PILOT STUDY OF CONTACT SENSITIZATION TO RUBBER ALLERGENS AND BISPHENOL A AMONGST DENTAL STUDENTS

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Abstract
Objectives: The aim of this study has been to evaluate the rate of contact sensitization to some rubber allergens and to bisphenol A (BPA) amongst students of dental medicine and dental patients. Material and Methods: A total of 50 participants were included in the study: 40 students of dental medicine exposed to the studied rubber allergens and BPA-based dental materials during the course of their education; 10 dental patients without occupational exposure to the latter substances served as a control group. All of them were patch-tested with the studied rubber allergens and bisphenol A. Results: Highest was the sensitizing action of carba mix, followed by benzoyl peroxide and mercapto mix. The sensitization rate for carba mix was significantly higher for dental students as well as for the whole studied population, if compared to the one for thiuram mix (Chi² = 12.9, p < 0.001; Chi² = 13.9, p < 0.001), bisphenol A (Chi² = 8.9, p < 0.001; Chi² = 11.9, p < 0.001), toluenesulfonamide formaldehyde resin (Chi² = 10.7, p < 0.001; Chi² = 13.9, p < 0.001) and benzoyl peroxide (Chi² = 4.7, p = 0.03; Chi² = 5.8, p = 0.016), and for dental patients, if compared to the one for mercapto mix (Chi² = 7.07, p = 0.008). Concomitant positive skin patch-test reactions to carba mix and to benzoyl peroxide, and to all the studied allergens were established. Conclusions: Carba mix could be outlined as a sensitizer of paramount importance for dental students as well as for dental patients. Benzoyl peroxide was the second ranked sensitizer for dental students. Positive skin patch-test reactions to bisphenol A and toluenesulfonamide formaldehyde resin were established only among the group of dental students.

Key words:
Allergic contact dermatitis, Dentistry students, Carba mix, Mercapto mix, Thiuram mix, Bisphenol A

Funding: the study was granted by the Medical University – Sofia – project No. 4-C/2014 “Pilot investigation of urinary bisphenol A in students of dental medicine, students from Dental Technician School and in dental professionals, exposed during the practical education.” Project manager: Prof. Angelina Ilieva Kisselova-Yaneva, D.D.S., Ph.D., D.Sc.
Received: March 1, 2016. Accepted: April 12, 2016.
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INTRODUCTION
Dental professionals are occupationally exposed to numerous chemical substances as ingredients of various dental materials, medicines and disinfectants, many of which represent both allergens and irritants. This exposure starts as early as during the course of their practical education in dentistry and lasts all over their professional activity. As a preventive measure to protect them against dermal exposure and development of occupational contact dermatitis, the use of proper protective gloves is needed.
On the other hand, this could impose a risk of sensitization to some of their ingredients. Among the various substances known to cause occupational allergic contact dermatitis, additives to rubber comprise a conspicuous and meaningful subgroup. They are either remnants from the production process, e.g., vulcanization accelerators, or added to enhance the technical properties of the final product such as plasticizers, colors, antioxidants or antiozonants [1]. Rubber accelerators are chemicals used for speeding up the manufacturing process of rubber (vulcanization). This process makes untreated natural rubber latex suitable for its use in the manufacture of many rubber products. Nearly all rubber compounds contain rubber accelerators. The vulcanizers (accelerators) may occur both in occupational and non-occupational context [2]. A considerable amount of unreacted accelerator remains in the cured rubber product, migrates to the surface and comes into contact with the skin [3].
Contact allergy caused by rubber allergens after repetitive skin contact is frequent [4]. The highest-frequency work-related allergens are thiuram mix, carba mix, and epoxy resin. Work sources include commercial and agricultural fungicides and pesticides, conveyor belts, gas masks, protective rubber aprons, rubber hoses, seals and cables, earphones, electrical cords, rubber tires and tubes, safety goggles, shock absorbers, springs. Main applications of rubber in medical devices being a source of rubber allergens in dentistry include dental dams, examination and surgical gloves, rubber stoppers in medical syringes. Consumer exposures are numerous as well – household/recreational rubber products, clothing and footwear, cosmetics, healthcare products, etc. [5]. Contact dermatitis among dental professionals is a worldwide problem. A high prevalence of occupational contact dermatitis is seen with frequent “wet work” that is common in dental practice [6]. In addition to the fact that frequent glove use contributes to decreased barrier integrity, gloves constitute the significant source of chemicals inducing allergic contact dermatitis.
An increased prevalence of sensitization to thiurams among nurses with occupational contact dermatitis has been known since the 1990s. Contemporary data is important to allow appropriate preventive measures and identification of contact allergy trends. Although the prevalence of latex allergy has been reduced by decreasing powder and protein content of gloves, the use of rubber accelerators such as carbamates and thiurams still persists in latex and nitrile gloves [7].
Bisphenol A (BPA) is a chemical substance that has been used for hardening plastics for more than 40 years. It is produced in large quantities to be used primarily in the production of polycarbonate plastics and epoxy resins. It may be found everywhere – in water bottles, lining of canned foods, medical devices, resin based on epichlorhydrin and bisphenol A to be used in adhesives, surface coatings, electrical insulation, laminates, surface coatings, paints and inks, polyvinyl chloride (PVC) products, vinyl gloves, etc. [8]. Some dental sealants and composites may also contribute to BPA exposure [9]. Bisphenol A is a known endocrine disruptor [10]. Its capacity as a contact sensitizer in occupational and non-occupational exposures [11] as well as in dental patients [12,13] has also been recognized. No data in the available literature was found to concern the frequency of contact sensitization to rubber allergens and BPA among dental students.
The purpose of this study has been to evaluate the frequency of contact sensitization to some rubber allergens and to bisphenol A among students of dental medicine and dental patients.

MATERIAL AND METHODS

Subjects
A total of 50 participants were included in the study: 40 students of dental medicine exposed to the studied rubber allergens and BPA-based dental materials during the course of their education (Group A); 10 dental patients without occupational exposure to the latter substances served as a control group (Group B). General characteristics of the studied subjects are presented in the Table 1.

Skin patch testing
Skin patch testing with carba mix (3.0% pet), thiuram mix (1.0% pet), mercapto mix (2.0% pet), benzoyl peroxide (1.0% pet), toluenesulfonamide formaldehyde resin (10.0% pet) and bisphenol A (1.0% pet) (Chemotechnique Diagnostics, Vellinge, Sweden) was performed according to the Jadassohn & Bloch classical methods for diagnosis of contact allergy, by placing the allergens in IQ Ultra hypoallergenic patches of Chemotechnique Diagnostics (Vellinge, Sweden) (IQ Chambers®). Lack of anti-allergic medication constituted a mandatory condition before placing the patches and during the testing. Patches with allergens were applied on the back of the tested individuals, reading of the test was performed on day 2, several hours after removing the patches, with control revision on day 3. Interpretation of reaction sites was based on the method recommended by the International Contact Dermatitis Research Group (ICDRG). Interpretation key based on recommendations by the ICDRG was applied.

The study was approved by the Medical Ethics Board at Medical University – Sofia, Bulgaria. All the participants were informed about the purpose of the study and gave their written informed consent before its commencement.

Statistics
The statistics were calculated by means of the SPSS 19.0. The following statistics available for cross-tabulation were used: Fisher’s exact test for statistical significance, \( \chi^2 \) test. Values of \( p < 0.05 \) were accepted as statistically significant.

RESULTS
Distribution by gender was not uniform, with predominance of men in the investigated population but without statistical significance (\( \chi^2 = 3.9, p = 0.321 \)). The mean

<table>
<thead>
<tr>
<th>Variable</th>
<th>students of dental medicine (group A)</th>
<th>patients without occupational exposure (group B)</th>
<th>total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age [years] (M±SD)</td>
<td>25.30±9.1</td>
<td>47.70±17.6</td>
<td>29.78±14.3</td>
</tr>
<tr>
<td>Sex [n (%)]</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>males</td>
<td>19 (47.5)</td>
<td>4 (40.0)</td>
<td>31 (100.0)</td>
</tr>
<tr>
<td>females</td>
<td>21 (52.5)</td>
<td>6 (60.0)</td>
<td>19 (100.0)</td>
</tr>
<tr>
<td>Total [n (%)]</td>
<td>40 (80.0)</td>
<td>10 (20.0)</td>
<td>50 (100.0)</td>
</tr>
</tbody>
</table>

M – mean; SD – standard deviation.
As shown in the Table 2, highest was the frequency of positive skin patch-test to carba mix, and lowest – to thiuram mix. Furthermore, a significantly higher frequency of sensitization to carba mix if compared to other allergens was established for both Group E and the whole studied population. Summary of data is presented in the Table 3 and the Table 4.

Table 2. Positive skin patch-test reactions to the studied allergens among the studied groups taking part in evaluating the rate of contact sensitization to some rubber allergens and to bisphenol A (BPA)

<table>
<thead>
<tr>
<th>Allergen</th>
<th>Respondents with positive reaction [n (%)]</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>students of dental medicine (group A) (N = 40)</td>
</tr>
<tr>
<td>Carba mix</td>
<td>17 (42.5)</td>
</tr>
<tr>
<td>Thiuram mix</td>
<td>3 (7.5)</td>
</tr>
<tr>
<td>Mercapto mix</td>
<td>7 (17.5)</td>
</tr>
<tr>
<td>Benzoyl peroxide</td>
<td>8 (20.0)</td>
</tr>
<tr>
<td>Toluene sulfonamide formaldehyde resin</td>
<td>4 (10.0)*</td>
</tr>
<tr>
<td>Bisphenol A</td>
<td>5 (12.5)*</td>
</tr>
</tbody>
</table>

* Positive reactions were observed only in the group of dental students.

Table 3. Statistical significances concerning the frequency of sensitization to carba mix compared to the other studied allergens for the students of dental medicine exposed to the studied rubber allergens and BPA-based dental materials during the course of their education

<table>
<thead>
<tr>
<th>Allergen</th>
<th>Chi²</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thiuram mix</td>
<td>12.90</td>
<td>0.001</td>
</tr>
<tr>
<td>Bisphenol</td>
<td>8.90</td>
<td>0.001</td>
</tr>
<tr>
<td>Toluene sulfonamide formaldehyde resin</td>
<td>10.70</td>
<td>0.001</td>
</tr>
<tr>
<td>Benzoyl peroxide</td>
<td>4.65</td>
<td>0.030</td>
</tr>
</tbody>
</table>

Table 4. Statistical significances concerning the frequency of sensitization to carba mix compared to the other studied allergens for the studied groups – students of dental medicine exposed to the studied rubber allergens and BPA-based dental materials during the course of their education and dental patients without occupational exposure to the latter substances served as a control group

<table>
<thead>
<tr>
<th>Allergen</th>
<th>Chi²</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thiuram mix</td>
<td>13.89</td>
<td>0.001</td>
</tr>
<tr>
<td>Bisphenol</td>
<td>11.90</td>
<td>0.001</td>
</tr>
<tr>
<td>Mercapto mix</td>
<td>7.07</td>
<td>0.008</td>
</tr>
<tr>
<td>Toluene sulfonamide formaldehyde resin</td>
<td>13.89</td>
<td>0.001</td>
</tr>
<tr>
<td>Benzoyl peroxide</td>
<td>5.80</td>
<td>0.016</td>
</tr>
</tbody>
</table>
No statistically significant differences regarding the frequency of sensitization to the investigated rubber allergens and BPA and the type of exposure (during the process of practical education in dentistry or during treatment with relevant dental materials), age and gender of the studied population were revealed.

DISCUSSION

Numerous studies clearly indicate the importance of rubber allergens as contact sensitizers among health care workers. Carba mix, thiuram mix, mercapto mix, toluene-sulfonamide formaldehyde resin and benzoinperoxide are considered to be among the most important sensitizers. Carba mix is a mix of substances used as activators and accelerators for natural rubber, styrene-butadiene, and butyl rubber. It could be found in rubber products such as: household/recreational rubber products – anti-slip carpet backing, elastic bands, rubber garden hoses and kitchen gloves, rubber handled sports equipment, rubber swim caps and goggles; clothing and footwear – elastic in underwear and swimwear, rubber boots, sport shoes, slippers, rubber insoles of shoes, elasticized waistbands; cosmetics and healthcare products – rubber make-up sponges, rubber latex condoms [14].

Warburton et al. (2015) [4] analyzed data from 12 European countries collected by the European Surveillance System on Contact Allergies (ESSCA) network between 2009 and 2012. The prevalence of allergy to carba mix was 2.29%, and was significantly increasing. They conclude that inclusion of carba mix in the European baseline series may be appropriate [4]. The latter statement is confirmed by our findings. Notably, highest was the rate of sensitization to carba mix in the group of dental students – 42.5% vs. 30% in the control group of dental patients, without statistical significance. Moreover, the rate of positive skin patch-test reactions to carba mix was significantly higher if compared to the other studied by us rubber allergens and to BPA and concomitant positive reactions were established with each of the studied allergens and with BPA. We could outline carba mix as a sensitizer of paramount importance for dental students as well as for dental patients.

The thiurams are regarded as the most important class of contact allergens among the vulcanizers, partly due to cross-reactivity (allergy) with corresponding dithiocarbamates, which are used for similar purposes [3]. Thiuram mix is a mix of substances used as additives to rubber products to prevent degradation of the rubber and improve its function. The applications of thiuram mix are similar to the one of carba mix. This type of substances may be added to products such as gloves, shoes, insoles sponges, cables, tyres, elastics, handles, balloons, toys, rubber in underwear, wallpaper. Some ingredients are also used as anticorrosive agents and seed disinfectants.

The main sources of thiuram mix in medicine and in dental practice include rubber gloves, bandages and medical devices, renal dialysis equipment [14]. Bauer et al. (2015) [15] analyzed data of all employed patients patch-tested between 2003 and 2013 in the German Departments of the Information Network of Departments of Dermatology and established by means of the significantly increased prevalence ratio (PR) (indicating risk) for: thiuram-mix, mercapto-mix, zinkdiethyldithiocarbamate, mercapto-benzothiazole (MBT), mercapto-mix without MBT, and epoxy-resin, being the predominant occupational allergens at least associated with a doubled risk (PR ≥ 2) for acquiring occupationally allergic contact dermatitis. The highest risk increase was identified for employees in the health services. Schwensen et al. (2016) [6] conducted a retrospective observational study of the patch-test results of 1402 healthcare workers with contact dermatitis in Denmark between 2007 and 2014. They reported the potential problem of contact allergy to thiurams among healthcare workers with contact dermatitis [6]. Similar results were obtained by Molin et al. (2015) [16], who found significantly increased rates of sensitization
produce a lace-like appearance [17]. In medicine, it is used in non-prescription drugs for the treatment of acne and as a keratolytic [18]. Benzoyl peroxide is an important bone cement component as well. Uncommonly, allergic reactions to orthopaedic joint implants due to benzoyl peroxide have been reported [19–21]. Benzoyl peroxide is also used as an initiator in dental applications [22]. Lynde et al. (2014) [23] retrospectively reviewed the charts of patch-tested patients diagnosed with burning mouth syndrome. Benzoyl peroxide was among the most common detected allergens (1%). In our study, surprisingly to some extent, benzoyl peroxide was the second ranked sensitizer, especially for dental students – the sensitization rate 20% vs. 10% for the group of dental patients. No similar results were found in the available literature, and we can’t make a definite conclusion about the role of exposure during the educational course in dentistry for the high sensitization rate for benzoyl peroxide. Further studies are needed.

Toluene sulfonamide formaldehyde resin was found to be among the top 20 allergens with the highest number of relevant positive patch-test reactions in a recently published study [24]. Toluene sulfonamide formaldehyde resin is a modifier and adhesion promotor for film forming natural and synthetic resins. It occurs in vinyl lacquers, nitrocellulose compositions (e.g., nail lacquers), PVA adhesives, veterinary medicaments, bandages and medical devices, renal dialysis equipment, cements [14].

As mentioned above, mercapto mix was recognized to be among the predominant occupational allergens at least associated with a doubled risk for acquiring allergic contact dermatitis. The findings from our pilot study confirm the importance of mercapto mix as a sensitizer for students of dental medicine, with the rate of sensitization 17.5% vs. 10% for the group of dental patients. Nevertheless, no statistically significant differences were revealed by the between-group analysis. Benzoyl peroxide is a widely-used initiator, curing and cross-linking agent in polymerization processes (primarily in the curing of unsaturated polyester resins, production of polystyrene and related resins, styrene polymers and other resins). It is an oxidizer used for bleaching edible oils, flour, bread and other food. It is used in the embossing of vinyl flooring, in special fast-drying printing inks, in printing pastes, and as a burn-out agent for cellulose acetate in mixed fabrics with viscose, silk or cotton to produce a lace-like appearance [17]. In medicine, it is used in non-prescription drugs for the treatment of acne and as a keratolytic [18].

Benzoyl peroxide is an important bone cement component as well. Uncommonly, allergic reactions to orthopaedic joint implants due to benzoyl peroxide have been reported [19–21]. Benzoyl peroxide is also used as an initiator in dental applications [22]. Lynde et al. (2014) [23] retrospectively reviewed the charts of patch-tested patients diagnosed with burning mouth syndrome. Benzoyl peroxide was among the most common detected allergens (1%). In our study, surprisingly to some extent, benzoyl peroxide was the second ranked sensitizer, especially for dental students – the sensitization rate 20% vs. 10% for the group of dental patients. No similar results were found in the available literature, and we can’t make a definite conclusion about the role of exposure during the educational course in dentistry for the high sensitization rate for benzoyl peroxide. Further studies are needed.

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Mercapto mix consists of the following chemicals: mercaptobenzothiazole, n-cyclohexylbenzothiazylsulfenamide, dibenzothiazyl disulfide and morpholinylmercaptobenzothiazole. They are used as vulcanizers and accelerators in the production of rubber-soled shoes and heavy-duty rubber gloves and are present in both natural and synthetic rubber products, similar to those listed above. They could be as well ingredients of cutting instruments oils, greases, coolants, anti-freezes, anti-corrosives, fungicides, adhesives, veterinary medicaments, bandages and medical devices, renal dialysis equipment, cements [14].
is produced through the reaction of methyl methacrylate and diglycidylether of bisphenol A [25]. The intraoral polymerization of dental composites is not 100% and unpolymerized residual monomer may leach out to the surrounding oral mucosa [26]. In spite of the widespread use of bis-GMA, intraoral reactions are not common [27]. Bisphenol A-diglycidyl ether methacrylate sensitivity is a rare cause of allergic contact stomatitis in dental patients because they are briefly exposed to the resin before it is polymerized and becomes non-allergenic [28].

As mentioned above, BPA is used as a starting substance in the production of ingredients found in many dental composites and sealants, e.g., bis-GMA [29]. So, residual BPA, not chemically converted into bis-GMA, is likely present in trace amounts in any dental material containing these ingredients of dental composites and sealants degrading in the oral cavity by the action of salivary enzymes [30]. Thus, dental treatment with composite restorations could be a possible source of exposure to BPA for each individual being currently or potentially dental patient [31] as well as for occupationally exposed dental professionals and for dental students during their educational course.

No data was found in the available literature concerning the sensitization to epoxy resin/BPA among dental students. Intriguing data was obtained in this pilot study – positive skin patch-test reactions to BPA were detected only among the group of dental students, the sensitization rate being relatively high – 12.5%. This finding from our pilot study also needs further confirmation.

Since this is a pilot study, the sample is too small to give categorical statement. Further work is needed to validate reliability of these findings.

CONCLUSIONS
This pilot investigation demonstrated the relatively high rate of sensitization to the rubber allergens and BPA among dental students. Carba mix could be outlined as a sensitizer of paramount importance for dental students as well as for dental patients. Benzoyl peroxide was the second ranked sensitizer for dental students. Positive skin patch-test reactions to bisphenol A and toluenesulfonamide formaldehyde resin were established only among the group of dental students.

The provision of adequate information on occupational chemical hazards should start as early as during the first years of education in dentistry in order to protect dental students and dental professional from occupationally-induced health disorders.

REFERENCES


