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Percutaneous or intradermal BCG vaccine?

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Abstract

Objective: To compare the intradermal and percutaneous routes of BCG administration.

Sources of data: A review of the literature published between 1987 and 2002 was carried out in the MEDLINE and Lilacs databases. The following key words were used: BCG vaccine/administration, adverse effects, efficacy, tuberculosis/prevention and control. Some articles published before 1987 were included because of their relevance to the topic.

Summary of the findings: There are no clinical studies comparing the efficacy of intradermal and percutaneous BCG. Percutaneous BCG causes a weaker reaction, however it is also less efficient in stimulating gamma-interferon production by Th1-lymphocytes, which is considered as the best marker of the anti-tuberculin immune response.

Conclusions: *In vivo* and *in vitro* studies suggest a better immune response with intradermal BCG. The intradermal method should be recommended for BCG administration.

J Pediatr (Rio J). 2004;80(2):93-8: BCG vaccine/administration, adverse effects, efficacy, tuberculosis/prevention and control.

Introduction

Despite the fact that the BCG vaccine is one of the most widely used globally, tuberculosis remains one of the most significant public health problems [1,2]. In Brazil, in the year 2000, 94,360 new tuberculosis cases were registered, even though there was a significant fall in the number of tuberculous meningitis cases among infants related to increase vaccine coverage.[3]

A number of different factors have been blamed for the BCG vaccines's variable efficacy; factors related to the host, the environment, the strain from which the vaccine is produced and the dose and method of administering the vaccine.[1,2]

The BCG vaccine was first used, orally, in 1921, and later came to be administered cutaneously (BCG-ID or BCG-PC), due to the better induction of delayed type hypersensitivity (DTH) response to the tuberculin skin test, lower cost and reduced number of adverse events.[4-8]

The BCG vaccine used in Brazil contains live Calmette Guérin bacilli, attenuated and lyophilized, from the Moreau-Rio de Janeiro strain. The Fundação Ataulpho de Paiva (RJ) produces intradermal BCG vaccine, available in 1 mg, 2 mg or 5 mg offerings (yielding 10, 20 or 50 doses) and concentrated vaccine for percutaneous use in 40 mg packaging. Both versions must be reconstituted according to the manufacturer's instructions before administration at the level of the insertion of the deltoid muscle.

While the Health Ministry recommends the administration of 0.1 ml of the BCG vaccine (1 mg/ml), intradermally,[3] in many private clinics, percutaneous BCG is used.

As we have pointed out, the vaccine produced for percutaneous use is concentrated (40 mg). The percutaneous vaccine must be made up with 1.0 ml of saline (40 mg/ml, contains 200 million live bacilli). It is recommendable to spread a drop of reconstituted vaccine over an area 1.5 cm wide by 3 cm long at the insertion of the deltoid muscle, using the edge of the multipuncture device itself, to be used for the percutaneous administration.

The multipuncture unit is a cylinder-like device with small needles, which should be pressed firmly against the skin, within the area where the vaccine was spread. It is also recommended that pressure is applied to the skin, in an adjacent area, a second time and once more spread the vaccine across the area.

The patient should not expose the area to sunlight or liquids until the vaccine has tried.

In contrast with the intradermally technique, the percutaneous method does not allow the dose to be estimated since it is much more difficult to evaluate the dose injected into the *stratum corneum*. Furthermore, variable application techniques and differing percutaneous vaccine administration devices can alter the number of bacilli actually given and change the response to vaccination.[6-9]

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This article will describe the results of a literature review of the controversies surrounding the issue of the best BCG administration method, giving emphasis to factors related to adverse events and the immunoresponse to the two techniques. Bibliographical searches were run on the MEDLINE and Lilacs databases using the keywords, "BCG vaccine" (administration, efficacy, adverse events) and tuberculosis (control and prevention). Articles published in English, Portuguese and Spanish during the last 15 years (1987 to 2002) were included.

Criteria on which to base the choice of BCG administration method

Globally, the intradermal method is used most widely. This is also the only method recognized by the WHO and the Brazilian Health Ministry.[3] In some countries, however, the percutaneous method is preferred because of concern about adverse events associated with BCG-ID and because the technique is easier to perform.[6,9-16]

The choice of vaccination method should take into account both the clinical effectiveness and safety of the respective vaccines. The best way to compare the effectiveness of a particular method is through clinical trials, preferably prospective studies that are double-blind controlled and involve large numbers of individuals.[7]

The choice of BCG administration method has traditionally been made based on an analysis of the reactogenicity profile of the vaccines and on their immunological capacity to induce a immunologic response of cutaneous hypersensitivity. There are no comparative studies of the effectiveness of different BCG administration techniques.[6]

In the Discussion that follows results will be presented from studies that have analyzed adverse events associated with the different BCG administration techniques, from tests of BCG immunoresponse both *in vivo* and *in vitro*, and, finally, from studies that have compared the ID and PC techniques.

Adverse events associated with the BCG vaccine

The intradermal BCG vaccine causes a local reaction which evolves over a long period (10 weeks). While the scar is still healing there may be oozing at the point of vaccination, right-side axillary lymphadenopathy occurs in between 1% and 10% of cases and suppuration complications in 0.1%. More than 95% of those vaccinated with BCG-ID exhibited a scar (4 to 7 mm in diameter).[3,5,6,17]

Adverse events associated with the BCG vaccine are rare, but may include local or systemic complications. Ulcers larger than one cm, subcutaneous abscesses, localized suppurating lymphadenitis occur in 0.4 out of every 1,000 vaccinations, appearing during the first six months post-vaccination. Hypertrophic or keloid scarring occur in 4 out of every million vaccinations. Systemic complications and fatal dissemination are rare (< 1.5 per million).[3,5,6,17]

Local and systemic reactions vary according to the bacterial strain employed and are more common among neonates than among adolescents. Local adverse events are associated with technical difficulties such as problems with subcutaneous placement and vaccination technique.[6,10,18,19]

The Moureau-Rio de Janeiro strain that is employed in Brazil induces a good delayed type hypersensitivity (DTH) skin test response and rarely causes local or systemic adverse reactions.[3,18,20-26] A study of 132 Brazilian neonates recorded just 12 local reactions (0.9%), four (0.3%) of which were suppurating adenites, six (0.45%) prolonged ulcers, and two (0.15%) lupoid reactions.[18]

In South Africa, the rate of occurrence of local reactions after BCG-ID use was 3% of 9,763 newborn babies. Adverse events occurred with much lower frequency at tertiary health centers than at centers located in peripheral regions, suggesting that these events were mainly related to technical problems with vaccine administration.[19]

More than 95% of those who receive the BCG-ID vaccination exhibit scarring. When the vaccine is administered more than 8 hours after preparation, DTH skin test response is reduced and scars are formed over a smaller area than when the vaccine is used less than eight hours after preparation.[18]

In Brazil, the majority of children vaccinated with BCG-ID exhibit scarring,[20-26] and it is recommended that those who do not exhibit any local reaction within six months be re-vaccinated.[3,17] While this method is highly useful, in practice "scar reading" is not always reliable.

The size of the scar caused by BCG, and also the DTH skin test response, are reduced with neonates than with school-age children and adults and the size of the scar also reduces over time.[18,24]

The sensitivity of scar reading at three months post BCG-ID administration is elevated for newborn children (93%) and reproducible in 94% of cases. When scar reading is performed 4 years post vaccination, sensitivity is greatly reduced (80%).[27] In Brazil, the correlation between scar reading and a record of BCG-ID was 80%.[24]

Vaccines using dead bacilli may leave scars. However, scar reading is not a good method for assessing vaccine coverage or the quality of the vaccine administered.[4,7,27,28]

The United States, Japan and some European countries have adopted the BCG-PC technique because of the ease of administration and the lower number of adverse reactions even though adverse events are very rare in association with BCG-ID.[4,9,10,16,29,30]

In England, 95% of babies vaccinated with BCG-ID exhibited scarring in contrast with 63% of those vaccinated with BCG-PC. The intradermal technique was considered difficult by 35% of doctors and, since DTH skin test response results were similar for both groups (68% and 73%, respectively), they concluded that BCG-PC had a greater acceptance for large-scale use.[11] Furthermore, the number of abscesses was 10 times greater after BCG-ID than after BCG-PC (2.8% and 0.29%).[13,14]

In Japan, the percutaneous method is preferred due to the elevated incidence of keloid reactions after BCG-ID. In a study of 34,516 children vaccinated with BCG-PC, 0.4% presented ganglions larger than 1 cm and 0.02%

suppurating adenitis. In the majority of cases adenitis had onset 4 to 6 weeks after vaccination and spontaneously withdrew within two months.[6]

Despite adverse events being less frequently associated with BCG-PC than with BCG-ID, it is important to point out that supperative complications are also rare after BCG-ID[6] and that even BCG-PC use can result in complications. There is evidence that complications are dependent on the strain and dosage employed.[6,30]

BCG vaccine effectiveness

The effectiveness of the BCG vaccine varies between 0 and 80%. Protection is greater against severe forms and outcomes such as tuberculous meningitis, miliary tuberculosis and death (70%) than against pulmonary manifestations (50 to 55%).[1,2,30-33]

Protection is generally lower in regions where disease incidence coefficients are extremely high. Nevertheless, even in areas of extreme poverty and high levels of malnutrition, the BCG vaccine, administered during the first few months of life, offers protection against tuberculous meningitis.[34-36]

There are no comparative studies of the effectiveness of BCG administration methods (BCG-ID and BCG-PC).[6,19]

In the USA and United Kingdom, where the BCG-PC vaccine has been adopted, there are low incidences of tuberculosis and the disease is not considered a significant public health problem.[13,14,16,29-33] In Japan, despite the BCG-PC vaccine being widely used, the incidence of tuberculosis is even lower than in the USA and there is currently debate on the need to maintain the vaccination program.[15,34] The studies of BCG-PC published in these countries, therefore, do not provide any data on the effectiveness of the vaccine.

In South Africa, where BCG-PC was routinely used up until the end of the nineties, tuberculosis prevalence remained elevated and, recently, the transition was made to intradermal administration, with no significant increase in adverse events occurring.[19]

A number of reasons, further to the methodological differences between studies, have been suggested as being responsible for the variations between vaccine effectiveness results obtained; of these, the most often quoted are quality and conservation of the vaccine (different vaccine strains, cold chain, ultra-violet light exposure), vaccine administration method, genetic and nutritional variations between populations, latitude, exposure to other mycobacteria and differences in the local prevalence of other diseases such as AIDS.[1-3,30-42]

Until to the end of the seventies, comparative studies of the two techniques were based on *in vivo* immunoresponse evaluations (cutaneous delayed-type hypersensitivity response - DTH skin test response); more recently, data has been published from studies which have investigated the immunoresponse *in vitro*, by means of assaying number of the different cytokines produced by the Th1 and Th2 lymphocytes, which appear to have a closer correlation with protective immunization.[40,43,44]

Methods for evaluating the response to BCG

Tests performed in vivo

The BCG vaccine induces cellular immunity, which is much more difficult to evaluate than the humoral type.

At the end of the nineteen-thirties, it was found that both BCG-ID and BCG-PC were capable of inducing a positive DTH skin test response, which lasted for a minimum of three years. This being the case, it was concluded that the BCG-PC vaccine was as "effective" as the BCG-ID at stimulating immunity and that it had the additional advantages the ease of application and the fact that it did not leave scars.[7]

At the start of the seventies, however, debate began over the validity of tuberculin-based test results as accurate markers of protection against tuberculosis, based on the fact that DTH skin test response varies according to the strain used, the population vaccinated, the type of instrument used for vaccination and the administration technique; in the case of the percutaneous vaccine, the number of needles and punctures made by different apparatus also make a difference.[4,33]

When the results of studies performed with children are compared with the results of studies involving adolescents, great variation is observed in DTH skin test response, being always lower among young children[14] than among adolescents and adults.[12]

Six to nine weeks after vaccination with BCG-ID or BCG-PC, 16% of neonates vaccinated with BCG-PC and 97% of those vaccinated with BCG-ID exhibited positive DTH skin test response assays.[14] The occurrence of localized abscesses post vaccination was 10 times greater among the group that received the BCG-ID vaccine.[36,37]

In contrast with the newborn children, the majority of the adolescents vaccinated with both BCG-ID and BCG-PC returned positive DTH skin test response results, although, while BCG-ID left scars on 100% of those vaccinated, just 17% of those that received the BCG-PC vaccine exhibited some type of local reaction.[12]

The BCG vaccines that are prepared for percutaneous use contain 40 to 50 times more viable bacilli than do vaccines formulated for intradermal use. It is believed that the low rates of adverse events associated with BCG-PC vaccines are due to the number of bacilli administered.[30]

Both DTH skin test response assay results and adverse event rates are directly related to the number of viable bacilli effectively inoculated. In the United Kingdom, inadvertent application of a vaccine formulated for percutaneous use intradermally was responsible for an elevated rate of local abscess formation.[30]

The Moureau-Rio de Janeiro strain used in Brazil is highly immunogenic, both in school-age children and infants, although among infants, the vaccine induces less DTH skin test response stimulation.[3,17,21-25,37]

There are large variations between individuals in terms of DTH skin test response and, if tuberculin testing is repeated, some individuals exhibit reversal of previous results while others return the same results as at first testing.[21]

The reliability of cutaneous testing is not always good; in up to 5% of cases a second DTH skin test response made by the same observer will lead to a reversal of the previous classification.[45] Furthermore, over time, the response to tuberculin testing diminishes, although, if there is exposure to *M. tuberculosis* or other mycobacteria, DTH skin test response will normally increase again.[21,46]

While DTH skin test response has been much used in research to assess the response to BCG vaccination, this test is not considered an accurate marker for immunity. Studies performed on experimental animals and on humans demonstrated that DTH skin test response has no relationship with protection and that individuals who react strongly to PPD may be less well protected against tuberculosis compared to those who do not exhibit a positive DTH skin test response.[21,29,31,37] Furthermore the post vaccination response to tuberculin testing is highly variable, depending on the strength of the vaccine employed, the strain, the age at which vaccination took place and the time that has passed since vaccination.[18,21,28] Therefore, new methods are needed to evaluate the capacity of BCG vaccines to stimulate protective immunoresponses.[37,38,40-47]

Towards the end of the nineties, the first questions were raised about the benefits of the percutaneous technique; instigated by the fact that this technique is associated with reduced induction of both DTH skin test response and of specific lymphocyte *M. tuberculosis* response, as assessed by IF-γ and IL-2 assay, nowadays accepted as better markers of protection.[6,40-44,46]

Tests performed in vitro

A number of different cytokines are produced by T-CD4+ lymphocytes and can be assayed *in vitro* in order to analyze the response to mycobacteria antigens. Th1 lymphocytes primarily produce interferon-gamma (IFγ) and Interleukin-2 (IL-2), while the Th2 lymphocytes produce IL-4, IL-5, IL-6 and IL-10. In experimental animals, the production of IF-γ and IL-2 has been associated with resistance to a number of different intracellular parasites and IF-γ increases the capacity of macrophages to destroy *M. tuberculosis.*[47]

The precise protective mechanisms acting against tuberculosis are not yet entirely understood, however, there is evidence that the best marker for evaluating the effectiveness of vaccines against tuberculosis is IF-γ production; for the following reasons:

- IF-γ is essential to tuberculosis protection in experimental animals;
- patients with advanced or moderate forms of tuberculosis produce less IF-γ;
- the use of IF-γ can be of benefit to people suffering from multiresistant tuberculosis;
- healthy people living with tuberculosis patients are found to have elevated IF-γ levels;
- after a positive DTH skin test response tuberculin test IF-γ levels rise;
- in pleural tuberculosis, which exhibits a tendency towards spontaneous resolution, IF-γ levels become elevated within the cells of pleural fluid and the blood.[47]

While these factors suggest that IF-γ ought to be the best marker for tuberculosis immunity, there are large variations in the production of this cytokine among both healthy and sick people. Levels of IF-γ may be elevated in healthy adults who have been in contact with sick people, in adults with tuberculosis with minimal lesions and in children with progressive tuberculosis. Adults with moderate or severe stages of the disease exhibit low IF-γ product ion levels, but it is not yet known if this lowered IF-γ production is the cause or the consequence of the progression of the disease.[47,48]

It is believed that tuberculosis protection is more closely linked to IF-γ titers and to the titers of cytokines produced by Th2 lymphocytes than to the absolute titers of each cytokine.[49]

The response to BCG varies according to previous mycobacteria sensitization. Tests performed *in vitro* have demonstrated that, among adults with a negative reaction to tuberculin testing, a response to soluble *M. tuberculosis* antigens appears a week after BCG administration; the response to antigens form the cell wall membrane only occurs 4 weeks post vaccination, coinciding with Mantoux test conversion and a response to antigens prepared by the cytolysis of *M. tuberculosis* appear a year after vaccination. Adults who have been previously sensitized, exhibit a wide spectrum of reponses to a the various *M. tuberculosis* antigens.[49] These results, obtained *in vitro*, confirm the classic clinical findings that responses to BCG are accelerated when people who have been previously sensitized are vaccinated.[22,23,26,49]

The BCG vaccine induces a Th1 and Th2 response, but, the action of cytotoxic lymphocytes is modulated by the cytokines that are produced by the Th1 lymphocytes. The results of recent studies, performed with both humans and animals, suggest that previous exposure to mycobacteria antigens is associated with increased cytotoxic activity and that, in order to be reliable, the BCG vaccine should be administered before the recipient has been exposed to *M. tuberculosis* or other mycobacteria.[39,43,49]

Among adults, BCG induces a strong Th1 response, however, studies performed with laboratory animals have indicated that early exposure to bacterial antigens may induce a response that predominantly involves Th2 lymphocytes, which are more associated with hypersensitivity that with protective immunity. This fact is of great concern when it is remembered that the BCG vaccine is recommended for newborn babies.[50]

There has yet been little study of the evaluation of newborn babies' immunoresponse to BCG. however it has already been demonstrated that, while BCG-ID stimulates cytotoxic lymphocytes, the response is modulated and there is high IF-g production and low IL 5 production.

In newborns, BCG-ID is capable of inducing specific cytotoxic lymphocyte generation, against mycobacteria and cytotoxic activity, as assessed by IL5 and IL10 production is greater among children who exhibit less IF gamma production.[49] Furthermore, children vaccinated at between 0 and 4 months with BCG-ID exhibit Th1 cell memory, maintaining high IF-g production and low IL 5 production.[50]

The relationship between DTH skin test response and IF-γ production

In countries such as Sweden and the United Kingdom, where there is little exposure to mycobacteria, there is a positive correlation between tuberculin testing conversion and IF-γ production.[29,38] This is no longer true when BCG vaccine responses are evaluated in individuals who live in communities that are heavily exposed to *M. tuberculosis* or other mycobacteria, such as in Africa (Malawi).

The magnitude of the immunoresponse, as measured by the increase in DTH skin test response and IF-γ production, before and after BCG vaccination was compared for English and African patients. Before vaccination, positive DTH skin test responses and IF-γ production were more common among the Africans (46% and 61%) than among the English (13% and 22%).

One year after vaccination, IF-g was similarly distributed across the two populations. The DTH skin test response increased for both populations, although more intensely so in the United Kingdom and the correlation between DTH skin test response and IF-γ production was only confirmed in the United Kingdom, suggesting that exposure to mycobacteria interferes significantly with the immunoresponse to the BCG vaccine.[38]

It is believed that the protection afforded by the BCG vaccine is better correlated with the magnitude of the post-vaccination immunoresponse than with absolute IF-γ or DTH skin test response values. It is probable that sensitization due to exposure to other mycobacteria will prove to be the most important factor controlling the variable protection afforded by the BCG vaccine among different populations.[38]

The BCG-ID vaccine has a greater capacity for stimulating cell-mediated immunoresponse than does the BCG-PC; 83% of adults vaccinated by BCG-ID present a positive DTH skin test response compared with 40% of those vaccinated by BCG-PC. Furthermore, it has been confirmed that the *M. tuberculosis* specific lymphoproliferative response only increases to a significant extent when vaccination is by BCG-ID.[40]

Delayed type hypersensitivity skin test responses and lymphoproliferative responses to the various proteins secreted by *M. tuberculosis* were evaluated for individuals who had been vaccinated at fpuir different BCG-PC dosages. Individuals vaccinated at low BCG doses did not exhibit IF-γ production and only 10% presented positive DTH skin test responses 8 weeks after vaccination, while 95% and 100% respectively of those vaccinated with standard and high doses of BCG presented elevated IF-γ and positive DTH skin test responses. These results prove that low doses of the BCG vaccine are ineffective at stimulating TH1 immunoresponse.[42]

With adults, the DTH skin test response is more intense after BCG-ID and correlates with other protective immunity markers such as IF-γ and specific lymphoproliferativeresponses.[46]

The response to the BCG vaccine varies in accordance with a number of different factors. In children, both DTH skin test responses and cytokine production in reaction to different strains of the BCG vaccine, administered sub or percutaneously, during the neonatal period or at ten weeks, were more intense for groups vaccinated with the Japanese strain, by BCG-ID and among children vaccinated at ten weeks.[49]

Despite some authors having suggested that vaccinating children after two months may improve the immunoresponse to the BCG vaccine,[49] it has already been demonstrated, in Brazil, that even low birth weight neonates exhibit a good immunoresponse after receiving BCG-ID.[26] We did not find any studies evaluating the Th-1 cellular response after BCG-PC in our country.

An analysis of comparative studies of BCG-ID and BCG-PC leads to the following conclusions:

- stimulation of a DTH skin test response and the production of cytokines associated with Th1 lymphocytes are more reliable with BCG-ID;[4,12,49]
- low dosages of the BCG vaccine are unreliable for inducing protective responses, as assessed by IF-γ production;[42]
- it is probable that the differences between the results found when comparing the two techniques are the result of the lower dosage inoculated percutaneously;[6-9,30-42]
- adverse events associated with BCG are most common in association with poor administration technique;[4,9,18]
- the use of scarring to test whether the vaccine has "taken" is only of use after BCG-ID since BCG-PC does not leave scars;[9,18,19]
- the presence of scars should be verified during infancy since the scar reduces in size over the years;[27,29]
- BCG-ID gives better protection against the more severe forms of the disease, which principally occur during childhood, such as meningitis tuberculosa;[2,3,32,33,35]
- to date, there is no evidence to prove that re-vaccination is effective for increasing the levels of protection against tuberculosis afforded to adolescents and adults.[28,33,41,47]

Recommendations

In Brazil, increased vaccination coverage with BCG-ID has been clearly associated with reduced tuberculous meningitis in infancy. While the BCG-PC vaccine containing the Moureau-Rio de Janeiro strain has been licensed in Brazil, it has been little used and its effectiveness cannot therefore be confirmed.

Advances in the fields of immunology and molecular biology have stimulated research into new vaccination techniques for tuberculosis, however it is unlikely that BCG-ID will be substituted by a new vaccine in the next few years.[51-54] Whilst more reliable vaccines are not available, the intradermal method should be preferred to the percutaneous method as it is more reliable in inducing a protective immunoresponse.

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