P05-059
Levels of mercury in amalgam tattoos

G. Guzzi[1,*], F. Spadari[2], G.P. Bombeccari[2], I. Bolengo[1,*], L. Brambilla[3], S. Ferrucci[3], A. Ronchi[4], P.D. Pigatto[5], F. Pallotti[6]

[1] *Italian Association for Metals and Biocompatibility Research – A.I.R.M.E.B., Dental Toxicology, Milan, Italy*
[2] *Ospedale Maggiore Policlinico Fondazione Ca' Granda IRCCS, University of Milan, Department of Biomedical, Surgical and Dental Sciences, Unit of Oral Pathology and Medicine, Milan, Italy*
[3] *Fondazione IRCCS Ca' Granda – Ospedale Maggiore Policlinico, Operative Unit of Dermatology, Milan, Italy*
[4] *IRCCS Maugeri Foundation and University of Pavia, Italy, Pavia Poison Control Center and National Toxicology Information Centre, Toxicology Unit, Pavia, Italy*
[5] *IRCCS Galeazzi Hospital, University of Milan, Milan, Italy, Department of Biomedical, Surgical and Dental Sciences, Unit of Oral Pathology and Medicine, Milan, Italy*
[6] *Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, University of Milan, Unit of Anatomical Pathology, Milan, Italy*

Question: Mercury is thought to be deposit in amalgam tattoo, which are oral localized black-bluish pigmentation, but there are no reports documenting the presence of inorganic and/or organic mercury in amalgam tattoos in the oral cavity in humans.

Methods: The content of total mercury in amalgam tattoo due to mercury released from mercury-containing amalgam fillings was determined in 5 specimens of amalgam tattoo. We analyzed at about 50–200 mg wet weight of tissue for each specimen and fixed in 10 percent formaldehyde, and compared with the levels of total mercury of 20 gingiva specimens without amalgam tattoo.

Results: Total mercury levels were elevated in all specimens of amalgam tattoo we analyzed (5 of 5), as it greatly exceeds the normal range 2 μg/g (toxic level, >200). The mean mercury level in amalgam tattoo was 82.12 μg/g as compared with 0.204 micrograms per gram in controls. We found that the content of the total mercury in amalgam tattoo specimens ranged from 3.2 to 358 μg/g. In one amalgam tattoo tissue with the highest mercury level was also observed a high level of silver (Ag) 94.5 μg/g (threshold limit value).

Conclusions: Amalgam tattoo had elevated total mercury levels.

http://dx.doi.org/10.1016/j.toxlet.2015.08.492

P05-060
Quantification of buprenorphine and the metabolites of methadone and heroin in hair of patients in rehabilitation programs by GC–MS

D. Lima[1,2,*], F. Marques[2], A. Duarte[2], C.M. Afonso[3], F. Carvalho[1], R. Dinis-Oliveira[1,*]

[1] *University of Porto, Laboratory of Toxicology, Porto, Portugal*
[2] *Fluminense Federal University, Department of Analytical Chemistry, Niterói, Brazil*
[3] *University of Porto, Center of Medical Chemistry, Porto, Portugal*

Hair analysis provides information about use of drugs over a long period. In the present study a methodology aimed the simultaneous quantification of buprenorphine, of methadone and its metabolites, 2-ethylidene-1,5-dimethyl-3,3-diphenylpyrrolidine (EDDP) and 2-ethyl-5-methyl-3,3-diphenyl-1-pyrrolidine (EMDP), and of heroin metabolites, morphine and 6-acetylmorphine (6-AM), in hair samples collected from drug abusers enrolled in methadone treatment program for heroine addition. Ethylmorphine was used as internal standard. Washing, spiking and extraction studies of hair drugs were developed. After decontamination with *n*-hexane, an amount of 30 mg of powdered real hair samples were placed in a tube and 1 mL of hydrochloric acid 0.1 mol/L. After overnight incubation at 60 °C the solution was neutralized with 1 mL of NaOH 0.1 mol/L and buffered with 1 mL of phosphate buffer pH 7.0. Samples were centrifuged at 4000 rpm for 5 min, the supernatant being collected and purified by solid-phase extraction using mixed-mode extraction cartridges (MCX). The analytes were eluted with 2 mL of a mixture of methanol:ammonia (5%). The obtained solutions were evaporated to dryness under a stream of nitrogen and subsequently resuspended with 50 μL derivatizing N-methyl-N-(trimethylsilyl) trifluoroacetamide (MSTFA). Blank, standards and samples were analyzed by gas chromatography/electron impact-mass spectrometry (GC-EI/MS). Selectivity, linearity, analytical limits (i.e., limit of detection and low limit of quantification – LOD and LLOQ), carryover, precision, accuracy and recovery were evaluated. A number of 10 hair samples from human subjects following a long-term methadone therapy were analyzed by this method. All analytes were identified except EMDP, possibly because its represents less than 10% of methadone metabolites and hair excretion may not be substantial. Methadone and morphine were found in all samples, 6-AM in four samples and EDDP in three samples of the samples analyzed. In conclusion, the developed method can be applied in forensic and clinical routine during therapeutic rehabilitation programs.

http://dx.doi.org/10.1016/j.toxlet.2015.08.493

P05-061
Gynecomastia associated with mercury-containing dental amalgam fillings

I. Bolengo[1,*], A. Ronchi[2], P.D. Pigatto[3], G.P. Bombeccari[4], F. Spadari[4], G. Guzzi[1]

[1] *Italian Association for Metals and Biocompatibility Research – A.I.R.M.E.B., Dental Toxicology, Milan, Italy*
[2] *IRCCS Maugeri Foundation and University of Pavia, Pavia Poison Control Center and National Toxicology Information Centre, Toxicology Unit, Pavia, Italy*
[3] *IRCCS Galeazzi Hospital, University of Milan, Milan, Italy, Department of Biomedical, Surgical and Dental Sciences, Unit of Oral Pathology and Medicine, Milan, Italy*
[4] *Ospedale Maggiore Policlinico Fondazione Ca' Granda IRCCS, University of Milan, Department of Biomedical, Surgical and Dental Sciences, Unit of Oral Pathology and Medicine, Milan, Italy*

Question: We present a case where gynecomastia was associated with overexposure to mercury-containing amalgam. July 2002, a 31-year-old man was referred to us because of permanent sensation of metallic taste. He has simply not taste for water and he had chronic fatigue. December 2001, he had a painful enlargement of the left breast that was diagnosed as idiopathic gynecomastia. He showed 7 mercury fillings and one gold crown on his upper right first molar.

Methods: Patient received an extensive laboratory investigation.

Results: Levels of mercury in chewing gum-stimulated whole saliva was 211.6 μg/l [limit values: were elevated in association with the dyspepsia, and was a clear-cut evidence of toxic local effect on papillae gustative due to mercury exposure.

Conclusions: Our case report documented a temporal relation between mercury amalgam-replacement and the resolution of signs and symptoms of gynecomastia.

http://dx.doi.org/10.1016/j.toxlet.2015.08.494