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## PREVALENCE OF REACTIVE TUBERCULIN SKIN TEST IN DENTAL HEALTHCARE WORKERS AND STUDENTS

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### Abstract

**Introduction.** The risk for active tuberculosis (TB) infection among dental healthcare workers (DHCWs) is controversial. Specifically, TB incidence is generally low among general dental practitioners, but it is higher than in the general population among hospital/prison dentists, DHCWs who work in endemic areas, students during clinical training with direct patient contact.

**Aim.** To investigate prevalence of reactive tuberculin skin test (TST) among dental students and DHCWs in an Italian dental university hospital.

**Material and methods.** DHCWs (n=76), dental (n=92) and dental hygienist (n=13) students, administrative staff (n=15, reference group), who were not previously vaccinated against TB, provided their consent to undergo the Mantoux test, the standard recommended TST. Unadjusted and adjusted odds ratios (ORs) of reactive TST for working categories and for years of practice in the dental hospital were assessed through logistic regression analysis.

**Results.** Prevalence of reactive TST estimates were 0.0%, 17% (95% confidence interval, 95CI, 11-26%), 20% (95CI, 12-30%) and 7% (95CI, 1-30%) among dental hygienist students, dental students, dental healthcare workers and administrative staff, respectively. The adjusted ORs for working categories were non-significant, but they were significant at 95% level for the years of exposure (OR, 1.09 for each year of practice at the hospital; 95CI, 1.01-1.19).

**Conclusion.** High prevalence values could be due to cross immunization from non-tubercular mycobacteria, frequently found in dental unit waterlines, or boosting from repeated TST occasions. However, these data suggest that the risk for TST conversion is associated with the years of practice in dental hospitals.

**Key words:** tuberculosis, tuberculin skin test, dental healthcare worker, dental student, dental hygienist, hospital

### Introduction

Mycobacterium tuberculosis is carried in airborne particles called droplet nuclei that are generated when persons with infectious tuberculosis (TB) disease cough, sneeze, shout or

talk. During these events, a multitude of droplets lower than 50 µm in size from the upper aero-digestive tract, known as aerosol, are emitted in the environment and rapidly desiccate, thus becoming droplet nuclei. They are 1-5 µm in size, are suspended in air for hours and are carried through air currents from a room to another. The probability of a person exposed to M. tuberculosis to become infected depends on the concentration of infectious droplet nuclei in the air and the duration of the contact with the source of infectious TB disease. Environmental factors, such as long stay in confined spaces, inadequate ventilation or recirculation of air containing infectious droplet nuclei, along with poor general health status of exposed individuals, may increase the likelihood of transmission. The persons at highest risk for M. tuberculosis infection are, therefore, individuals who share air space in households or in other indoor environments with persons with active pulmonary TB disease<sup>1</sup>. After initial infection, immune response generally prevents further multiplication and spread of M. tuberculosis and the subjects do not show symptoms of the disease, although bacteria can remain alive in lungs or other organs for years. This condition is known as latent TB infection, which can be diagnosed by reactive tuberculin skin test (TST). Some 5% of infected individuals develop active TB disease

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soon after infection, while another 5% develop active TB later in life in consequence of conditions, such as AIDS or malnutrition, associated with immune depression. Symptoms of active TB disease include productive cough, night sweats, fatigue, malaise, fever and weight loss. These are the only contagious TB subjects<sup>2,3</sup>.

The risk for *M. tuberculosis* infection transmission in dental healthcare settings could be associated with aerosol production, which is frequent during dental therapy as demonstrated since more than forty years ago<sup>4</sup>. In the year 1970, professional toothcleaning with ultrasonic scaler, which generates abundant aerosol and splatter, was performed on the labial surfaces of the mandibular anterior teeth of patients with suspected active TB disease. The purpose was to determine whether in these circumstances droplet nuclei containing *M. tuberculosis* were produced in the environment. These microorganisms were not detected in air samples collected immediately after the dental prophylaxis. This experiment, which would not be feasible nowadays, suggests that production of *M. tuberculosis* containing droplet nuclei from patients with active TB disease probably occurs only when fluids from the lower respiratory tract are emitted, while during dental therapy fluids from the upper respiratory tract and, most of all, saliva are emitted<sup>5</sup>. However, the risk for *M. tuberculosis* infection and TB transmission to patients and staff cannot merely be assessed through the investigation of the spread of microorganisms in the dental environment, but it is necessary to study the ascertained cases of transmission during dental treatment<sup>6</sup>. Actually, an outbreak of TB was reported in the year 1982 at two community dental clinics: primary lesions developed in fifteen patients following dental extractions. Thirteen of the lesions were oral TB, while one case was an active TB disease which involved mouth and lungs. The dental surgeon who did not routinely use protective barriers was the source of infection<sup>7</sup>. Such a case could not occur nowadays since almost all dental healthcare workers use masks and other protective barriers and, if affected by active TB disease, are prevented from working. This situation is common with many other infectious diseases<sup>8,9</sup>. A dentist with active pulmonary TB disease who treated at least 450 patients, including 60 children, during the period elapsed between the appearance of the

first symptoms and the final active TB diagnosis, did not transmit *M. tuberculosis* to any of his patients probably because he wore mask routinely<sup>10</sup>. The TB risk for patients is minimal if dental healthcare workers adopt few simple preventive measures<sup>3</sup>, but a different issue is the risk for dental staff. Indeed, TB was considered an occupational hazard for this category of healthcare workers few years ago<sup>11</sup>. A survey made in England and Wales in 1988 and 1993 seeking to determine whether healthcare workers were at increased risk of TB, reported that there were 48 health professionals with diagnosis of active TB and the risk for this disease was almost ten times higher than in the general population. However, only one of the diseased individuals was a dentist<sup>12</sup>. Conversely, according to another survey made in 1995 among the members of the American Association of Hospital Dentists, 12% of respondents reported that there was at least one case of TST conversion by dental healthcare providers within the past year at their institution<sup>13</sup>. These data suggest that the risk of *M. tuberculosis* infection would be particularly high among dental healthcare workers who practice in hospitals.

## *Aim*

The aim of this study was to investigate prevalence of reactive TST in a sample of dental students, and dental healthcare associates and professionals in a large university hospital in Italy.

## *Material and Methods*

This study was made in the year 2008 in a large dental hospital in the centre of Italy, where dental students perform their clinical training. The clinical and administrative staff was invited to participate and to provide their informed consent, while the internal review board of the hospital approved the study. In addition, dental students at their fourth and fifth (i.e., last) course years, as well as dental hygienist students at their second and third (i.e., last) course years were invited to participate. Criteria for eligibility were that subjects must not be affected by diabetes, kidney diseases, Crohn's disease, rheumatoid arthritis, cancer chemotherapy,

conditions which may produce unclear TST results. Volunteers were also excluded if they had a history of severe reaction to a previous TST, were vaccinated against TB with the Bacille of Calmette-Guérin (BCG) a strain of *Mycobacterium bovis*, or had fever the day of the testing occasion. The remaining participants provided their data and underwent the TST.

The TST used was the Mantoux test (Biocine Test PPD Liof 2D 5UI/D -Novartis Vaccines and Diagnostics, Siena Italy- this product is not available anymore in Italy since April 2009). TST tests are based on the fact that infection with *M. tuberculosis* produces a delayed-type hypersensitivity skin reaction to certain components of the microorganism. These components are contained in extracts of culture filtrates and are the core elements of the classic tuberculin Purified Protein Derivative (PPD), which is then used for skin testing. The standard recommended tuberculin test, known as Mantoux test, is administered by injecting a 0.1 mL of 5 tuberculin units of PPD intradermally in the forearm, choosing an area free of abnormalities and away from veins. Reaction in the skin is due to sensitized T cells which induce induration, that is, a hard, raised area with clearly defined margins around the injection site. The Mantoux test was preferred to the widespread and cheaper Tine test, a multiple puncture test, because the latter provides a higher number of false positive results and tends to overestimate TST positives<sup>14</sup>.

Skin tests were read 48-72 h after the injection when the size of the induration was maximal. Reactive (positive) TST among students and the administrative staff with normal immune system must show at least one induration greater than 15 mm, while among dental healthcare workers, indurations  $\geq 10$  mm were considered reactive TST. Since in this study the response to the TST was crucial, it was necessary to obtain the most reliable results as possible. For this reason, the test was performed by

one trained healthcare worker with more than ten years of experience and a specific TST caliper was used to read the induration size. These two measures, guaranteed the consistency of test results<sup>15</sup>.

Differences in mean age, gender distribution and average years of hospital practice (i.e., exposure) among the four categories of dental healthcare workers, dental students, dental hygienist students and administrative staff members were statistically analysed using the  $\chi^2$  test (cell  $\chi^2$  test for pairwise differences) and the analysis of variance -ANOVA (Bonferroni-Dunn post hoc test for pairwise differences). Prevalence of reactive TST with 95% confidence interval (95CI) and the unadjusted odds ratios (ORs) for reactive TST were estimated within the four categories, with the administrative staff as reference group. Logistic regression analysis also was used to estimate the ORs for reactive TST test within the four categories, adjusted for gender, age and years of exposure. In order to prevent that excess collinearity may artificially inflate the coefficient estimates, Pearson's correlation coefficient between the two continuous variables, that is, age and years of exposure, was estimated. In the event that coefficient was higher than 0.5, the two variables were used in two different regression models. A level of significance of 95% was chosen.

## Results

Four dental healthcare workers were not included because they reported to be vaccinated with BCG. The sample, therefore, included 76 dental healthcare workers, 15 administrative staff members, 92 dental students and 13 dental hygienist students. Gender distribution was similar in the four categories, with males prevailing on females, excluding dental hygienist students who were 77% females, a statistically significant difference with the other catego-

**Table 1.** General characteristics of the sample.

category	number	males	females	age	years of exposure
Dental hygienist student	13	23.1%	76.9%	31.0 $\pm$ 8.5	1.3 $\pm$ .5
Dental student	92	72.8%	27.2%	26.3 $\pm$ 4.0	2.8 $\pm$ 0.4
Dental healthcare worker	76	65.8%	34.2%	33.6 $\pm$ 6.6	9.5 $\pm$ 6.4
Administrative staff	15	60.0%	40.0%	34.8 $\pm$ 8.7	0.0

Gender:  $\chi^2_{3df}$  test = 12.79;  $p=0.005$  – pairwise differences: dental hygienist students with the three other categories,  $p<0.05$

Age: ANOVA F-test = 24.97;  $p<0.0001$  – pairwise differences: dental students with the three other categories,  $p<0.05$

Years of exposure: ANOVA F-test = 52.02;  $p<0.0001$  – pairwise differences: healthcare workers with the three other categories,  $p<0.05$

ries (Table 1). Mean age was approximately of thirty years in all groups excluding dental students (26 years, statistically significant difference with the remaining groups). Years of exposure to the dental hospital settings was zero in the administrative staff and nine years in the healthcare workers, significantly higher than in the student categories.

Prevalence of reactive TST in the administrative staff, used as reference unexposed category, was almost 7% (Table 2) with wide confidence interval due to the small size of this

did not differ appreciably between model 1 and model 2. In addition, the analysis of the whole model fit, assessed through the likelihood ratio test, provided similar values of the statistic test ( $\chi^2_{25df}=11.63$ , model 1;  $p=0.04$  -  $\chi^2_{25df}=12.24$  model 2;  $p=0.03$ ), thus suggesting that the difference between models was minimal. The only variables significantly associated with reactive TST were age (OR=1.08, 95CI, 1.01-1.15 for each increasing year) and years of exposure (OR=1.09, 95CI, 1.01-1.19 for each increasing year).

**Table 2.** Prevalence of reactive tuberculin skin test (TST) among the four categories and unadjusted odds ratio for positive test.

category	reactive TST	95CI	unadjusted OR	95CI
Dental hygienist student	0.0%	0.0%	0.36	0.01-9.62
Dental student	17.4%	11.0%-26.4%	2.95	0.36-24.07
Dental healthcare worker	19.7%	12.3%-30.0%	3.44	0.42-28.26
Administrative staff	6.7%	1.2%-29.8%	1.00	-

group. Similar situation for dental hygienist students, with 0% reactive TST. Prevalence was considerably high among dental students (17%) and healthcare workers (20%). These values led to high unadjusted ORs for reactive TST of 3 and 3.4, respectively.

The Pearson’s correlation coefficient between age and years of exposure was 0.65 (Z-value, 10.58;  $p<0.0001$ ) (data not in Table). It was, therefore, impossible to include these variables in the same logistic regression model, because this option would produce excess collinearity. Two models were then designed including the four working categories, gender and age (model 1) and the four working categories, gender and years of exposure (model 2). The results, displayed in Table 3, refer to model 1 for all the coefficient estimates excluding for

As for the latter variable, the probability to be TST positive at the Mantoux test increased by 1%-19% for every working year in the dental hospital. The variable years of exposure was, therefore, more meaningful than the working category.

This important result induced to investigate the effect of years of exposure to the hospital setting on reactive TST probability. This variable was split into four categories of exposure, namely, 0-2 (reference category), 3-5, 6-10, >10 years. The unadjusted and adjusted ORs for reactive TST in these categories were assessed using simple and multiple logistic regression analysis. The covariates included in the multivariate model were gender and working category. The results, displayed in Table 4, suggest that the risk for reactive TST was marginally

**Table 3.** Multiple logistic regression analysis (adjusted OR) of the association between reactive TST and the four working categories, gender, age and years of exposure to the dental healthcare setting.

variables	adjusted OR	95CI
Dental hygienist student	10-6	-
Dental student	7.03	0.70-70.23
Dental healthcare worker	4.67	0.54-40.64
Administrative staff (reference)	1.00	-
Gender (female, reference)	0.71	0.30-1.65
Age (continuous)	1.08*	1.01-1.15
Years of exposure (continuous)	1.09*	1.01-1.19

\* $p<0.05$

Whole model fit. Likelihood ratio  $\chi^2_{25df} = 11.63$ ;  $p=0.04$  [model with years of experience and without age];  $\chi^2_{25df} = 12.24$ ;  $p=0.03$  [model with age and without years of experience]

the covariate “years of exposure” which was estimated through model 2. The coefficient estimates for the working categories and gender

significant (p-values ranging between 0.05 and 0.07 – data not in Table), within the first ten years, while it was statistically significant and

high (unadjusted OR, 9.38, 95CI, 2.26-38.90; adjusted OR, 10.10, 95CI, 2.38-42.85) when the years of exposure were more than ten.

among 474 dentists in the Indian Health Service and the Federal Bureau of Prisons (US), 7.6% resulted reactive for TST and 3.6% reported

**Table 4.** Simple logistic regression analysis (unadjusted OR) of the association between reactive TST and the four categories of exposure to the healthcare setting. Multiple logistic regression analysis (adjusted OR) including gender and the four working categories as covariates.

years of exposure to the healthcare setting	unadjusted OR	95CI	adjusted OR	95CI
Years 0-2 (reference category)	1.00	-	1.00	-
Years 3-5	3.43	0.94-12.45	3.70	0.99-13.77
Years 6-10	3.97	0.87-18.13	4.23	0.91-19.62
Years >10	9.38*	2.26-38.90	10.10*	2.38-42.85

\*p<0.05

Whole model fit. Likelihood ratio  $\chi^2_{3df} = 11.47$ ; p=0.009 [simple regression model];  $\chi^2_{7df} = 16.73$ ; p=0.01 [multiple regression model]

## Discussion

The present study is one of the papers presented at the workshop "Advances in Infection Epidemiology and Control in Dental Healthcare Settings", Department of Public Health and Infectious Diseases, Sapienza University, Rome, Italy on February 9th, 2013<sup>16-22</sup>.

The main limit of this study lies in the small sized sample, particularly in the control group. Indeed, the administrative staff included only 15 subjects, which resulted in low level of precision of the risk estimates. Therefore, high point risk estimates, such as those displayed in Tables 3-5, had wide confidence intervals and did not allow to make robust inference regarding the risk for TST conversion in dental hospitals.

Nevertheless, there were two important results, which could not be ignored. High prevalence of reactive TST in last course year dental students (17%, Table 2) and the increasing risk for TST conversion with the increasing years of practice in the hospital.

These data are comparable with published data regarding dental healthcare workers. The cited survey made in 1995 by the American Association of Hospital Dentists reported that cases of TST conversion within their institution were reported by 12% of respondents. This, of course, does not mean that there was a 12% conversion rate in hospital dentists from US, but that conversion was not unusual<sup>13</sup>. An Italian survey made in the year 2005 reported that 17% dental healthcare workers, including those vaccinated against M. tuberculosis, from a dental hospital were reactive for TST assessed using the Tine test<sup>23</sup>. According to a survey

that conversion occurred after graduation, suggesting that infection was acquired during practice among aborigines or in prisons<sup>24</sup>. Another survey made in the year 1995 compared self-reported reactive TST subsequently treated with anti-tuberculosis medication occurred during the period of practice among Air Force dentists and lawyers. 2.4% and 1.5% dentists and lawyers, respectively, reported to have required such medication<sup>25</sup>. Another study made among dentists working in US-Mexico border counties of Texas where TB prevalence was higher than in other US counties reported an annual TST conversion rate of 1.7%<sup>26</sup>. These data collectively suggest that the risk for reactive TST among dental healthcare workers depends on the characteristics of patients, specifically, relatively high risk in hospitals, as also reported in the present study<sup>13,23</sup> or in areas where TB prevalence is high<sup>26</sup> low risk for general dental practitioners<sup>24,25</sup>.

Paradoxically, dental students are more frequently investigated than dental healthcare workers, probably because in most countries dental students are invited to check their TB status before admission or before beginning their clinical training with direct patient contact. In addition, negative TST subjects are invited to undergo one or more BCG vaccinations, because clinical training is often made in hospitals, where patients are more likely to be affected by active TB. A previous study from Italy reported that 14% dental students at their last-course years were positive at the Tine test, including some vaccinated subjects as compared with 2% of controls who were not healthcare workers<sup>23</sup>. Another study from Japan reported that 60% medical/dental students were positive

at the Mantoux test, but 92% of the sampled subjects were vaccinated with BCG between one and four times. Despite such a high vaccination rate, 1% of them was considered suspicious of having an episode of TB infection<sup>27</sup>. A survey performed in 1994 in US, where dental students were invited to read the results of their Mantoux test by themselves, reported that 7.4% of them declared to be reactive to TST<sup>28</sup>. In the same year, 8.7% US dental students resulted reactive TST before starting their clinical training, with conversion rates of 4.2% during the first year and of 2.2% during the second year<sup>29</sup>. A last survey from US in 1994 reported a conversion rate of 10.6% after the first year of intensive clinical training<sup>30</sup>. Collectively, these data suggest that dental students are at risk for TST conversion and, perhaps, for *M. tuberculosis* infection during the period of clinical training, with conversion rates or prevalence values at the end of this period, which may be as high as in dental healthcare workers.

It is important to highlight, however, that such a high prevalence of positive TST among dental healthcare workers and dental students could be due to cross immunization between *M. tuberculosis* and other environmental mycobacteria, defined non-tubercular mycobacteria (NTM) isolated from water, as cases of TST conversion attributable to *Mycobacterium scrofulaceum* and *Mycobacterium intracellulare* are reported among medical students<sup>31</sup> and

aerosols coming from dental unit waterlines are often contaminated by NTM<sup>32</sup>. Another possible problem of TST is that in individuals who undergo frequent tests, such as healthcare workers and medical/dental students, "boosting" may occur. Boosting is a T-cell response due to repeated injections of PPD antigens. Such a response induces TST conversion even in absence of BCG vaccination or *M. tuberculosis* infection<sup>33</sup>. Therefore, the high prevalence reported in this and other studies among dental healthcare workers and dental students could be partly due to cross immunization from NTM or boosting from repeated TST occasions.

## ***Conclusion***

The data from this survey and from the literature suggest that prevalence of reactive TST among dental healthcare workers from hospitals and dental students during clinical training is relatively high. Such a high prevalence increases with the years of practice in dental hospitals. However, these data do not necessarily mean that the risk for *M. tuberculosis* infection is high because the cases of active TB among these working categories are few, and because frequent inhalation of NTM from aerosol generated by dental equipment and repeated TST occasions may artificially induce reactive TST conversion in absence of *M. tuberculosis* infection.

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