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The Immunological Basis for Immunization Series

Module 3:

Tetanus

GLOBAL PROGRAMME FOR VACCINES AND IMMUNIZATION
EXPANDED PROGRAMME ON IMMUNIZATION

World Health Organization
Geneva

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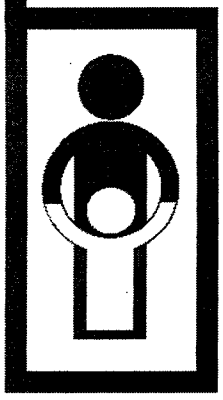
Dr Artur M. Galazka
Medical Officer
Expanded Programme on Immunization



GLOBAL PROGRAMME FOR VACCINES AND IMMUNIZATION
EXPANDED PROGRAMME ON IMMUNIZATION

World Health Organization
Geneva

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The Immunological Basis for Immunization **series is available in English and French (from the address below). It has also been translated by national health authorities into a number of other languages for local use: Chinese, Italian, Persian, Russian, Turkish, Ukranian and Vietnamese. The series comprises eight independent modules:**

Module 1: General Immunology
Module 2: Diphtheria
Module 3: Tetanus
Module 4: Pertussis
Module 5: Tuberculosis
Module 6: Poliomyelitis
Module 7: Measles
Module 8: Yellow fever

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Expanded Programme on Immunization
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Contents

Preface

This series of modules on the immunological basis for immunization has grown out of the experience of persons working with the WHO Expanded Programme on Immunization (EPI). The EPI was established in 1974 with the objective of expanding immunization services beyond smallpox, with emphasis on providing these services for children in developing countries.

Six vaccine-preventable diseases have been included within the EPI since its beginning: diphtheria, measles, pertussis, polio, tetanus, and tuberculosis. To protect newborns against neonatal tetanus, tetanus toxoid is administered to the mother either during her pregnancy or prior to pregnancy during the childbearing years.

Two more vaccine preventable-diseases will be addressed by the EPI during the 1990s. The World Health Assembly has set the target of including yellow fever vaccine in the EPI by 1993 in countries where this disease poses a risk. Hepatitis B vaccine is being added gradually, with the target date of 1997 for incorporation of this vaccine in the immunization programme in all countries.

Titles of the nine modules in this series are listed inside the front cover of this module. They are intended to provide information on the immunological basis for WHO-recommended immunization schedules and policies. They have been prepared for the following main audiences:

- immunization programme managers, whose questions and concerns caused this series to be written,
- consultants and advisers on immunization activities,
- teachers of courses on immunization at the university level and facilitators of workshops,
- medical and nursing students as part of the basic curriculum,
- laboratory scientists providing diagnostic or research services for vaccine-preventable diseases, and
- scientists involved in basic research aimed at improving the delivery of vaccines or providing improved vaccines.

Other modules in this series and additional materials on the EPI are available from the Expanded Programme on Immunization, World Health Organization, 1211 Geneva 27, Switzerland.

Tetanus

1. Tetanus Toxin

Tetanus is caused by the action of a highly potent neurotoxin, tetanospasmin, which is produced during the growth of the anaerobic bacterium *Clostridium tetani. Cl. tetani* is not an invasive organism; infection with *Cl. tetani* remains localized. The disease usually occurs through infection of a skin injury with tetanus spores. Tetanus spores introduced into an area of injury convert to tetanus bacilli in the presence of necrotic tissue with reduced oxygen potential. Neonatal tetanus occurs through infection of the umbilicus when the cord is cut with an unclean instrument or when substances heavily contaminated with tetanus spores are applied to the umbilical stump.

Tetanus toxin produced by *Cl. tetani* bacteria migrates to its site of action in the central nervous system by retrograde transport along nerves. Tetanus toxin is neurotropic: it binds to gangloside-containing receptors at the nerve termini. Once bound to neuronal tissue, tetanus toxin cannot be affected by tetanus antitoxin. Toxin accumulates in the central nervous system, where it blocks the release of inhibitory neurotransmitter substances, such as glycine and gamma-aminobutyric acid, in neural synapses.

Tetanus toxin is very toxic; the estimated human lethal dose is less than 2.5 ng per kg. Tetanus toxin is synthesized inside *Cl. tetani* bacterial cells as a single polypeptide chain of 150 000 molecular weight. In artificial laboratory conditions, the toxin obtained from the culture supernatant consists of light and heavy polypeptide chains connected by a disulphide bond. The toxin molecule can be degraded into polypeptide fragments by enzymes such as papain. Some of the polypeptide fragments are non-toxic and have been studied as potential candidates for vaccines.

2. Tetanus Toxoid and the Nature of Immunity Against Tetanus

2.1 Tetanus toxoid-induced immunity

Tetanus toxin can be inactivated by formaldehyde to yield tetanus toxoid. Tetanus toxoid is used as a monovalent vaccine to immunize adults or as a component of combined diphtheria-pertussis-tetanus (DPT) vaccine or diptheria-tetanus (DT) vaccine for immunization of children. A combined tetanus-diphtheria (Td) vaccine for adults contains the normal amount of tetanus toxoid and a reduced amount of diphtheria toxoid. Tetanus toxoid is adsorbed onto aluminium salts (aluminium hydroxide or aluminum phosphate) to increase its antigenicity. Tetanus toxoid is stable and can withstand exposure to room temperature for months and to 37°C for a few weeks without a significant loss of potency *(Galazka 1989).*

Work is in progress on the development of a slow-release tetanus toxoid that may provide long-lasting immunity with only one injection. This research involves incorporation of tetanus toxoid into injectable and biodegradable microspheres made of well-tolerated polymers. Following injection of the slow-release product, the tetanus toxoid would be released from the injection site at pre-determined intervals.

Tetanus toxoid induces the formation of specific antitoxins. These antibodies play an important role in protection against tetanus. Immunity to tetanus is antibody-mediated and depends on the ability of antitoxins to neutralize tetanus toxin. Tetanus antitoxins, like diphtheria antitoxins, belong to the IgG class; they easily pass through the placenta and are distributed throughout the bloodstream and extravascular spaces. Antitoxin in tissues can neutralize toxin produced in an infected wound. Antitoxin which passes to the fetus through the placenta following active immunization of the mother can prevent neonatal tetanus.

Immunity to tetanus toxin is induced only by immunization; recovery from clinical tetanus does not result in protection against further attacks. A small amount of tetanus toxin, although enough to cause the disease, is insufficient to stimulate antibody production. Therefore, all patients with clinical tetanus should be immunized with tetanus toxoid, either at the time of diagnosis or during convalescence.

2.2 Controversies around "natural immunity" against tetanus

It has been proposed that "natural immunity" against tetanus can be induced by a sublethal dose of tetanus toxin or by fragments of tetanus toxin released from tetanus bacilli located in the digestive

tract, as a result of ingesting tetanus spores *(Dastur 1981, Matzkin & Regev 1985, Tenbroeck &* Bauer *1923, Veronesi et al. 1975, 1981).* Some authors report finding tetanus antitoxin in the sera of persons who were not immunized *(Tenbroeck 1923)* or who claim not to have been immunized with tetanus toxoid *(Dastur et al. 1981, Matzkin & Regev 1985, Veronesi et al. 1981, 1983).* Tetanus antibodies have been also found in the sera of unimmunized animals *(Veronesi et al.* 1983). Tetanus toxin can be adsorbed from the gastrointestinal tract. The rate of adsorption depends on the concentration of toxin, the species, and the age and condition of the mucosal lining *(Fedinec 1981).* Guinea pigs fed a suspension of tetanus spores or tetanus toxin for more than nine months have shown a rise in tetanus antibody level up to 0.1 IU/ml *(Veronesi et al. 1975).*

There are many unanswered questions about "natural immunity" against tetanus and much of the evidence is open to criticism. Tetanus organisms are widely distributed in nature. Proponents of the natural immunity hypothesis believe that "natural immunity" to tetanus occurs in developing countries due to the presence of tetanus bacilli in the intestinal tract. However,. early studies showed that tetanus bacilli carriers were as likely to be found in England and the USA as in China. Furthermore, the carrier state (transient or established) does not protect animals from tetanus infection, nor does it cause the appearance of detectable quantities of antitoxin in the serum *(Coleman 1931).*

Colonization of the intestine with tetanus spores is the basis for the theory of "natural immunity". Experiments in mice suggest that *Cl. tetani* bacteria colonize the intestinal tract poorly *(Ebisawa 1987).* Horses often carry tetanus organisms in their intestines, but they are extremely susceptible to tetanus toxin originating from injury. Little is known about factors influencing the production of tetanus toxin in the intestine, the nature of the antigen (whole toxin molecule, toxin detoxified in the stomach, or toxin fragments), the functional state of the intestinal barrier to tetanus toxin, or its adsorption from the intestine.

Lack of history of artificial immunization is not proof of its absence. In New Guinea in 1962 to 1964, women were asked about previous immunization prior to entry into a tetanus toxoid study and only those with a negative history were accepted. A few of the participants showed preinjection antitoxin, but antenatal clinic records showed that all had received tetanus toxoid in the previous two to three years. The immunity in these women living in an environment heavily contaminated with tetanus spores and associated with a high neonatal tetanus morbidity was, in this particular study, due to immunization *(MacLennan et al. 1981).*

Some authors claiming the existence of "natural immunity" to tetanus have used *in vitro* techniques to test for tetanus antibodies, such as passive hemagglutination *(Dastur et al. 1981, Ray et al.* 1978) or ELISA *(Matzkin & Regev 1985). By* these methods, the titers were very low, in the range of one-thousandth to one-hundredth part of a unit per ml, and may reflect the activity of antibodies other than antitoxin. Such very low titers do not necessarily provide evidence for immunity against tetanus (see section 3). Studies in African schoolchildren *(Rey 1981),* Indian military recruits *(Menon et al. 1976),* persons taking care of horses *(Lahiri 1939),* pregnant women in New Guinea *(MacLennan et al. 1975),* and healthy persons in Upper Volta *(Breman et al. 1985)* have demonstrated that populations in developing countries with a high level of exposure to tetanus spores usually lack tetanus neutralizing antitoxins.

If natural immunity is of epidemiological significance in developing countries, then the percentage of immune persons should increase with age. This is not the case *(Misra & Rao 1988, Ray et al. 1978).* The data for healthy unimmunized persons do not show a clear dependency between age and the presence of tetanus antibodies *(Metzkin & Regev 1985).*

The implications of "natural immunity" are understood quite differently. One group believes that naturally immunized people are sensitized by contact with tetanus toxin and will respond as primed persons when tetanus toxoid is administered parenterally *(Veronesi 1981).* Another group speculates about a tolerant state to tetanus toxoid resulting from chronic clostridial contamination of the small intestine *(Dastur 1981).* Neither speculation is confirmed by experimental data; in most studies in developing countries the response to primary immunization has not been significantly different from that in industralized countries.

Even if asymptomatic colonization and infection of the intestine with tetanus organisms occurs in some areas of the developing world, it is unlikely that natural immunity has any practical importance in controlling tetanus.

2.3 "Transplacental immunization" hypothesis

IgG antibodies produced by the immunized mother are transferred across the placenta to the fetus and provide transient, passive protection of the newborn against tetanus. Some authors suggest "transplacental immunization" as a different mechanism of neonatal protection. According to this concept, the fetus is actively immunized with tetanus toxoid transported transplacentally *(Gill et al. 1983, 1985, Vanderbeeken et al. 1985).* The main argument quoted

is the presence of tetanus IgM antibodies at birth in infants of mothers who were reimmunized with tetanus toxoid in pregnancy. It seems unlikely that non-soluble, adsorbed tetanus toxoid injected into the mother can penetrate from the mother's bloodstream to the placenta, cross the placenta, and pass into the fetal circulation to reach the lymphoid tissue of the fetus. In these studies IgM antibodies were measured by radioimmunoassay and it is not certain whether they represent tetanus-specific neutralizing antibodies (see section 3). Available data do not provide evidence that transfer of tetanus toxoid across the placenta results in active immunization of the fetus.

3. Techniques to Measure Antibody Response

Tetanus toxin does not produce a dermonecrotic effect on human skin or a cytopathogenic effect on tissue culture. Therefore, assays based on these effects — so useful in diphtheria — cannot be used for measuring tetanus antibody activities. Tetanus antibodies may be measured by *in vivo* or *in vitro* techniques.

3.1 Neutralization test *in vivo*

The *in vivo* neutralization test directly measures the biological activity of tetanus antitoxin by demonstrating the toxin-neutralizing property of serum in laboratory animals, usually mice. The neutralization test is expensive, time consuming, requires well trained personnel, a large number of animals, and a relatively large amount of serum. The neutralization test is a sensitive test which can detect an antitoxin level of one-thousandth IU/ml. This test is primarily a measure of serum IgG antitoxin.

Methods for titration of tetanus antitoxin can be divided into three groups *(Peel 1980).* Methods in the first group are based on the assumption that there is a close relationship between the average time of death of mice and the amount of non-neutralized toxin still present in the serum-toxin mixture. The exact time of death of the mice is measured over a five day period following injection of mixtures of different dilutions of serum and a test dose of toxin. A system of log values (correction factors) is used for each death time to calculate the extrapolated antitoxin titer *(Galazka et al. 1971, Ipsen 1942, Kyselova et al. 1968).* This method requires few materials, but requires frequent observations over a five-day period.

In the second group of methods, end-point readings are based on the proportions of mice dying and surviving at the end of a defined period after injection of the serum-toxin mixture, usually four days *(Barile et al. 1970, Eckmann 1963, Glenny & Stevens 1938, Gottlieb et al. 1964, Wilkins & Tasman 1959).* The major disadvantage of this method is that a relatively large volume of serum (1 to 2 ml) is required for titration when the serum has a low antitoxin concentration.

The third group of methods uses as an end-point the differentiation between paralysis of mouse leg injected with the serum-toxin mixture and complete neutralization of symptoms of tetanus. As differentiation between a minimal degree of paralysis and complete neutralization is influenced significantly by observer variation, the detection of this type of end-point tends to be difficult and innacurate. This method requires only a small amount of serum, but it can only detect titers of 0.02 IU/ml or higher *(Chen et al. 1956, Taylor & Moloney 1960).*

The accuracy and sensitivity of the neutralization test are influenced by the nature of toxin used (crude or purified), the toxin test (L+) level, and the weight of the mice *(Gupta et al. 1985, Peel 1980).*

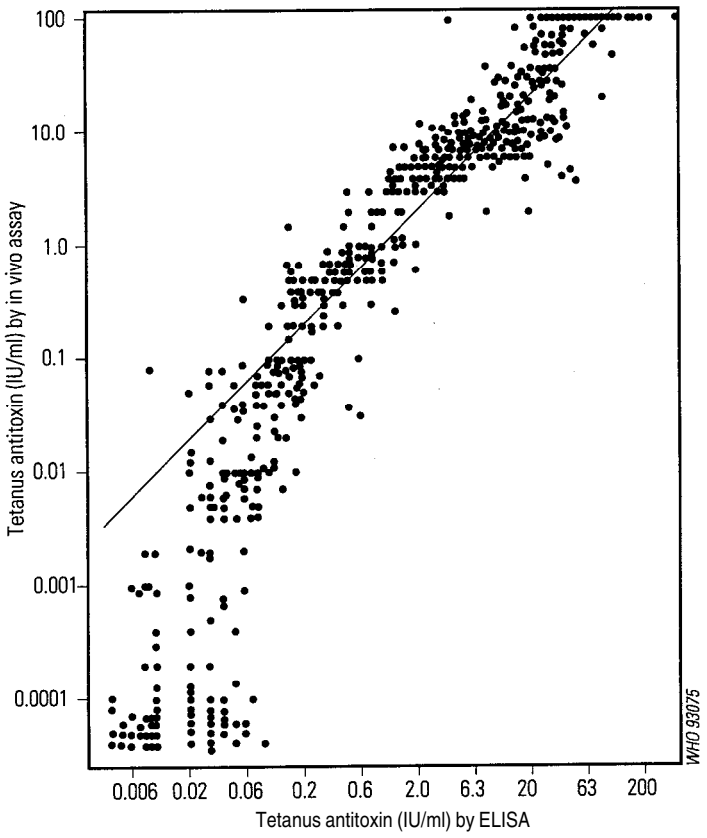
3.2 *In vitro* techniques

The interaction between tetanus antibody and tetanus toxin (or toxoid) may be measured *in vitro* by the passive hemagglutination test (HA), the enzyme-linked immunosorbent assay (ELISA), or the radioimmunoassay (RIA). These techniques are simple, sensitive, rapid, and inexpensive, but they are generally less specific than the *in vivo* neutralization method. *In vitro* techniques are more sensitive in detecting IgM antibodies than IgG antibodies, particularly in the early period of the primary response. Therefore, the results of *in vitro* techniques should be interpreted carefully and verified against the *in vivo* neutralization method.

3.2.1 Passive hemagglutination

The design of the passive hemagglutination (HA) test is simple: carrier red cells sensitized with tetanus toxoid agglutinate in a specific way in the presence of tetanus antibodies. The HA test has been widely used to assess the immune status of various age, sex, socioeconomic, and professional groups *(Aguzzi et al. 1980, Bistoni et al. 1978, Chapman & Davis 1973, Crossley et al. 1979, Fara et al. 1980, Galazka & Kardymowicz 1989, Gold et al. 1973, Kishimoto et al. 1980),* to evaluate the duration of immunity post-immunization *(D'Arca et al. 1980, Riberio et al. 1980),* and to compare the effectiveness of different vaccines and immunization schedules *(Durand et al. 1978, Feeley et al. 1979, Gasparini et al. 1980, Nazari et al. 1976, Ruben et al. 1978, Thorley et al. 1975).* The HA test has also been used for screening serum or plasma donated for production of human antitetanus immunoglobulin *(Rubin et al. 1980)* and

Figure 1. Tetanus antitoxin levels measured in 727 sera by ELISA and by an *in vivo* assay *(Simonsen et al. 1986).*



for detecting tetanus antibody in the serum of injured persons *(Bytchenko 1975, Galazka et al. 1971).*

The HA test can be conducted in a moderately equipped laboratory by staff trained in basic laboratory skills. Results of an HA test can be determined within one hour *(Galazka et al. 1971, Pitzurra et al. 1983).*

The main disadvantage of the HA test is its preferential sensitivity for IgM antibodies. IgM antibody is incapable of neutralizing tetanus toxin *(Ourth & MacDonald 1977),* but it successfully combines with tetanus toxoid resulting in a positive HA test. Although the correlation between the HA test and the neutralization test is generally high, differences of up to tenfold or more have been reported between the results of the two techniques in individual serum samples. These differences were especially noted at low levels of antibodies.

3.2.2 ELISA

In the indirect ELISA test, antibody in the test solution is allowed to react and form a complex with tetanus toxoid, which has been passively adsorbed to a plastic surface. An enzyme-labelled antibody against bound tetanus antibody (usually anti-IgG) is then attached to the antigen-antibody complex. The amount of enzyme bound, which indicates the amount of antibody in the test serum, can be measured by consumption of a suitable enzyme substrate. Usually, the substrate is chosen so that when it reacts with enzyme there is a color change which can be assessed visually or photometrically. The ELISA test is commonly used to assess tetanus antibody titers *(Lau 1987, Layton 1980, Sedgwick et* al. *1983, Sirnonsen et al. 1986b).* A simple modification of the ELISA test has been proposed for rapid assessment of immunity to tetanus in injured persons *(Chandler et al. 1984).*

In persons with an incomplete vaccination history, the ELISA test has been reported to be unreliable, as it often gives values far higher than the *in vivo* results *(Brabin et al. 1984, Gentili et al. 1985, Melville et al. 1983).* This is probably due to the presence of nonspecific or low avidity antibodies. The extent of these discrepancies is inversely proportionate to the neutralizing antibody content in the samples. The lowest ELISA value reliably predictive of protective antibody activity in serum, regardless of vaccination history, has been found to be 0.16 IU/ml (Figure 1).

Since use of the indirect ELISA test to quantify tetanus neutralizing antibodies in human sera is limited by marked overestimation in low titer sera, a modified ELISA test has been developed. The basis for this new test, called the antigen-competition ELISA, is the assumption that the binding of antitoxin to antigen adsorbed to the surface of microtiter wells is different from the binding of antitoxin to toxoid or toxin in solution *(Fey & Stifler-Rosenberg 1977, Simonsen et al. 1987b).* In the antigen-competition ELISA test, the antibody to be tested is allowed to react with antigen in solution, which prevents antibody binding to toxoid adsorbed to the insoluble carrier. The method is called the antigen-competition ELISA test since there is competition between free toxoid in solution and toxoid adsorbed to insoluble carrier. The results of the antigen-competition ELISA test correlate well with results of the *in vivo* neutralization test.

The toxin-binding inhibition (ToBI) test is another modification of the ELISA test. Tetanus toxin is preincubated with serum dilutions and the mixture is exposed to antitoxin-coated plates *(Hendriksen et al. 1988, 1989).* The ToBI test is based on detection of unbound toxin in a toxin-antitoxin mixture and therefore is similar to the neutralization test. The difference between the ToBI test and the neutralization test is the way in which free toxin is detected: in the ToBI test, toxin is detected by the enzyme-labelled antitoxin; while in the neutralization test, direct toxic effects are observed in mice.

Better correlation with *in vivo* tests has been obtained with the ToBI test than with the standard ELISA test. However, experience with the antigen-competition ELISA test and the ToBI test is limited and further data on the relationship between results of these tests and the neutralization test are needed, especially for sera with a low antibody content.

3.2.3 Other tests

Radioimmunoassay (RIA) tests have been used to titrate tetanus antibodies. There are several possible modifications of the RIA test: tetanus toxoid can be coupled with an insoluble sorbent, such as cellulose or agarose *(Stiffler-Rosenberg & Fey 1975),* or adsorbed passively onto a plastic surface as in the ELISA test. The specific antibodies bind to the antigen immunosorbent and are quantified by measuring the incorporation of isotope-labelled human antiglobulin attached to the antigen-antibody complex. The sensitivity of the RIA test is high and the results correlate well with values obtained by the HA test *(Wang et al.* 1982) and the ELISA test *(Layton 1980, Stiffler- Rosenberg* 1977). However, the reagents and equipment needed for the RIA test are expensive, the isotope labels may decay rapidly so that the conjugate has a short shelf life, the technique can be used only by highly trained personnel, and the radioactive material represents a small but potential health hazard.

Other methods, such as latex agglutination and various gel diffusion techniques, are simple and economical, but suffer from low sensitivity. They may be useful in detecting high antitoxin levels in donor blood used to produce human tetanus immunoglobulin.

4. Protective Level of Tetanus Antibodies

4.1 Protective level of antitoxin

The amount of circulating antitoxin needed to ensure complete immunity against tetanus is not known for certain. Establishment of a fixed level of tetanus antitoxin does not take into consideration variable conditions of production and adsorption of tetanus toxin in the anaerobic area of a wound or a necrotic umbilical stump. A given serum level could be overwhelmed by a sufficiently large dose of toxin. Therefore, there is no absolute protective level of antitoxin and protection results when there is sufficient toxin-neutralizing antibody in relation to the toxin load *(Passen et al. 1986).*

Immunological memory and the ability to respond quickly to booster doses of tetanus toxoid may be as important as the level of circulating antibody in determining the outcome of infection with tetanus spores.

Therefore, although the protective role of tetanus antitoxin is well documented, the establishment of a protective level has been somewhat arbitrary. Antitoxin activity is expressed in international units (III) and a tetanus antitoxin level of 0.01 IU/ml serum, is considered the minimum protective level. This "protective" level is based on animal studies that correlate antitoxin levels with symptoms or death. Experimental human data are limited and direct observations on "protective" levels of antibody are rare.

Wolters and Dehmel injected themselves with a dose of tetanus toxin equivalent to 2 or 3 human doses (calculated by weight, based on guinea pig experiments). Their postvaccination levels of serum antitoxin were 0.004 to 0.005 IU/ml and they did not suffer from tetanus after intramuscular administration of tetanus toxin (according to *Ullberg-Olson 1976).* The interpretation of this extraordinary experiment is limited by ignorance about what is a real "human dose" of tetanus toxin.

There are at least three reports of tetanus occurring in persons with antitoxin levels greater than 0.01 IU/ml. Goulon et al. *(1972)* studied the serum neutralization antibody level in 64 tetanus patients prior to serotherapy; in 54 patients (84%) the antitoxin level was lower than 0.01 IU/ml, but in 9 patients the antitoxin level ranged between 0.01 and 0.1 IU/ml and in one patient it was between 0.1 and 1 IU/ml. The severity of the disease was inversely proportional to the antitoxin level. The most severe illnesses and 5 deaths were in patients with antitoxin levels of 0.002 IU/ml or less.

Berger et al. *(1978)* reported one patient with an antitoxin level of 0.04 IU/ml at the onset of tetanus. Passen et al. *(1986)* described a case of severe, generalized tetanus in a person who had been fully immunized in childhood and who had received booster injections eight and four years before the disease. The antitoxin level was 0.16 IU/ml at the onset of the disease. The prognosis in this patient was considered poor because of the short incubation period, rapid progression from the initial symptoms to generalized spasms, and severe disease manifestation at admission. His survival and rapid recovery may have been the consequence of partial protection from pre-existing neutralizing antibody, good antibody response to toxoid doses given during the acute illness, and his young age and good general health.

4.2 Misuse of the term "protective" level of antibodies

The term "protective level" is often misused when it is assumed that the level of 0.01 IU/ml as determined by HA, ELISA, or RIA is equivalent to the same level of antitoxin determined by the neutralization method. This is not necessarily true, since *in vitro* techniques, including the HA test and the classical ELISA test, tend to show falsely increased titers in the range of "protective" titers (see section 3.2). The use of the term "protective level" is especially dangerous in studies on natural immunity or in assessment of individual titers of wounded persons. It is therefore advisable to use a level of antibody deter-

Table 1. The neonatal tetanus mortality rate and the clinical efficacy of two doses of tetanus toxoid (TT2) in preventing neonatal tetanus, as determined by community-based surveys in 6 countries.

Country	No. of live births surveyed	Neonatal tetanus mortality rate/1000 live births				Reference
		Overall rate	Rate for cases born of mothers		Efficacy of TT2 (%)	
			Immunized	Nonimmunized		
Burma	6 000	6.8	1.5	11.1	86	Stroh et al. 1986
Egypt	12 000	4.8	0.8	6.0	88	EPI 1987
Ethiopia	2 010	4.5	0	5.8	100	Maru et al. 1988
India	4 344	3.5	1.3	6.1	79	Kumar et al. 1986
Indonesia	4 971	10.7	1.4	12.5	89	Arnold et al. 1986
Iran	2 655	6.0	0	9.2	100	Sadeghi-Hasanbadi 1987

mined by in *vitro* techniques that is equivalent to 0.01 IU/ml determined by the *in vivo* technique. Usually 0.1 IU/ml is considered as a safe estimation. In laboratories using the antigen competition ELISA test or the ToBI test the tetanus antibody level corresponding to 0.01 IU/ml of neutralizing antibody will vary according' to the technique used.

5. Effectiveness of Tetanus Toxoid

5.1. How effective is tetanus toxoid?

The effectiveness of tetanus toxoid has been convincingly demonstrated in many field trials and in hospital-based studies. A double-blind, controlled field trial in a rural area of Colombia conducted in the 1960s showed that adsorbed tetanus toxoid administered to women of childbearing age provides substantial immunity against neonatal tetanus. Control babies had a neonatal tetanus mortality rate of 78 per 1000 live births, whereas no neonatal tetanus cases occurred in babies of mothers given two or three doses of tetanus toxoid *(Newell et al. 1966, 1971).* A reduction in neonatal tetanus mortality following the implementation of programmes to immunize women of childbearing age, and especially of pregnant women, has also been observed in Bangladesh *(Black et al. 1980, Rahman et al. 1982),* Haiti *(Berggren et al. 1983),* Mozambique *(Cliff 1985, Expanded Programme on Immunization 1988),* and Sri Lanka *(Expanded Programme on Immunization 1982).* Surveys of neonatal tetanus mortality provide data about mortality rates for children born to vaccinated and nonvaccinated mothers; these data are useful in assessing the tetanus toxoid vaccine efficacy. In most studies, tetanus toxoid vaccine efficacy ranged from 80% to 100% (Table 1).

5.2 Reported failures of tetanus toxoid immunization

Although immunization with tetanus toxoid is one of the most effective prophylactic procedures, several apparently genuine failures to achieve protection following tetanus toxoid immunization have been reported (Table 2). As Edsall mentions in his excellent review *(1959),* some of these reports are inadequately documented, some cases follow incomplete immunization, several occurred years after primary or basic immunization, and only a few had received basic immunization plus a booster injection.

The number of reports from Asia and Africa describing the failure of tetanus toxoid to prevent neonatal tetanus in infants of immunized women has increased recently. These data were obtained from hospital-based studies (Table 3). This problem becomes increasingly prominent as immunization coverage rises, since the proportion of neonatal tetanus cases in infants of immunized mothers will increase, even with high tetanus toxoid vaccine efficacy. In India, the number of neonatal tetanus cases hospitalized decreased from 88 in 1984 to 19 in 1989, but the proportion of mothers of neonatal tetanus cases immunized with at least 2 doses of tetanus toxoid increased from 20% in 1987 to 32% in 1989 *(Deivanayagam et al. 1991).* From 1984 to 1989, the reported coverage with tetanus toxoid immunization increased in India from 33% to 69%. Similar changes were observed in Senegal *(Diop et al. 1991).*

There are several explanations for reports of neonatal tetanus cases occurring in infants of women claiming to be immunized:

1. Inaccurate immunization history. Maternal immunization status is often based on verbal history, not written documentation. In many countries, written records are not given to mothers or they do not retain records they are given. In some countries pregnant women receive many

Table 2. Tetanus cases and deaths reported in persons immunized with tetanus toxoid, 1946 to 1992.

Reference	Tetanus toxoid immunization status	No. of cases	No. of deaths
Boyd 1946	Primary series or incomplete	7	3
	Routine booster(s)	9	2
	Emergency booster	1	0
Hall 1948	Uncertain	1	1
	Primary series	1	0
	Emergency booster	1	0
Hedrick 1951	Primary series	1	0
	Routine booster	1	0
Boyer et al. 1953	Primary series or incomplete	9	7
	One booster, 3 years previously	1	1
Long 1954	Primary series	2	0
	Routine booster, 3 months previously	1	0
	Emergency booster	4	2
Moss et al. 1955	Routine booster	2	1
	Emergency booster	1	0
Christensen & Thurber 1957	Primary series, 10 years previously	1	0
Peterson 1965	Emergency booster	1	0
Spittle 1973	Several boosters	1	0
Berger et al. 1978	Uncertain	1	0
	Primary series, 15 years previously	1	0
Baptist 1984	Incomplete primary series	1	1
Passen et al. 1986	Boosters 8 and 4 years previously	1	0

injections unrelated to tetanus toxoid immunization and this may lead to confusion about tetanus toxoid immunization status.

2. Inappropriate immunization schedule. Many women report late in pregnancy for antenatal care. Consequently, immunization is begun late and the second dose of tetanus toxoid is given too close to delivery for the mother to develop an immune response that will protect the newborn.
3. Low potency vaccine. The tetanus toxoid itself may not be potent due to problems of manufacture, storage, or transport. For at least one developing country, the epidemiologic and laboratory data indicated sub-standard potency of locally produced tetanus toxoid.
4. Poor maternal immune response. In most studies performed in developing countries, two doses of tetanus toxoid stimulated the development of tetanus antibody levels considered protective in at least 80% of women. Some mothers, however, may have an antibody response below the protective level ("poor responders" - see section 6.1).
5. Inadequate placental transfer. Recent studies suggest than in areas where mothers' immunoglobulin levels are excessively high due to continued antigenic stimulation, placental transfer of antibodies may occur at lower rates, leaving the newborn inadequately protected.
6. Excessive toxin exposure. The load of tetanus toxin produced in a heavily contaminated umbilical cord stump may be so large that the modest immunity generated from two doses of tetanus toxoid is overwhelmed.

5.3 Factors influencing the response to tetanus toxid

Two conditions that may influence the immune response to tetanus toxid are malaria and AIDS. In many areas where neonatal tetanus is widespread, malaria is also endemic.

The response of malaria-infected pregnant women to tetanus toxoid immunization is similar to that of non-pregnant healthy adults *(Brabin et al. 1984).* No differences in antibody response to tetanus toxoid was seen in persons who had received long-term malaria chemoprophylaxis, compared with control persons *(Gilles et al. 1983, Monjour et al. 1982).* In

Table 3. Tetanus toxoid (TT) immunization history of mothers whose infants developed neonatal tetanus (NT), based on hospital data.

Country	Reference	No. of NT studied	Maternal history: number of doses of TT			
			0	1	2	3
Angola	Grudeborn 1987	199	188	0	11[a]	0
Egypt	El-Sherbini 1991	74	55	19	0	0
Egypt	Gad et al. 1986	324	324	0	0	0
India	Bildhaiya 1983	74	73	0	0	1[b]
India	Deivanayagam et al. 1991	19	13	0	3[c]	3[c]
India	Ghosh 1990	30	21	5	4	0
India	Kumar et al. 1988	385	363	0	22[d]	0
India	Mathur et al. 1980	50	50	0	0	0
India	Verma et al. 1989	76	49	5	12[e]	10
Mozambique	Cliff 1985	175	173	0	2	0
Nigeria	Einterz & Bates 1991	237	234	"several"	1	0
Nigeria	Grange 1991	419	411	8	0	0
Nigeria	Owa & Makinde 1990	52	35	5	11	1
Nigeria	Oyedeyji et al. 1982	104	97	3	3	1

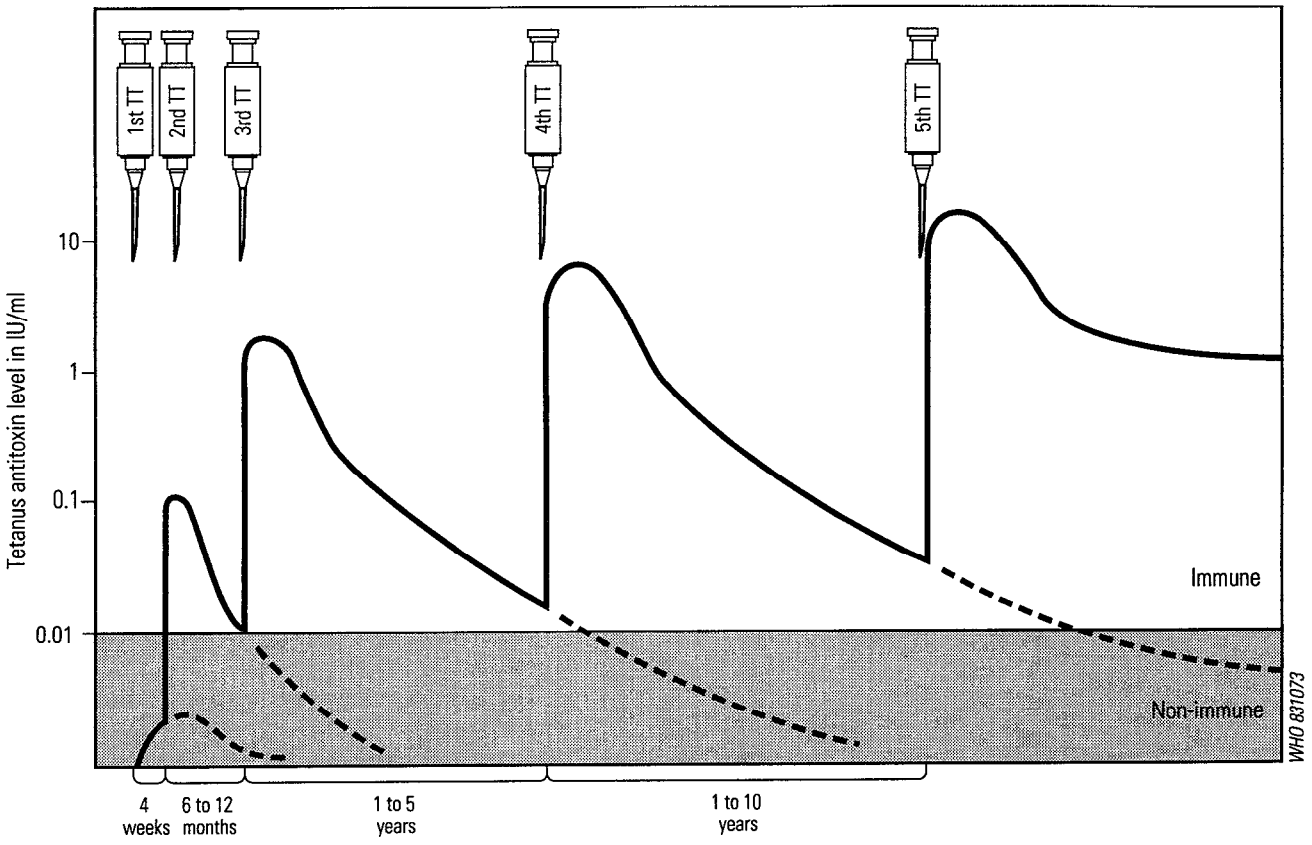
[a] *Immunized during pregnancy with TT.*
[b] *Immunized in childhood with DPT vaccine.*
[c] *Out of 3 mothers who received 2 doses of TT the second dose was given in the 9th month of pregnancy in two mothers; out of 3 mothers who received 3 doses, the third dose was given in the 9th month of pregnancy in one mother.*
[d] *22 mothers "fully" immunized.*
[e] *One mother received the second dose two days before delivery.*

other studies, however, a decreased response was observed following one or two doses of tetanus toxoid in children with parasitemia from an acute attack of malaria *(Edsall et al. 1975, Greenwood et al. 1972).*

The main concern in HIV-infected persons is the effectiveness of immunization. Persons with symptomatic HIV infection have several immunological abnormalities, including hypergammaglobulinemia, decreased CD4 lymphocytes, poor T-lymphocyte response to mitogen stimulation, and altered humoral immunity. In such persons, abnormal primary and secondary antibody responses may result in decreased efficacy of immunization. Abnormalities of the immune response become more severe with advancing disease *(Blanche et al. 1986).* HIV infection interferes with antibody responses to antigen encountered after infection has occurred, but affects less severely the antibody responses of lymphocytes "educated" prior to infection *(Borkowsky et al. 1987).* Defects in antibody response may be more evident in children, who have a more limited antibody repertoire before the effects of HIV on immune responsiveness become apparent *(Pinching 1991).* Antibody responses of children with AIDS to diphtheria and tetanus toxoids and to pneumococcal vaccine were absent or lower than those of age-matched controls *(Bernstein et al. 1985, Borkowsky et al. 1987, Blanche et al. 1986).* In contrast, HIV-infected parents of these children had protective diphtheria and tetanus antibodies resulting from childhood immunization *(Borkowsky et al. 1987).* A recent study suggests that most children infected with HIV during the perinatal period have a satisfactory immune response to tetanus and diphtheria toxoids during the first two years of life. However, from age 2 to 4 years tetanus and diphtheria antibody levels fell more rapidly in HIV-infected children *(Borkowsky et al. 1992).*

The tetanus response varies in HIV-infected adults. A decreased response to a booster dose of tetanus toxoid has been reported in asymptomatic *(Teeuwsen et al. 1987)* and symptomatic HIV-infected adults *(Opravil et al. 1991).* In another study, tetanus toxoid immunization of 21 HIV-infected military recruits produced antibody responses equal to those in uninfected controls. However; only 11 of the 21 individuals had a serological response to diphtheria toxoid compared with 18 of 21 controls *(Rhoads et al. 1987).* In studies in Africa and Haiti, HIV- infected women had the same levels of tetanus antibody after two doses of tetanus toxoid given during pregnancy

Figure 2. Antibody response to tetanus toxoid (TT).



as seronegative women *(Baende et al, 1989* — according to *Onorato & Markowitz 1992, Halsey et al. 1988).*

Tetanus toxoid, as a monovalent vaccine or as a component of combined vaccines, is recommended for HIV-infected children or adults, regardless of the presence or absence of symptoms of AIDS. A similar policy is recommended for all inactivated bacterial and viral vaccines.

6. Development of Immunity Following Immunization

6.1 Immune response to immunization

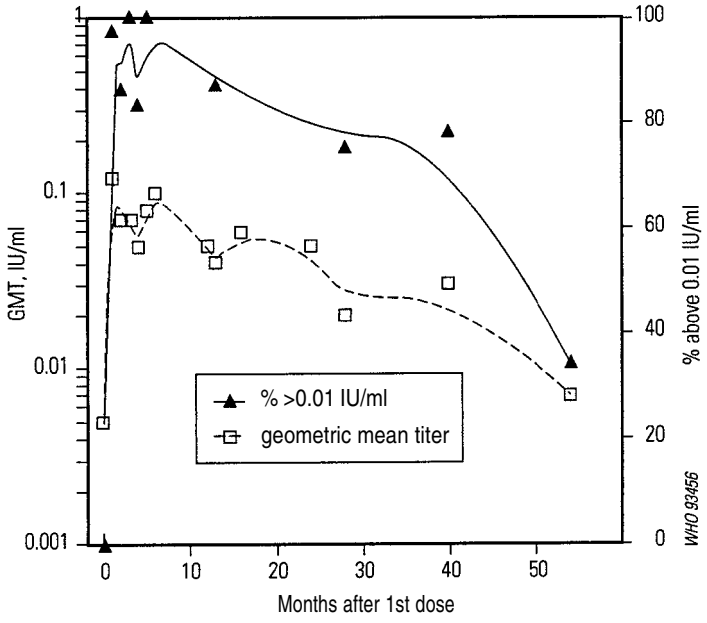
A schematic picture of tetanus antitoxin response of adults following primary and booster immunization with tetanus toxoid is shown in Figure 2. The degree and duration of immunity increases with the number of tetanus toxoid doses given. One dose of tetanus toxoid ensures little, if any, protection. Two to four weeks after the second dose the mean level of tetanus antitoxin usually exceeds the minimum "protective" level of 0.01 IU/ml, although the percentage of poorly protected persons ("bad responders") can still be up to 10%. Immunity declines with time. After one year the percentage of poorly protected persons may increase to 20% and the mean titer may fall to the threshold level. A study in Papua New Guinea showed that 78% of women immunized during pregnancy with two 10 Lf doses of adsorbed tetanus toxoid had antitoxin levels above 0.01 IU/m1 for at least 3 years; the mean antitoxin level was about 0.03 IU/ml (Figure 3).

The infants of poorly responding women may be at risk of tetanus. For this reason, a third dose of tetanus toxoid should be given during the subsequent pregnancy or 6 to 12 months after the initial two doses.

A third dose of tetanus toxoid induces plentiful antitoxin production, with mean levels between 1 and 10 IU/ml. The level of immunity induced by a course of three injections is high and durable. One month following the third dose the percentage of bad responders is negligible and the protective level lasts for at least 5 years.

After the third dose, each additional dose given with at least a one year interval increases the tetanus antitoxin level and prolongs the duration of immunity. Immunity will last for 10 years after the fourth

Figure 3. Geometric mean titer and the percentage of pregnant women with 0.01 IU/ml or more of tetanus antitoxin after two doses of adsorbed tetanus toxoid, Papua New Guinea *(MacLennan et a/. 1965, Hardegree et al. 1970).*



dose and for at least 20 years after the fifth dose.

In children, three primary doses of DPT vaccine induce an antibody level above the minimum protective threshold, with a mean level above 0.2 IU/ml *(Anderson et al. 1988, Barkin et al. 1984, Edwards et al. 1989, Kimura et al. 1991, Pichichero et al. 1986).* Factors influencing the height of the immune response in children and adults, apart from the number of doses, are discussed in section 7.

6.2 Duration of immunity following various immunization schedules

Immunization of infants with 3 doses of DPT vaccine will provide tetanus immunity for one to three years (Figure 4). Usually, three doses of tetanus toxoid received as an infant are counted as two doses received as an adult. Reinforcing the infant immunization with a fourth dose given somewhere between the 15th and 24th month of life will prolong tetanus immunity for another five years, e.g. until 6 or 7 years of age. A fifth dose of tetanus toxoid (given as Td or DT vaccine) at school entry will provide immunity for another 10 years, e.g. until 17 or 18 years of age. An additional dose at school leaving or during military service will assure sufficient immunity for at least two more decades *(Bizzini et al. 1978, Christenson & Bottiger 1987, Simonsen et al. 1986a).*

In Sweden, females under 20 years of age routinely receive 4 doses of DT vaccine. In contrast, Swedish males receive an additional booster dose of DT as military recruits at the age of 18 years and the effect of this additional dose is seen for several decades (Figure 5).

The EPI recommendation to give five doses of tetanus toxoid to women of childbearing age is based on the United States military experience with tetanus toxoid during World War II. The American record was particularly impressive, with only 12 cases of tetanus occurring in over 2.5 million injured soldiers in the US Army and Navy. The two services established basic immunity with 3 doses of fluid (non-adsorbed) toxoid (Army) or two doses of alum-precipitated toxoid (Navy), and a booster dose

Figure 4. Expected duration of tetanus immunity after different immunization schedules.

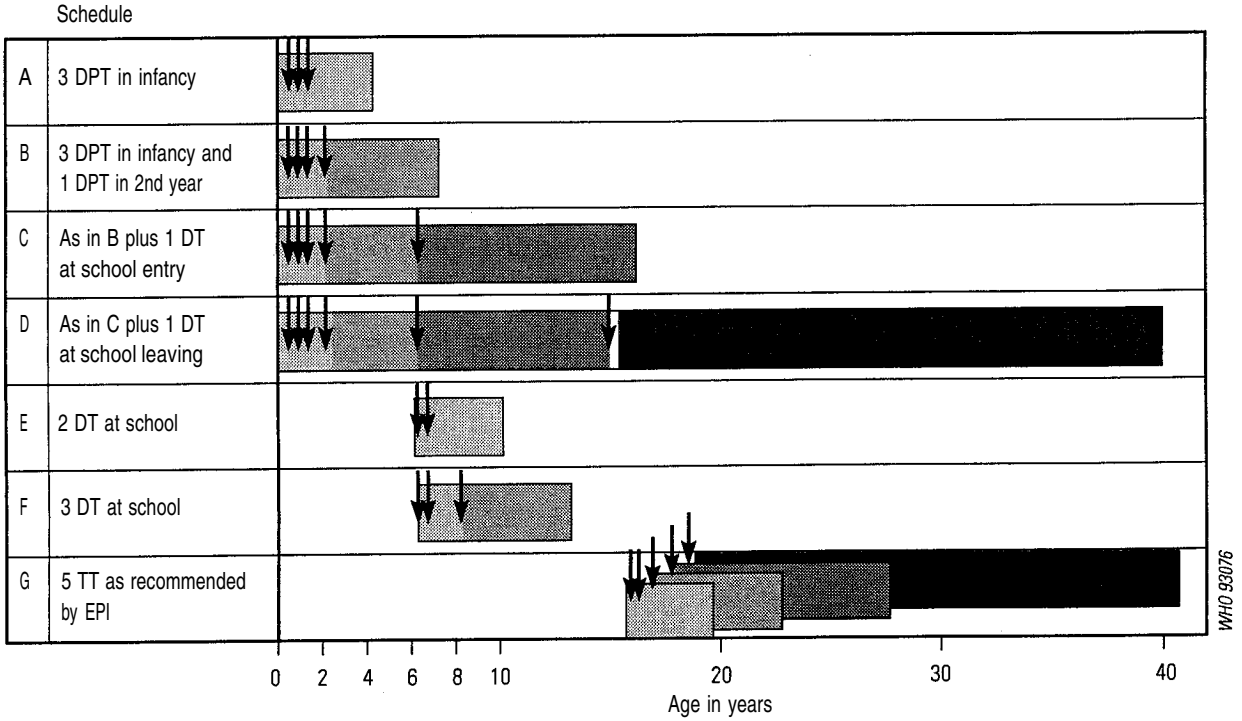
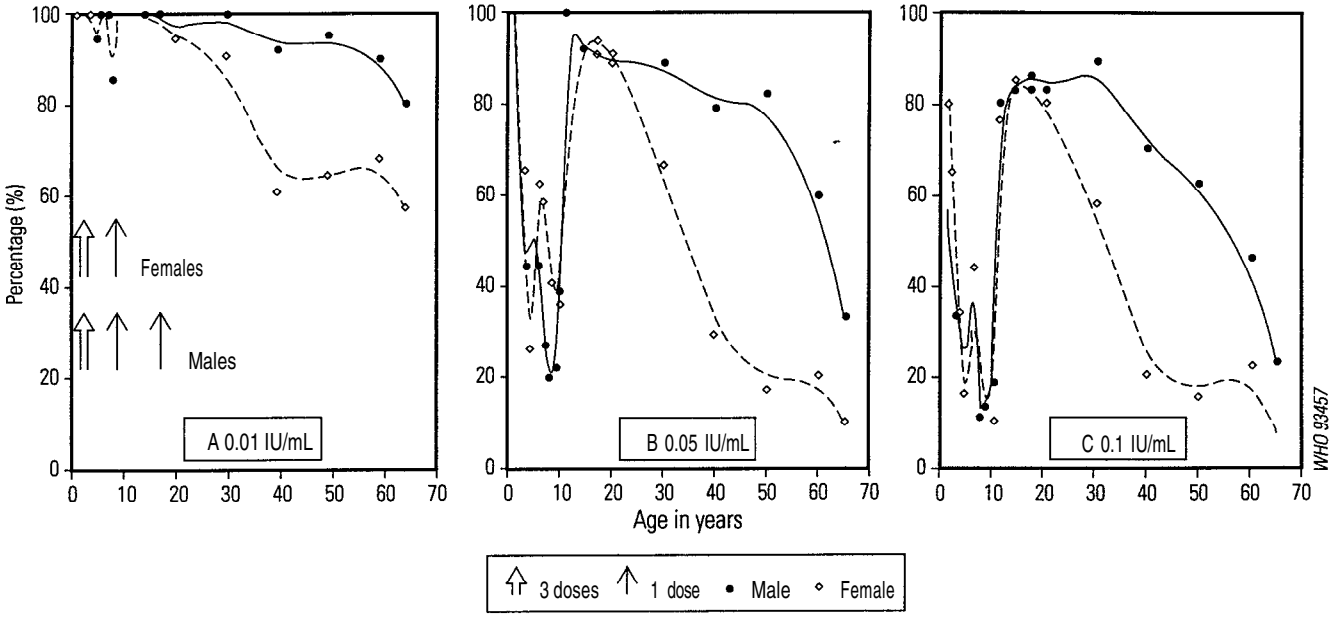


Figure 5. Tetanus immunity by age at three levels of antibodies, (ELISA), Sweden 1983 to 1985 *(Christenson & Bottinger 1987).*



after one year. Subsequent booster doses were given either before departure for combat (Army) or at four-year intervals (Navy). In addition, both services administered an emergency booster dose as soon as possible after wounds or other injuries of the type ordinarily incurring the risk of tetanus. In 1949, the US Army, Navy, and Air Force jointly adopted the procedure of administering two doses of adsorbed tetanus toxoid one month apart, a third dose approximately one year later, and booster doses every four years and after injury *(Looney et al. 1956).*

In the 1960s the duration of tetanus immunity in American military veterans was determined in several studies. Most of veterans surveyed retained their tetanus immunity and, furthermore, they were able to respond to a booster dose of tetanus toxoid (Table 4).

There are insufficient data on the duration of tetanus immunity following three doses of DPT vaccine given early in infancy. Early observations of Scheibel from Denmark (Table 4) suggested long-lasting immunity following two doses of DT vaccine

Table 4. Tetanus antibody levels in persons up to 30 years since last dose of primary series of tetanus toxoid and after a booster dose.

Country	Primary series	Subjects	Years since last dose	Antibody level before booster		Time after booster	Antibody level after booster		Reference
				GMT IU/ml	% >0.01 IU/ml		GMT IU/ml	% sera with titer	
USA	during military service	veterans	15		87.5	4 days 7 days 14 days		86 > 0.01 94 > 0.01 100 > 0.1	McCarroll et al. 1962
USA	during military service	veterans	14 to 18	0.08	87	7 days 14 days 21 days	1.2 12.0 15.7	92 > 0.1 96>1.0 100 > 0.1	Goldsmith et al. 1962
USA	during military service	veterans	14 to 21	0.11	100	14 days	about 30*	99 > 1.0	Gottlieb et al. 1964
Denmark	3 DT	children	4 to 8	0.38	98.7				Scheibel et al. 1962
Denmark	3 DT	children	10 to 14	0.35	95.8				Scheibel et al. 1966
Denmark	3 DT in childhood 3 DT in childhood	adults adults	14 to 18 26 to 30		89 72	20 years		100 > 0.01	Simonsen et al. 1986a
Denmark	4 DPT in childhood	military recruits	19	0.09					Simonsen et al. 1987a
Denmark	3 DT in childhood	adults	25	0.08		9 years	2.5		Simonsen et al. 1987a

* Arithmetic mean.

reinforced with a third dose. Further Danish studies showed that three doses of a combined DT-polio vaccine given at 5,6, and 15 months of age induced protective levels of tetanus antibodies lasting for 14 to 18 years in 90% of persons, 20 to 25 years in 85% of persons, and 26 to 30 years in 72% of persons *(Simonsen et al. 1987a, Simonsen 1989)*. Reimmunization as late as 30 years after primary immunization induced a strong and long lasting antibody response. However, these data from Denmark cannot be generalized, since different vaccines and different immunization schedules were used.

Some studies suggest the possibility of an insufficient response to a booster dose given 15 years after the primary series *(Collier et al. 1979)*. The booster response may vary individually and a pronounced dispersion of antibody levels can be expected with time following a primary series.

6.3 Tetanus immunity in different age and sex groups

Protection against tetanus benefits only those who are immunized; unimmunized persons are at risk of tetanus from contaminated wounds, including the umbilical stump.

The introduction of tetanus toxoid into childhood immunization programmes has changed the pattern of immunity against this disease considerably. As shown in Figure 6, the highest level of immunity is seen in the younger segments of the population, who have been covered by routine immunization. This has resulted in a dramatic decrease in the number of tetanus cases reported in younger persons *(Christenson & Bottiger 1987, Galazka & Kardymowicz 1989, Simonsen et al. 1987c)*. There is a clear difference between levels of immunity among men and women

Figure 6. Tetanus immunity in men and women in different age groups (geometrical mean titers or percentage of persons with titers 0.01 to 0.1 IU/ml or higher) in several countries (Galazka 1988).

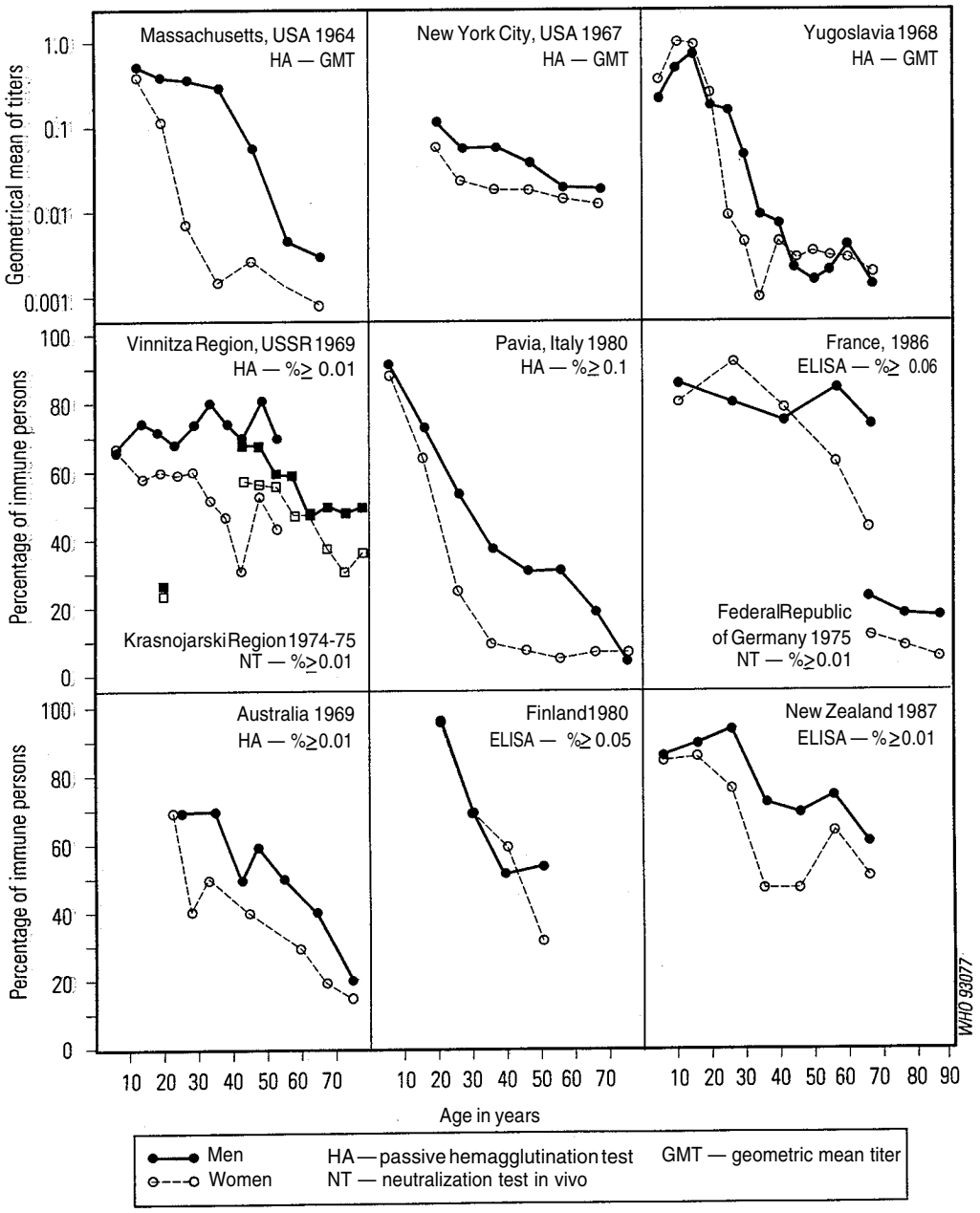
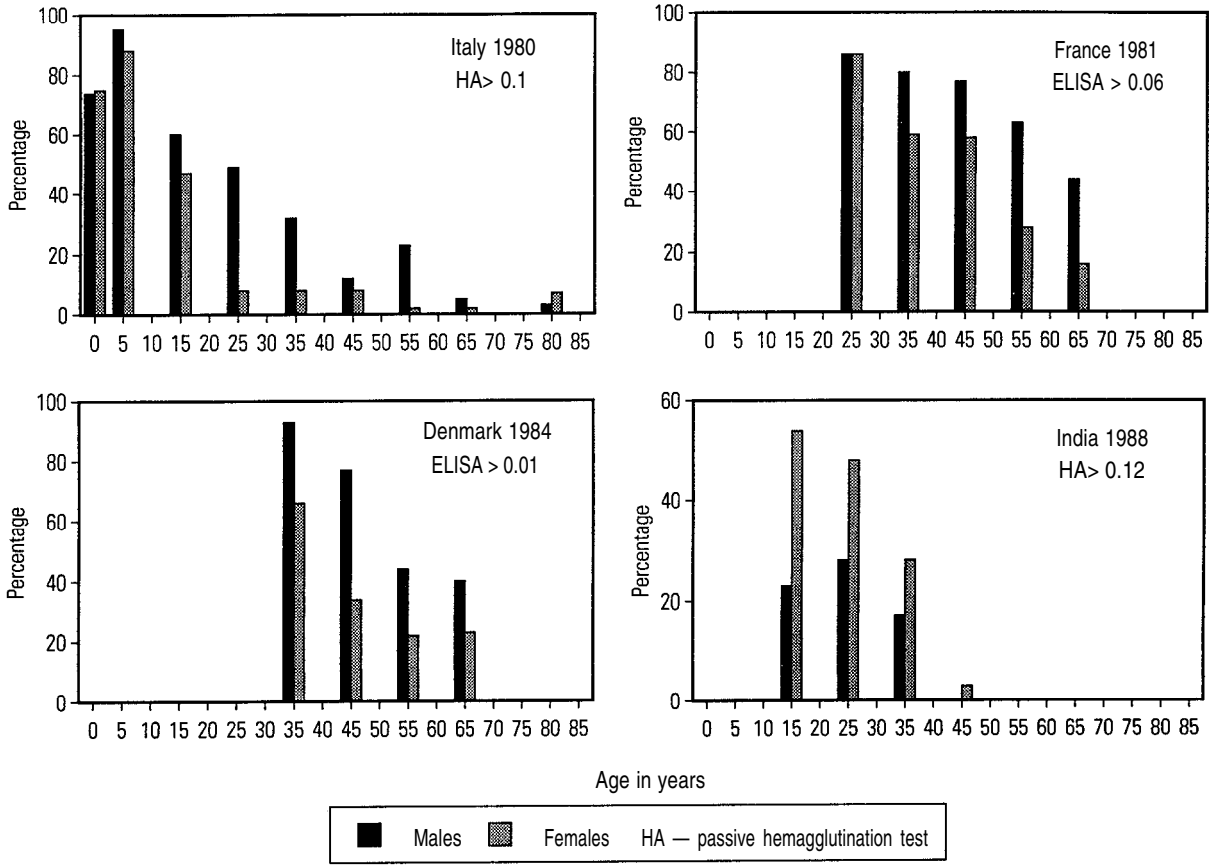


Figure 7. Percentage of persons with protective tetanus antibody levels, by age and sex, in four countries *(Bourland 1984, Gasparini et al. 1980, Kjeldsen et al. 1988, Misra & Rao 1988).*



of older ages. In developed countries, men are better protected, probably due to additional immunizations given during military service or professional activities. In India, a country where tetanus toxoid is given to women of childbearing age, and especially to pregnant women to prevent neonatal tetanus, the immunity status of women is better than that of men (Figure 7).

7. Placental Passage of Tetanus Antitoxin

7.1 The placenta as a selective organ

Tetanus antitoxin passively transferred from immunized mother to fetus provides transient protection of the newborn infant from tetanus. The human placenta regulates the transfer of antibodies from mother to fetus in a selective manner; transplacental transfer is restricted to IgG immunoglobulin. Fetal IgG antibody levels rise progressively from the fourth month of pregnancy until term. At birth, the infant usually has a total tetanus antibody concentration equal to, or sometimes higher than, his mother. Early studies found that the tetanus antitoxin levels in cord serum and maternal serum were usually equal, although in 20% to 30% of cases the cord serum had lower a titer than the maternal serum. Recently, it was observed that the cord/maternal ratio of tetanus antibodies is higher in European than in African settings *(Gendrel et al. 1990a, 1990b).* This may be linked to high immunoglobulin levels in African mothers exposed to multiple antigenic stimuli. Transplacental transfer of tetanus antibodies was better in the low range of maternal IgG than in the high range of maternal IgG.

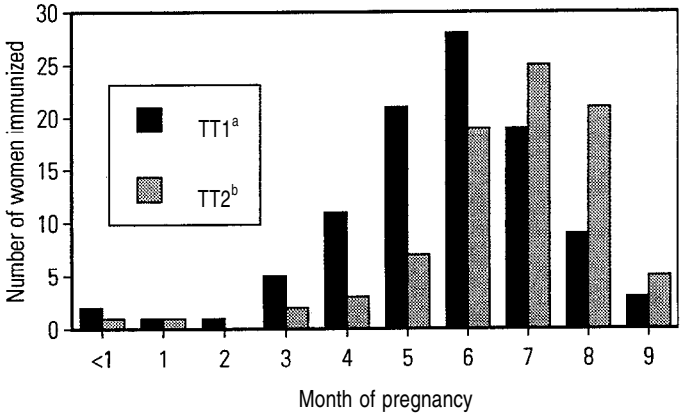
7.2 Influence of interval between TT doses and between the last dose and delivery on the amount of antitoxin transferred to the fetus

The ratio of antitoxin in maternal serum to antitoxin in cord serum depends on the intervals between doses of tetanus toxoid and the interval between the last dose and delivery. Longer intervals between doses of tetanus toxoid in the initial series increase the height and duration of the immune response (Table 5). Long intervals between doses of toxoid are best for achieving the optimal immunological results. However, in reality, pregnant women in developing countries often report to health centers

Table 5. Tetanus antitoxin level in cord sera of newborns whose mothers were immunized with two doses of tetanus toxoid administered at different intervals *(Dhillon & Menon 1975).*

Interval between toxoid doses (weeks)	No. of samples tested	% distribution of antibody levels (UI/ml) in cord sera		
		> 0.01	> 0.1	> 1.0
4 to 8	238	70.6	37.0	8.4
9 to 12	210	81.1	62.4	15.7
13 to 16	133	92.5	71.4	22.6
over 16	142	90.8	73.9	39.4

Figure 8. Time of TT immunization during pregnancy, Lagos State, Nigeria *(EPI Review 1989).*



[a] Mean time for TT1 = 5.7 months
[b] Mean time for TT2 = 6.6 months.

for the first time and are immunized when pregnancy is already advanced, (Figure 8). Often, the second dose of tetanus toxoid is given just before the delivery, which diminishes the possibility of effective transfer of a significant amount of antibody from the mother to the fetus. The cord/maternal ratio of tetanus antibodies increases as the interval between the second dose and delivery is prolonged *(Stanfield et al. 1973).* These data strongly support the policy of starting immunization as early as possible in the pregnancy, to assure long enough intervals between doses and between the second dose and delivery.

7.3 Interference between passive antibodies and development of active immunity

The rate of decrease of tetanus antitoxin during the neonatal period *(Kryl et al. 1964, Sangpetchsong et al. 1985)* is similar to that for antibodies against *Neisseria meningitidis group A, Haemophilus influenzae type b* and *Streptococcus group B* induced by polysaccharide vaccines given to mothers during pregnancy *(Amstey et al. 1985, Baker et al. 1988, McCormick et al. 1980).* After one month, about 80% of antitoxin transferred from the mother is still present in the circulation of the newborn.

With an increasing proportion of women immunized with tetanus toxoid, more and more infants will have high levels of passively acquired tetanus antitoxin. Such passive immunity could suppress the development of active immunity following early administration of DPT vaccine. Results of one study showed some interference between passive immunity acquired from mothers immunized three times during pregnancy and active immunity following two doses of DPT vaccine administered at 2 to 6 months and 3 to 7 months *(Kryl et al. 1964).* The interference was accentuated in infants who had cord serum titers above 0.1 IU/ml. Data from Thailand on infants immunized at 3, 4, and 6 months of age show a suppressive effect of passive immunity after the first dose of DPT vaccine, but not following the two subsequent doses (Figure 9).

In developed countries, the majority of women of childbearing age are immune against tetanus. In the USA, the mean tetanus antibody level in cord serum is high, exceeding 10 IU/ml, when measured by the hemagglutination test *(Anderson et al. 1988).* With a half-life of about one month, the antibody level determined by the neutralization test declines to 0.3 to 0.5 IU/ml by the age of two months, when the first dose of DPT vaccine is administered *(Barkin et al. 1984, Edwards et al. 1989).* This level of passive immunity interferes with the first dose(s) of DPT, but the third dose of DPT exerts a strong antigenic stimulus (Figure 10).

8. Safety of Tetanus Toxoid

Tetanus toxoid is a very safe antigen. Although acute anaphylactic reactions were reported in the 1940s, it is believed that they were due to the presence of sensitizing agents (peptones) from the culture media. The improvement of manufacturing techniques, the development of culture media free from sensitizing agents, and the use of more purified products has considerably decreased the risk of untoward reactions.

Severe generalized reactions are extremely rare. Local minor reactions may occur in a small proportion of vaccinees. Reactions following tetanus toxoid have been reported in hyperimmunized persons, i.e. in persons immunized with multiple doses in the past and showing high antibody levels at the time of injection *(Collier et al. 1979, Edsall et al. 1967, Relihan 1969, White et al. 1973, White 1980).* These reactions are due to the presence of antibody, which forms complexes with the injected toxoid. These antibody-toxoid complexes attract complement and leukocytes to produce localized vascular damage, with ensuing local swelling, pain, and malaise *(Edsall et al. 1967).* Overuse of tetanus toxoid can also lead to polyneuropathy *(Holliday & Bauer 1983, Rutledge & Snead 1986).* The estimated incidence of polyneu-

ropathy is 0.4 cases per million doses of tetanus toxoid *(Quast et al. 1979).*

Some countries still have a policy of administering two doses of tetanus toxoid during each pregnancy. Since overuse of tetanus toxoid is undesirable, such a policy should be stopped and replaced by the five-dose schedule. An optimal and effective immunization schedule requires use of durable and portable records of tetanus toxoid immunization.

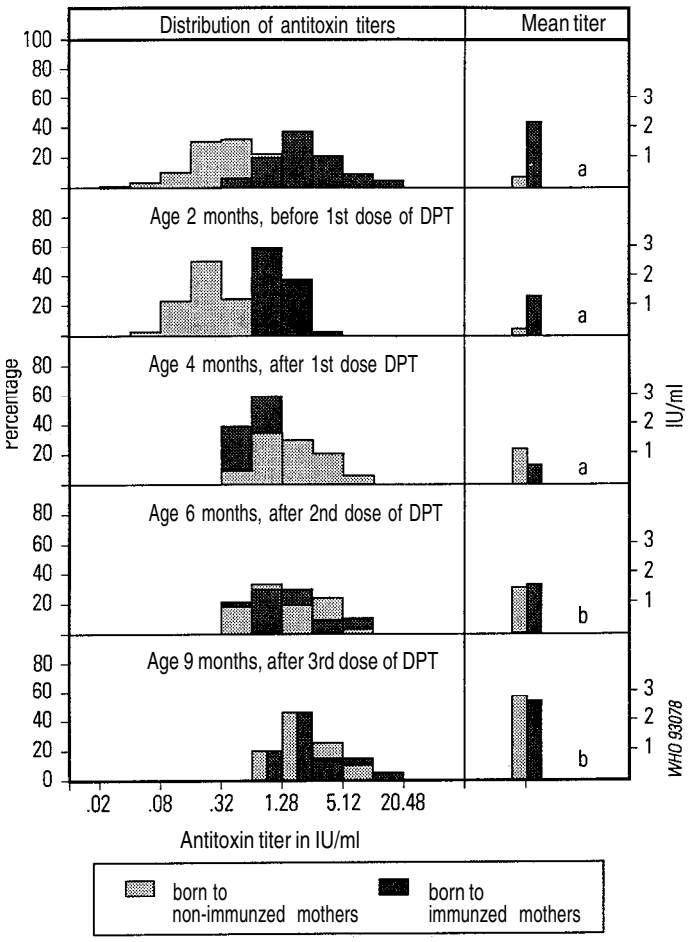
Tetanus toxoid is considered safe in pregnant women. There is no convincing evidence of risk to the fetus from immunizing the pregnant women with tetanus or diphtheria toxoids *(ACIP 1989).* In the United States, the only vaccines recommended for administration during pregnancy are tetanus and diphtheria *(American Academy of Pediatrics 1986, American College of Physicians 1990).* Three groups of investigators have studied tetanus toxoid use in early pregnancy. Freda *(1956)* observed no difference in the incidence of complications of pregnancy in a group of 107 pregnant women immunized against tetanus, cholera and typhoid, compared with pregnant women who were not immunized. Immunizations were administered during the first trimester in half of these women. Heinonen et al. *(1977)* analyzed the outcome of 9222 pregnancies in which women were immunized during the first four months with different vaccines, including tetanus toxoid. No significant risk of congenital abnormalities or abortion was found in immunized women. Schofield et al. in a study in Papua New Guinea *(1961)* gave non-adsorbed tetanus toxoid as early in pregnancy as possible and found no excess in the number of stillbirths.

EPI recommends giving the first dose of tetanus toxoid as early as possible during pregnancy. Usually women report late for antenatal care and consequently in most countries immunization does not start earlier than the fourth or fifth month of pregnancy.

9. Implications for Immunization Programmes

An optimal immunization schedule aimed at protecting the newborn against neonatal tetanus depends on the history of immunization of the mother with vaccines containing tetanus toxoid. When cohorts of women who are entering the childbearing years have not been immunized with tetanus toxoid in their infancy or adolescence, the implementation of a five-dose tetanus toxoid schedule is of utmost importance. The most effective strategy is immunization of pregnant women. This approach, however, is difficult to implement and by 1992 only 43% of pregnant women in the developing world had received at least 2 doses of tetanus toxoid *(EPI Information System 1993).*

Figure 9. Tetanus antitoxin titers in DPT-immunized infants whose mothers were immunized or not immunized against tetanus *(Sangpetchsong et al. 1985).*

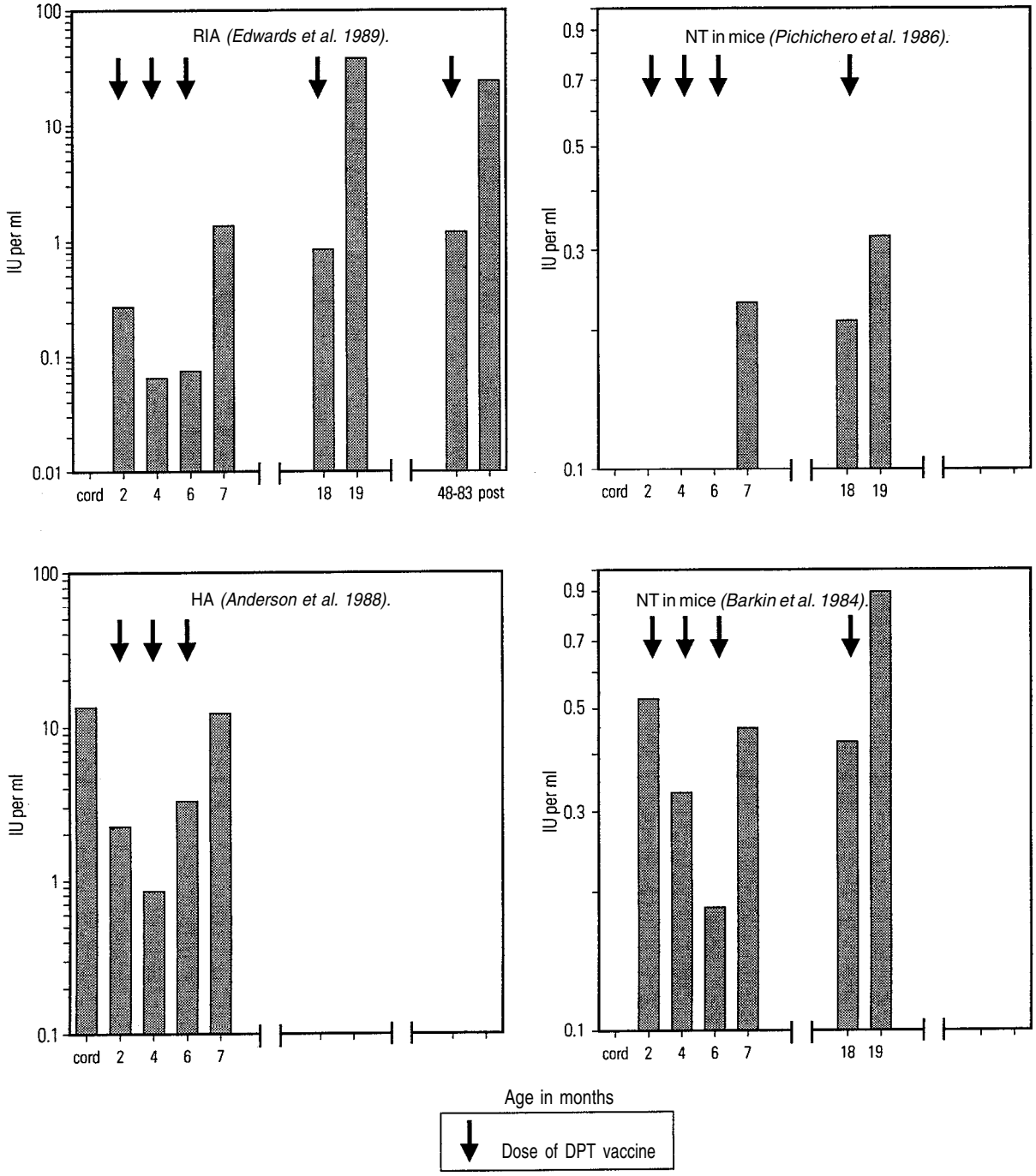


a p<.05. b Not significant.

Since 1987 the EPI Global Advisory Group has recommended tetanus toxoid immunization for all women of childbearing age *(Expanded Programme on Immunization 1988).* Tetanus toxoid immunization status should be checked whenever women contact health services, e.g. when they bring their children to an immunization session and when they attend curative services for themselves or their children. Another alternative is special outreach sessions, which can be held at markets, meetings, and in the workplace. All women should be asked for immunization records and immunized with tetanus toxoid, if needed. Every effort should be made to immunize these women with the first three doses of tetanus toxoid; this basic course will provide protection for at least 5 years. A fourth dose will prolong the duration of immunity for 10 years and a fifth dose will prolong it for 20 years.

An important approach may be immunization of school children. Children in the primary grades may be immunized with DT (child type) or Td (adult type); older children should receive tetanus toxoid or Td vaccine. The most practical immunization schedule usually involves two primary doses given at school

Figure 10. Tetanus antibody levels in children immunized in the USA with DPT vaccine at 2, 4, 6, 18 months and 4 to 6 years of age.



entry (with at least 4 weeks interval between these doses) and a third dose administered in the next grade (e.g. one year after the second dose). A fourth dose can be given at school leaving. In countries where such a schedule has been implemented for many years and where a high proportion of the female population attends school, revision of the immunization policy for adult women may be warranted. Theoretically, if a woman has received four doses of tetanus toxoid at school age, a single dose of tetanus toxoid given, in the first pregnancy will provide immunity lasting for the next two decades. The implementation of such a scheme must, however, be documented by immunization records retained by a high proportion of girls or by the results of serological studies.

In some countries, a large proportion of women reaching childbearing age may have received tetanus toxoid in childhood. The selection of an immunization policy will vary, depending on the schedule used and coverage levels achieved. Women who received only three or four doses of DPT in infancy, without additional booster doses, will most likely have lost circulating antitoxins before reaching the childbearing age (Figure 4); however, they will retain the capacity to respond to a tetanus toxoid booster dose.

Table 6. Guidelines for tetanus toxoid (TT) imunization of women who were immunized in the past.

Age at last vaccination	Previous immunizations	Recommended immunizations	
		At present contact/pregnancy	Later (at intervals of at least one year)
Infancy	3 DPT	2 doses of TT*	1 dose of TT
Childhood	4 DPT	1 dose of TT	1 dose of TT
School age	3 DPT + 1 DT/Td	1 dose of TT	1 dose of TT
School age	4 DPT + 1 DT/Td	1 dose of TT	none
Adolescence	4 DPT + 1 DT at 4-6 yrs + 1 TT/Td at 14-16 yrs	none	none

**At least 4 weeks between doses.*

The fall-off rate in immunity and the capacity to respond to a booster dose depends on the number of tetanus toxoid doses received, the age at immunization, and the interval between the primary series and booster doses. Women who received only three doses of DPT in early infancy or more than 10 years ago, may have diminished capacity to respond to tetanus toxoid booster. Therefore, it would be prudent to give them two doses of tetanus toxoid and complete the full immunization with one dose of tetanus toxoid in the next pregnancy or one year later: Women who can provide evidence of four doses of DPT vaccine in childhood will need only one dose of tetanus toxoid in the present pregnancy and one additional dose of tetanus toxoid in the next pregnancy (Table 6).

In areas where a high proportion of women were immunized in early childhood with three or four doses of DPT vaccine and reimmunized with additional doses of DT, Td or TT during school age, the immunization plan for pregnant women may be further abbreviated. Women who can present evidence of one booster dose during school age, in addition to early immunization with DPT vaccine, may need only one additional dose during the first pregnancy. Women who received more than one dose of TT-containing vaccine at school age, plus DPT immunization in childhood, may not need any additional doses of TT. When immunization records on the previous immunizations are available, the decision is easy for individual cases.

Decisions about changes in immunization policy in a community should be undertaken carefully after thorough analysis of data on coverage levels of various cohorts or serological data. In such situations, any decision should made made in favor of "the safer" policy alternative. It is better to give one booster dose in excess than to leave the mother and her child unprotected.

Abbreviations

DPT	diphtheria-tetanus-pertussis vaccine
DT	diphtheria-tetanus vaccine, for children
ELISA	enzyme-linked immunosorbent assay
HA	passive hemagglutination test
HIV	human immunodeficiency virus
IU	international units
RIA	radioimmunoassay
Td	preparation of diphtheria and tetanus toxoids with a low amount of diphtheria toxoid, for adolescents and adults
ToBI	toxin binding inhibition test
TT	tetanus toxoid
TT2	second dose of tetanus toxoid

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The **Global Programme for Vaccines and Immunization,** established by the World Health Organization in 1994, defines its goal as "a world in which all people at risk are protected against vaccine-preventable diseases". The Programme comprises three units:

Expanded Programme on Immunization
Vaccine Research and Development
Vaccine Supply and Duality

The **Expanded Programme on Immunization** focuses on the prevention of selected childhood diseases and, through support to national immunization programmes, aims to achieve 90% immunization coverage of children born each year. Its goals are to eradicate poliomyelitis from the world by the year 2000, reduce measles deaths and incidence, eliminate neonatal tetanus as a public health problem and introduce hepatitis B vaccine in all countries.

Vaccine Research and Development supports and promotes research and development associated with the introduction of new vaccines into the Expanded Programme on Immunization. This includes research and development of new vaccines, improvement of immunization procedures and support to epidemiogical studies.

Vaccine Supply and Quality ensures adequate quantities of high quality, affordable vaccines for all the world's children, supports the efforts of governments to become self-reliant as regards their vaccine needs, and assists in the rapid introduction of new vaccines.

The **Global Programme for Vaccines and Immunization** produces a range of documents, audiovisual materials and software packages to disseminate information on its activities, programme policies, guidelines and recommendations. It also provides materials for group and/or individual training on topics ranging from repair of health centre equipment to curricula guidelines for medical schools, nursing colleges and training of vaccine quality control personnel.

For further information please contact:

Global Programme for Vaccines and Immunization
World Health Organization • CH-1211 Geneva 27 • Switzerland
Fax: +41 22 791 4192/93 • E-mail: GPV@who.ch