experiments.

Objective: To evaluate the effects that BCG vaccination has on the serological status of allergic people.

Participants and Methods: This study included 20 volunteers with a history of allergic rhinitis who were required to undergo BCG vaccination by Italian law. Epicutaneous allergy testing with a panel of common seasonal and perennial inhalational allergens and 2 blood

withdrawals were performed. The serum total IgE levels and the serum allergen-specific IgE levels of each individual were measured just before BCG vaccination and again 4 months later. Total IgE levels were determined using the paper radioimmunosorbent test, and allergen-specific IgE levels were determined using the radioallergosorbent test.

Results: Total IgE and allergen-specific IgE levels were significantly decreased after BCG vaccination ($P = .004$ and $P < .001$, respectively).

Conclusion: BCG, an effective stimulus for cell-mediated immunity, deserves further study to evaluate its ability to modulate the immune response associated with allergic rhinitis.

Arch Otolaryngol Head Neck Surg. 2002;128:1058-1060

THELPER CELLS play a critical role in controlling immune responses. According to their pattern of cytokine production, T helper cells are classified into functionally distinct subsets, including type 1 (T_H1) and type 2 (T_H2) cells.¹⁻⁴ The T_H1 cells mainly produce interleukin (IL)-2, interferon γ , and tumor necrosis factor β , whereas the T_H2 cells produce IL-4, IL-5, IL-6, IL-9, IL-10, and IL-13. T helper type 1 cells are critical for the induction of cell-mediated immunity and pathological disorders such as diabetes. In contrast, T_H2 cells bias the response toward antibody production and allergy.⁵ Late phases of allergic response are often associated with an increased expression of T_H2 -type cytokines, such as IL-4 and IL-5. Interleukin 4 is essential for isotype switching toward IgE, while IL-5, together with IL-4, switches antibody response toward IgA.⁵⁻⁸ On the other hand, T_H1 cells have a down-regulatory role in the production of T_H2 cytokines by releasing interferon γ .^{5,7,9}

Differentiation between T_H1 and T_H2 is biased by antigen dose and structure, the concurrence of distinct costimulatory molecules, the kind of cells presenting the antigenic peptides, and the presence of particular cytokines in the microenvironment.¹⁰⁻¹² A T_H1 - T_H2 imbalance with an increase in T_H2 cells favors IgE production and the establishment of allergic reactivity.

In the last 20 years, allergies have increased in civilized countries.¹³ Residency in urban or industrial areas, changes in lifestyle, and the falling incidence of microbial infections may be responsible for this increase, to some extent. The diminishing incidence of tuberculosis could well be one of the many contributing factors. A marked production of T_H1 cytokines is a hallmark of the systemic reaction to mycobacteria. The subsequent inclination of the T_H1 - T_H2 balance toward T_H1 may be responsible for an inverse association between exposure to *Mycobacterium tuberculosis* and allergic diseases.^{14,15}

Even if it is unlikely that BCG vaccination will entirely replace the complex

From the Dipartimento di Fisiopatologia Clinica, I Clinica Otorinolaringoiatrica, University of Turin, Turin, Italy.

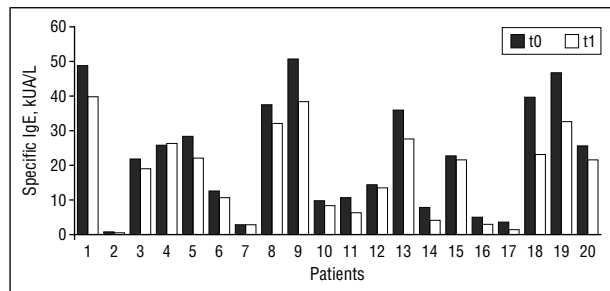


Figure 1. Specific IgE levels, calculated as the mean value for each patient, before (t0) and 4 months after (t1) BCG vaccination.

and long-lasting T_H1 stimulation produced by mycobacterial infection, it does mimic some of its characteristics. Children who have undergone tuberculin conversion after BCG vaccination seem to be more likely to lose their atopic symptoms.¹⁶ Moreover, a decrease in IgE production and the inhibition of allergic response and airway hyperreactivity after vaccination for mycobacteria have been observed in animal experiments.¹⁷⁻²⁰

Our goal was to evaluate the effects of BCG vaccination on the serological status of allergic persons.

PARTICIPANTS AND METHODS

From October to November 1997, volunteers were recruited at the Antituberculosis Dispensary, Turin, Italy. They were physicians, nurses, and medical students who presented with negative Mantoux test results and were required by Italian law to undergo BCG vaccination. Informed written consent was obtained for epicutaneous allergy testing with a panel of common seasonal and perennial inhalational allergens and, if the subject was enrolled in the study, for 2 blood withdrawals. Subjects with anamnesis of allergic rhinitis were probed with epicutaneous allergy testing and were recruited into the study only if the skin test was positive for at least one of the following allergens: mites, cats, dogs, mixed molds, grass, or trees. The inclusion criterion was the anamnesis of allergic rhinitis associated with a positive skin test result.

Subjects with a history of previous antiallergy vaccination, immunomodulatory treatments, and anti-influenza vaccination were excluded. A total of 17 women and 3 men (age range, 19-37 years) with a skin test that was positive for a series of ubiquitous inhalational allergens were enrolled in the study. Volunteers were required not to take any allergy medicines during the entire study.

The serum total IgE levels and the serum allergen-specific IgE levels of each individual were determined just before BCG vaccination and again 4 months later by means of commercially available enzyme immunoassay. The blood samples of each subject, collected before and after BCG vaccination, were labeled with a mark, coagulated at room temperature, centrifuged, frozen, stored at -20°C , and tested with a unique assay at the end of the collecting time. Total IgE levels (kU/L) were determined using a paper radioimmunosorbent test (Pharmacia, Uppsala, Sweden). Allergen-specific IgE levels (kU/L) for an inhalational (seasonal or perennial) allergen against which the volunteer was reactive were determined with a radioallergosorbent test (Pharmacia). The levels of serum allergen-specific IgE determined before and after BCG vaccination were calculated as the mean and SD of all the allergen-specific IgE values. Statistical analysis of IgE variations was performed using a paired *t* test. For each patient, the levels of specific IgE before and after BCG vaccination were calculated as the mean

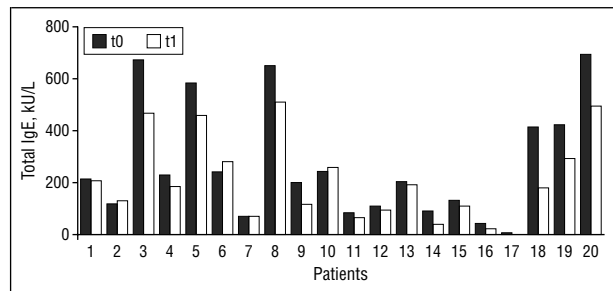


Figure 2. Values of total IgE before (t0) and 4 months after (t1) BCG vaccination.

value of the allergen-specific IgE values. The variation of serum specific IgE levels against perennial-only allergens was also evaluated.

RESULTS

All the subjects enrolled in the study reported an anamnesis of allergic rhinitis and demonstrated skin tests that were positive for at least 2 inhalational allergens, while the tests of 11 of the 20 volunteers were positive for perennial allergens.

Serum specific IgE levels, calculated as the mean value of the allergen-specific IgE values of each patient, were lower in 18 of the 20 subjects 4 months after BCG vaccination; only 2 cases demonstrated a slight augmentation (**Figure 1**). Total IgE levels in serum samples obtained 4 months after BCG vaccination were lower in 18 of the 20 volunteers (**Figure 2**).

The levels of serum specific IgE determined before and after BCG vaccination, calculated as mean $\pm$ SD of all the allergen-specific IgE values, were 25 ± 31 and 19 ± 27 kU/L, respectively. Serum total IgE levels determined before and after BCG vaccination were 269 ± 221 and 210 ± 162 kU/L, respectively. Allergen-specific IgE and total IgE levels were significantly lower 4 months after BCG vaccination ($P = .004$ and $P < .001$, respectively). Five volunteers displayed negative radioallergosorbent test values (<0.35 kU/L) for one or more allergens to which they were positive before BCG vaccination. The values of serum specific IgE against perennial-only allergens were significantly decreased ($P = .004$) 4 months after BCG vaccination.

COMMENT

The requirements imposed by Italian law on obligatory BCG vaccination of physicians, nurses, and medical students with negative Mantoux test results enabled us to collect data from an allergic adult population with a fully mature immune system before and after BCG vaccination. A significant difference between the serum allergen-specific and total IgE levels determined before BCG vaccination and 4 months later was evident from the analysis of the data.

The skin tests of the volunteers enrolled in the study were positive for both perennial and seasonal allergens. However, the levels of serum specific IgE against perennial-only allergens were also significantly decreased after BCG vaccination, thus indicating that the decrease

in specific IgE levels was not related to a seasonal variation in IgE serum levels. BCG vaccination induced a decrease in IgE serum levels; furthermore, it is notable that BCG vaccination decreases a preexistent T_H2 response in a nonspecific manner.

The anti-influenza vaccination was considered an exclusion criterion because it is usually performed in the fall, when mandatory BCG vaccination is largely performed. Therefore, it may have introduced an unrelated modulation of the immune system that adds an additional confusion factor.

The short-term effects of BCG vaccination were evaluated in terms of the variation in serum IgE levels in adult subjects suffering from allergic rhinitis. Controversial results are reported in other human studies on the effect of BCG vaccination in asthmatic subjects.²¹⁻²⁴ No significant difference in IgE levels was found in asthmatic patients who were intradermally injected with *Mycobacterium vaccae* compared with a placebo group.²¹ Wheezing BCG-vaccinated children demonstrated no significant differences in IgE levels when compared with a nonvaccinated group.²² The discrepancies in our findings may be accounted for by the age of the vaccinated subjects,²² the time of vaccination,²² and the period in which the effects were evaluated.^{21,22} By contrast, fewer symptoms and increased interferon γ production were described in rhinitic and asthmatic patients who were intradermally treated with *M vaccae*.²³ Furthermore, successful BCG vaccination appeared to inhibit the development of atopic disorders (including asthma) in children, although the preventive effect of BCG vaccination did not last long.²⁴ Our data supported the findings that BCG vaccine would be able to reduce a preexistent T_H2 response in persons with allergic rhinitis. A study of IgE serum levels in adult asthmatic subjects before and after BCG vaccination would be an excellent follow-up step to take.

More convincing data may emerge after long-term evaluation of the significance and persistence of IgE responses in a larger number of subjects; however, our preliminary findings indicate that BCG vaccination may be valuable for modulating the allergic profile in atopic subjects. An accurate symptom evaluation is not currently available. It is now being studied in detail on the basis of the present serum data. The forthcoming results will be described separately.

CONCLUSIONS

Expansion of T_H1 or T_H2 can be selectively induced using appropriate antigenic stimuli. The BCG vaccine is an effective stimulus for cell-mediated immunity and may be used to modulate immune response in atopic subjects to reduce allergic reactivity. The data from this study encourage us to continue the study of the effects of BCG vaccination on the magnitude and duration of IgE responses in a larger number of individuals in order to set a protocol for its use in atopic subjects.

Accepted for publication February 25, 2002.

This study was supported by grant 96.03034.CTO4 from the National Research Council.

We wish to thank Guido Forni, MD, for his valuable support, as well as all the personnel of the Antituberculosis Dispensary, Turin, Italy.

Corresponding author and reprints: Giovanni P. Cavallo, MD, Dipartimento di Fisiopatologia Clinica, I Clinica Otorinolaringoiatrica, University of Turin, Via Genova No. 3, 10126 Turin, Italy (e-mail: mt.eli@virgilio.it).

REFERENCES

1. Mosmann TR, Cherwinski H, Bond MW, Giedlin MA, Coffman RL. Two types of mouse helper T cell clone, I: definition according to profiles of lymphokine activities and secreted proteins. *J Immunol*. 1986;136:2348-2357.
2. Romagnani S. Type 1 T helper and type 2 T helper cells: functions, regulation and role in protection and disease. *Int J Clin Lab Res*. 1991;21:152-158.
3. Abbas AK, Murphy KM, Sher A. Functional diversity of helper T lymphocytes. *Nature*. 1996;383:787-793.
4. Paul WE, Seder RA. Lymphocyte response and cytokines. *Cell*. 1994;76:241-251.
5. Mosmann TR, Sad S. The expanding universe of T-cell subsets: Th1, Th2 and more. *Immunol Today*. 1996;17:138-146.
6. Leung DYM, Gheea RS. Regulation of the human IgE responses. *Int Rev Immunol*. 1987;2:75-91.
7. Pène J, Rousset F, Briere F, et al. IgE production by normal human B cells induced by alloreactive T-cell clones is mediated by IL-4 and suppressed by IFN- γ . *J Immunol*. 1988;141:1218-1224.
8. Kay AB. T lymphocytes and their products in atopic allergy and asthma. *Int Arch Allergy Appl Immunol*. 1991;94:189-193.
9. Maggi E, Parronchi P, Manetti R, et al. Reciprocal regulatory effects of IFN- γ and IL-4 on the in vitro development of human Th1 and Th2 clones. *J Immunol*. 1992;148:2142-2147.
10. O'Garra A, Murphy K. Role of cytokines in development of Th1 and Th2 cells. *Chem Immunol*. 1996;63:1-13.
11. Constant SL, Bottomly K. Induction of Th1 and Th2 CD4⁺ T cells responses: the alternative approaches. *Annu Rev Immunol*. 1997;15:297-322.
12. O'Garra A. Cytokines induce the development of functionally heterogeneous T helper cell subsets. *Immunity*. 1998;8:275-283.
13. Romagnani S. Atopic allergy and other hypersensitivities interactions between genetic susceptibility, innocuous and/or microbial antigens and the immune system. *Curr Opin Immunol*. 1997;9:773-775.
14. Rook GA, Stanford JL. Give us this day our daily germs. *Immunol Today*. 1998;19:113-116.
15. von Mutius E, Pearce N, Beasley R, et al. International patterns of tuberculosis and the prevalence of symptoms of asthma, rhinitis and eczema. *Thorax*. 2000;55:449-453.
16. Shirakawa T, Enomoto T, Shimazu S, Hopkin JM. The inverse association between tuberculin responses and atopic disorder. *Science*. 1997;275:77-79.
17. Wang CC, Rook GA. Inhibition of an established allergic response to ovalbumin in BALB/c mice by killed *Mycobacterium vaccae*. *Immunology*. 1998;93:307-313.
18. Herz U, Gerhold K, Grüber C, et al. BCG infection suppresses allergic sensitization and development of increased airway reactivity in an animal model. *J Allergy Clin Immunol*. 1998;102:867-874.
19. Bakir M, Tukenmez F, Bahceciler NN, Barlan IB, Basaran MM. Heat-killed *Mycobacterium bovis*-bacillus Calmette-Guérin-suppressed total serum IgE response in ovalbumin-sensitized newborn mice. *J Asthma*. 2000;37:329-334.
20. Nahori MA, Lagranderie M, Lefort J, et al. Effects of *Mycobacterium bovis* BCG on the development of allergic inflammation and bronchial hyperresponsiveness in hyper-IgE BP2 mice vaccinated as newborns. *Vaccine*. 2001;19:1484-1495.
21. Shirlcliffe PM, Easthope SE, Cheng S, et al. The effect of delipidated deglycolipidated (DDMV) and heat-killed *Mycobacterium vaccae* in asthma. *Am J Respir Crit Care Med*. 2001;163:1410-1414.
22. Grüber C, Kulig M, Bergmann R, et al. Delayed hypersensitivity to tuberculin, total immunoglobulin E, specific sensitization, and atopic manifestation in longitudinally followed early bacille Calmette-Guérin-vaccinated and nonvaccinated children. *Pediatrics*. 2001;107:E36.
23. Hopkin JM, Shaldon S, Ferry B, et al. Mycobacterial immunisation in grass pollen asthma and rhinitis. *Thorax*. 1998;53(suppl 4):S63.
24. Yoneyama H, Suzuki M, Fujii K, Odajima Y. The effect of DPT and BCG vaccinations on atopic disorders. *Arerugi*. 2000;49:585-592.