Immunity status against tetanus in young migrants: a seroprevalence study

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Summary. Background and aim of the work: Thanks to the highly effective vaccine, tetanus became sporadic in high-income countries with well-established primary childhood immunization programs, but it is common in low-income countries. The migrants, leaving countries with poor immunization programs or where vaccinations have been interrupted, may represent a new risk group for tetanus in host countries. A seroprevalence study was conducted to estimate the immunological status against tetanus in young migrants without vaccination documentation. Methods: After a careful assessment by vaccination services of the Local Health Authority, all migrants recently arrived in Italy were included in the serosurvey. Titers of anti-tetanus toxoid were measured using a commercial ELISA kit. Subjects were stratified by age and by WHO region. Antibody titers <0.10 IU/ml were considered to be seronegative, between 0.10 and 1.00 IU/ml as intermediate protection, and >1.00 IU/ml high protection. Results: From January 2004 to December 2019, 2,326 blood samples were collected. Mean age was 13.9 years with no differences between WHO regions. The percentage of the subjects without protective antibodies was 22.3%, with an intermediate level was 45.2%, with high titer was 32.5%. Among migrant coming from African and Eastern Mediterranean WHO regions, the highest percentages of seronegative titers and, at the same time, the low percentages of high protective levels were found. Titers decreased with age. Conclusions: The significant proportion of seronegative migrants and the decrease of protective titers increasing age, confirm the importance of the evaluation of the immunological status to employ the appropriate vaccination strategy.

Key words: tetanus, migrants, serological survey, seroprevalence, immunity, WHO region

Introduction

Tetanus is one of the rare diseases that is infectious but not communicable. Immunity to tetanus toxin is induced only by immunization, so the recovery from clinical tetanus does not result in protection against further attacks. On the contrary to what happens for diseases transmitted from person to person, the achievement of high vaccination coverage in children does not allow to obtain an indirect protective effect in the population. Therefore, tetanus can never be eradicated because it is impossible to eliminate spores from the soil and generally from the living environment (1-5).

The vaccine against tetanus allowed massive progress in controlling the disease. The epidemiology of tetanus has radically changed due to the availability of a highly effective vaccine since the 1930s. Tetanus became sporadic in several high-income countries, where well-established childhood primary immunization programs have made a major contribution in the drastic reduction in morbidity and deaths (6, 7). In these countries, however, most cases occur among unvaccinated elderly people (8-10).
A serosurvey study of six European countries in 2015 showed that 2–31% of people aged 65 had sub-protective antitetanus toxin antibody concentrations (11).

The disease remains an important public health problem in many parts of the world, particularly in low-income countries, where most of reported tetanus cases are birth-associated, as consequence of the unclean deliveries and umbilical cord care practices (6, 7). Moreover, these cases are indicators of inequity in access to immunization and to other maternal, newborn, and child health services (12, 13). The Maternal and Neonatal Tetanus Elimination (MNTE) initiative, i.e. a reduction of the incidence below one case for 1000 live births per district, over 1 year, is the common goal in all countries of the world (14, 15).

As of September 2019, 47 out of 59 countries identified as high-risk for maternal and newborn tetanus had fully eliminated the disease and over 154 million women were immunized against tetanus between 1999 and September 2019. However, 12 countries remain to be validated, of which 11 are in the African and Eastern Mediterranean regions (16).

During 2017, 82 tetanus cases were reported in 26 countries of the European Union (EU), with a notification rate of 0.02 cases per 100,000 population that is in the range reported since 2012. Italy together with Poland accounted for 54% of all notified cases. Italy, albeit with a slow and gradual reduction over the years, remains, at European level, the country with the highest number of cases, with an annual notification rate that remained stable between 0.08–0.1 /100,000 from 2013 to 2017. Of the 231 cases reported in Italy in this period, 78% occurred in the age group 65 years old and above (8-10, 17). In Italy, tetanus toxoid vaccine was introduced in 1938 and was initially compulsory only for military personnel. In 1963, it became mandatory for two-year-old children and for workers engaged in activities considered to be at high risk of infection, e.g. construction, farming, refuse collection and animal husbandry. From 1968, tetanus vaccination became mandatory for all newborns. According to National Vaccine Prevention Plan 2017-2019, tetanus vaccination schedule consists of a primary series of three doses of tetanus-diphtheria-acellular pertussis vaccine (DTPa) at the 3rd, 5th and 11th months of age, then two boosters at 6 and 12–18 years of age. Administration of additional booster doses is recommended for every 10 years of a combined tetanus-diphtheria-acellular pertussis vaccine (dTpa) (18).

In the last decades, migration flow towards Europe and Italy was highly intensified. In 2018, 30.4% of all the migrants at global level were in the European region. Within the European Union, Italy was at the third place with 8.7% of foreign legally resident citizens (19).

In 2018, the “Vaccine European New Integrated Collaboration Effort (VENICE)” survey group, conducted an extended survey among 30 countries in the European Union (EU) and European Economic Area (EEA), to map out immunization policies targeting irregular migrants, refugees and asylum seekers. The results from the survey showed that in the case of children/adolescent migrants, almost all (n.27) of the 28 countries having strategies for migrant immunization, offer all the vaccinations included in the National Immunization Programs, in line with the international recommendations (20).

In Italy, according to Law n.40/1998, regular foreign citizens are totally equated to Italian citizens as regards to all health services including preventive medical services, to safeguard individual and collective health (21).

The Italian Ministry of Health recommends to vaccine according to National Immunization Plan, based on age, all young migrants and adolescent who have insufficient documentation regarding prior vaccinations (22).

Due to its severity, tetanus poses a risk to unvaccinated or insufficiently vaccinated people. Since tetanus infection does not confer immunity, the migrants leaving countries with poor immunization programs or where vaccination series have been interrupted, can represent a new risk group for tetanus in host countries (20, 23–26).

The present study was undertaken to assess the immunity status against tetanus in young migrants who attended the Local Health Services to regularize their vaccination situation in line with the National Immunization Programme.
Methods

Study population

After a careful assessment by vaccination services of the Local Health Authority of Parma (a city with 190,000 inhabitants, in northern Italy), all migrants recently arrived in Italy, without or with incomplete vaccination documentation, were included in the serosurvey implemented between January 2004 and December 2019.

In this study, we evaluated foreign young children for quantitative determination of antibodies against Tetanus toxoid. According to the agreement “Good Clinical Practice Guidelines”, all samples were treated anonymously.

The migrants came from the six WHO regions: African Region (AFR), the Americas’ Region (AMR), the South-East Asia Region (SEAR), the European Region (EUR), the Eastern Mediterranean Region (EMR) and the Western Pacific Region (WPR).

Patients were reorganized into four age groups: less than 2 years, from 2 to 6 years, from 7 to 18 years and equal or more than 19 years according to Italian immunization schedule.

Serological analysis

Sterile human serum, kept at -20°C until the determination, was quantitatively analysed for antibodies IgG against Tetanus toxoid by using commercial ELISA kit (RIDASCREEN Tetanus IgG, R-Biopharm, Germany), and following the manufacturer’s instructions.

Titers of anti-tetanus toxoid ELISA <0.10 IU/ml were considered as seronegative, between 0.10 and 1.00 IU/ml as intermediate protection and >1.00 IU/ml as high protection (5, 27)

Statistical analysis

The data were described in terms of mean, standard deviation (SD), median, minimum and maximum values. The results were summarized in tables of frequency and the differences in the proportions were compared using Chi square test, with Yates’s correction of continuity when appropriate. The analysis of variance was applied when appropriate; otherwise, the median test was used to investigate any difference in the titers relative to the WHO region of origin or age class. P-values equal to or less than 0.05 were considered statistically significant. All statistical analyses were performed with SPSS 25.0 (IBM SPSS Inc., Chicago – IL).

Results

From January 1, 2004 to December 31, 2019, 2,326 blood samples were collected. The largest number was collected in 2011 years and the smallest in 2018 and 2019. In 176 cases, it was not possible to reconstruct the origin of the subjects. The remaining 2,150 subjects came from 85 different countries. Ten countries accounted for 65.8% of the samples. The African region, albeit with significant fluctuations over the years, provided the largest number of samples, with Senegal at 1st place (16.22%), followed by Ivory Coast and Ghana; India is at 4th place (8.48%) and the first European country was Albania at 9th place (2.5%) (Figure 1).

Overall, the AFR provided 49.4% of the samples, followed by EMR (13.4%), EUR (10.8%), SEAR (10.2%), AMR (8.7%) and WPR (7.5%). The average age was 13.19 years (SD 5.92) without statistically significant differences between WHO regions (Table 1). The median titer was 0.52 IU/ml. Overall on the WHO region of origin, 22.3% of the subjects had no protective antibodies (<0.10 IU/ml), 45.2% fell into the intermediate range and 32.5% of the subjects showed high titer of antibodies (> 1.00 IU/ml). The highest percentage of subjects without
protective antibodies was found in the AFR (28.2%) and in subjects coming from the EMR (28.0%). In these regions, there have also been low percentages of subjects with antibodies with a high protective titer: 27.2% and 27.0% respectively. Among the subjects from SEAR, the lowest percentage of non-protective titers was found (5.9%) (Table 2).

By stratifying the subjects by age group, the median titers were higher in early childhood, decreasing in the 2 successive age groups of pre-schoolers and children and adolescents (0.60 IU / ml, 0.56 IU / ml and 0.50 IU / ml, respectively), and then up in young adults. In particular, the median antibody titers in the age group 2-6 years and 7-18 years, were statistically significantly lower than titers observed in the age group > 19 years (p <0.05 with Bonferroni’s Test) (Table 3).

### Conclusions

Pediatric vaccination with diphtheria-tetanus-pertussis vaccine has traditionally been the cornerstone of Expanded Program on Immunization and is often used as an indicator of how well countries are providing routine immunization services.

The vaccination schedules in low-income countries are different from those adopted in Italy in compliance with WHO indications, i.e. many more doses for diphtheria-tetanus vaccination. Many factors, such as the difficult logistical situations in which the local health services operate, the difficulty of maintaining an optimal cold chain, the low immune level of the child population for concomitant diseases and malnutrition, the organizational impossibility of reaching all children at specific ages, may be causes of incomplete immunization.

Therefore, to ensure lifelong protection against tetanus, WHO recommends that all people should receive 6 doses (3 primary plus 3 booster doses) of tetanus toxoid-containing vaccine through routine childhood immunization schedules. The booster doses

### Table 1. Characteristics of the study sample

| Region | Subjects (No.) | Age Mean (SD) | Median | Min | Max |
|--------|----------------|---------------|--------|-----|-----|
| AMR    | 183            | 13.13 (5.37)  | 13.00  | 2   | 51  |
| AFR    | 1,057          | 13.39 (5.88)  | 14.00  | 1   | 55  |
| SEAR   | 217            | 12.36 (5.19)  | 12.02  | 2   | 35  |
| EUR    | 229            | 13.67 (7.26)  | 14.00  | 1   | 43  |
| EMR    | 286            | 13.10 (6.33)  | 13.40  | 1   | 40  |
| WPR    | 160            | 12.52 (4.63)  | 13.00  | 0   | 31  |
| Overall| 2,132*         | 13.19 (5.92)  | 13.60  | 0   | 55  |

*Subjects with both countries of origin and age data

### Table 2. Numbers and percentages of subjects with non-protective (0-0.1 IU/ml), intermediate (0.11-1.0 IU/ml), high (>1.00 IU/ml) tetanus antibody titers, by WHO Region

| WHO Region | Tetanus Antibody Titers (IU/mL) |
|------------|---------------------------------|
|            | 0 - 0.10 | 0.11 – 1.00 | > 1.00 |
| AMR        |   86     |       92    |   79   |
|            |   8.1%   |     49.5%   |  42.5% |
| AFR        |  1,063   |     474    |   289  |
|            |  28.2%   |     44.6%   |  27.2% |
| SEAR       |   219    |       98    |   108  |
|            |   5.9%   |     44.7%   |  49.3% |
| EUR        |   232    |       96    |   108  |
|            |  12.1%   |     41.4%   |  46.6% |
| EMR        |   289    |     130     |   78   |
|            |  28.0%   |     45.0%   |  27.0% |
| WPR        |   161    |       81    |   37   |
|            |  26.7%   |     50.3%   |  23.0% |
| Overall    |  2,150   |     971     |  699   |
|            |  22.3%   |     45.2%   |  32.5% |

### Table 3. Numbers and percentages of subjects with non-protective (0-0.1 IU/ml), intermediate (0.11-1.0 IU/ml), high (>1.00 IU/ml) tetanus antibody titers, by age class

| Age          | Subjects (No.) | Median Titer | Tetanus Antibody Titers (IU/mL) |
|--------------|----------------|--------------|---------------------------------|
| < 2 years    | 21             | 0.60         | 0 – 0.10 | 0.11 – 1.00 | > 1.00 |
|              |                |              | 0.00%  | 90.5%  | 9.5%  |
| 2 - 6 years  | 266            | 0.56         | 0.56 | 52    | 134   | 80 |
|              |                |              | 19.5%  | 50.4%  | 30.1% |
| 7-18 years   | 1,701          | 0.50         | 0.50 | 404   | 752   | 545 |
|              |                |              | 23.8%  | 44.2%  | 32.0% |
| ≥ 19 years   | 144            | 0.99         | 0.99 | 23    | 53    | 68  |
|              |                |              | 16.0%  | 36.8%  | 47.2% |
| Overall      | 2,132*         | 0.52         | 0.52 | 479   | 958   | 695 |

*Subjects with both countries of origin and age data
should be given at 12–23 months of age, 4–7 years of age and 9–15 years of age respectively.

In the EU/EEA countries, tetanus vaccination is recommended in infancy (3–4 doses in the first 2 years of life). All countries also recommend booster doses for children and teenagers after completing the priming vaccinations. Most of the Member States recommend a booster for adults who have reached 18 years of age or above.

In 2018, the global coverage rates for the third dose of the diphtheria, tetanus and pertussis (DTP3) vaccine reached 86%, up from 72% in 2000 and 20% in 1980. However, improvements have stalled over the current decade, and 83 countries have yet to achieve the Global Vaccine Action Plan target of 90% or greater coverage of DTP3 (28, 29).

Globally in 2018, 10 countries account for 11.7 of the 19.4 million under and un-vaccinated children in the world (60%). This list includes Nigeria, India, Pakistan, Indonesia, Ethiopia, the Philippines, Congo, Brazil, Angola and Vietnam, i.e. countries with moderate coverage and very large birth cohorts, and other countries with substantially lower coverage. In fact, coverage levels vary substantially across WHO regions: the gap between the best performer, the EUR, and the lowest performer, the AFR, is 18 percentage points (76% - 94% respectively) (7, 30, 31).

This epidemiological situation highlights the importance of the attention paid to foreign young population arriving in Italy without records of the main vaccine preventable diseases prior vaccinations. In particular, for tetanus, since protection is essentially based on artificially acquired immunity, the antibody dosage provides a useful indication of the immune status and can highlight any risk situation.

To our knowledge, from current literature, there is a paucity of data on tetanus immunity status of young migrants arrived in Italy, while more attention, in the epidemiological Italian context, is paid to the elderly population and to the professional categories at greater risk of tetanus infection (32-34).

Moreover, often, a limit in the interpretation of the results is represented by the differences in the laboratory tests used, in the cut-offs chosen as threshold values of effective protection (27).

From the literature, several prevalence and sero-prevalence studies about the vaccine-preventable diseases immunity status of migrants, also compared with the native population, attest to the importance of this topic (10, 11, 35-55).

In this study concerning 2,150 subjects coming from the 6 WHO regions and from 85 different countries, protective tetanus antitoxin levels (>0.10 IU/ml) were found in the 45.2% of subjects, and long-protective tetanus antitoxin levels (>1.00 IU/ml) in the 32.5%. To note that a significant percentage (22.3 %) of subjects were inadequately protected.

In a seroprevalence study conducted in the past on young Italian children, the percentage of subjects without protective antibody levels is comparable with our results. The Authors explain their data supported by documentation, as the result of non-compliance with the vaccination program (56).

It is well known that tetanus toxoid is a very effective antigen, and for persons whose primary vaccination schedule is completed, the effectiveness of the vaccine is over 95% (1, 2, 5).

In Italy, since 2013, there has been a decrease in vaccination coverage in children and low levels of adherence to vaccination in adolescence (55, 57, 58). In particular, the detection of the vaccination coverage for the single tetanus antigen, for vaccinations at 5-6 years and at 16 years (cohort 2012 and 2002) (data as at 31 December 2018) shows lower percentage values than those expected as optimal in the PNPV 2017-2019 (58).

From the analysis conducted stratifying the young migrants by age group, the median titers were higher in early childhood and decreasing in the age groups of pre-schoolers and children and adolescents. This evidence must be carefully evaluated, due to the decline of tetanus immune protection, if not strengthened by booster doses. The increase of antibody titers observed in young adult, could be attributed to appropriate tetanus prophylaxis practices during accidents, traumas. In this survey, the largest number of migrant (49.4 %) came from the African countries.

The highest percentage of migrants without protective antibodies and, at the same time, the low percentages of subjects with high protective levels come from AFR and EMR. Paxton et al. have also reported these results for east African immigrant children in
Australia (59). WHO estimates 8.5 million un-and unvaccinated children live in the AFR, almost as many as in all other regions combined (31).

In a wider European and global context, it is important to take this epidemiological situation into account, regarding risk of exposure and susceptibility of young migrants. The availability of serological investigations to assess the immunity status against many vaccine preventable diseases is an important tool for Public Health worldwide.

Conflict of interest: Each author declares that he or she has no commercial associations (e.g. consultancies, stock ownership, equity interest, patent/licensing arrangement etc.) that might pose a conflict of interest in connection with the submitted article

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