REVIEW AND ANALYSIS OF THE LITERATURE ON THE HEALTH EFFECTS OF DENTAL AMALGAM

EXECUTIVE SUMMARY

The Sponsor's charge to LSRO and the Expert Panel

The Trans-agency Working Group on the Health Effects of Dental Amalgam composed of representatives from the National Institutes of Dental and Craniofacial Research (NIDCR) of the National Institutes of Health, the Center for Devices and Radiological Health of the U.S. Food and Drug Administration, the Centers for Disease Control and Prevention, and the Office of the Chief Dental Officer of the Public Health Service requested that the Life Sciences Research Office (LSRO), acting as a subcontractor to BETAH Associates, Inc., undertake an independent third-party review. LSRO was asked to consider the peer-reviewed, primary scientific and medical literature published between January 1, 1996 and December 31, 2003 that contributed to an understanding and evaluation of the potential adverse human health effects that may be caused by dental amalgam. Unlike other recent reviews of the dental amalgam literature (Berlin, 2002), LSRO was not asked to provide policy recommendations or perform risk assessment or risk-benefit analyses. LSRO was simply asked to review the literature within the specified time period to determine if it supported hypotheses relating to adverse health effects. This review was undertaken in consultation with LSRO's Expert Panel that was composed of scientific experts in the fields of immunotoxicology, immunology, and allergy; neurobehavioral toxicology and neurodevelopment; pediatrics; developmental and reproductive toxicology; toxicokinetics and modeling; epidemiology; pathology; and general toxicology (Appendix A). No member of the Expert Panel expressed a public opinion regarding the potential adverse health effects of dental amalgam prior to or during the review period. The inclusion of the names of the Expert Panel in Appendix A does not imply that each individual endorsed all of the statements in this report. The LSRO accepts full responsibility for the study conclusions and accuracy of the report.

The dental amalgam controversy

Dental amalgam is a widely used restorative dental material. Most standard dental amalgam formulations contain approximately 50% elemental mercury. Mercury vapor is released from elemental mercury at physiological temperatures and is absorbed by the human body. Dental amalgam was introduced as a restorative material over 150 years ago. Periodically throughout its history concerns have been raised about the potential human health effects due to the inhalation and absorption of mercury vapor from dental amalgam. The debate about the safety of dental amalgam continues today.

In the last ten years the U.S. Public Health Service (USPHS) (U.S. Department of Health and Human Services, 1993 & 1997) and other agencies, including the European Commission (1998), the health agencies of Canada (Health Canada, 2004), Quebec (Conseil d'Evaluation des Technologies de la Sante du Quebec, 1997), and Australia (National Health and Medical Research Council, 1999), and the World Health Organization (WHO) (1997), have reviewed the safety of dental amalgam for human use. These scientific panels concluded that there was no scientifically relevant and definitive evidence that demonstrated a causal link between dental amalgam and adverse health effects, except in rare instances where individuals experienced local side effects or delayed hypersensitivity reactions. These reports, however, stressed that further research was required in many critical areas, due to a lack of conclusive scientific studies.

Despite the findings of these panels, the governments of Germany (German Ministry of Health *et al.*, 1997), Austria (Commission of the European Union *Ad Hoc* Working Group on Amalgam, 1998), and Canada (Health Canada, 2004) and its province of Quebec (Conseil d'Evaluation des Technologies de la Sante du Quebec, 1997) have recommended against the placement of dental amalgam restorations in certain patient populations that include but are not limited to children, pregnant women, and individuals with renal dysfunction or hypersensitivity to metals. In addition, the governments of Sweden and Denmark have banned and are currently phasing out all mercury-containing materials, including dental amalgam because of environmental protection efforts. In 2002, a literature review of the health effects of dental amalgam was undertaken for the Dental Material Commission of Sweden (Berlin, 2002). The review advocated that the timely elimination of dental amalgam from dental care would reduce the occurrence of hypersensitivity reactions and local side-effects observed in some dental patients, remove the occupational exposures to elemental mercury experienced by dental professionals, and prevent further environmental mercury pollution. All of the above mentioned recommendations and regulatory actions have provoked public debate in the United States about the safety of dental amalgam. This present literature review is designed to update the last USPHS review that was completed in 1997 (U.S. Department of Health and Human Services, 1997).

The approach to the charge

Based on the charge from the Sponsor, the literature review undertaken by LSRO and the Expert Panel was limited to peer-reviewed journal articles published between January 1, 1996 and December 31, 2003. Literature was primarily identified using abstract databases maintained by the National Library of Medicine (Appendix B). These references were supplemented with citations obtained from the Expert Panel; from bibliographic reference searching of review articles, books, and international scientific studies; and from literature recommendations submitted by the public in response to the Federal Register Request for Information on Dental Amalgam (Docket No. 03N-0169).

The Expert Panel and LSRO received both oral and written public comment from interested parties on the controversy surrounding the use of dental amalgam as a restorative material (Appendices C & D). Information was also presented to the Expert Panel and LSRO about the toxicokinetics of mercury, the material properties of dental amalgam, and the design of ongoing dental amalgam clinical trials by experts in their respective fields (Appendix C). Review articles published within the timeframe and a small number of seminal articles pre-dating this timeframe were used by the Expert Panel and LSRO as background material. Guidance documents from various international and federal agencies including, the World Health Organization (1991), the Agency for Toxic Substances and Disease Registry (1999), and the U.S. Environmental Protection Agency (EPA) (1997) as well as reports published by the National Research Council (2000) and the Institute of Medicine (2001; 2004) were also used as sources of information. Therefore, although the Expert Panel and LSRO were asked to consider only the peer-reviewed literature published since the beginning of 1996 in the weight of evidence deliberations, they were knowledgeable of prior studies and the current debates about the use of dental amalgam.

All peer-reviewed human, animal, and *in vitro* studies published since the beginning of 1996 that investigated the biochemical, behavioral, and/or toxicological effects resulting from exposure to dental amalgam, mercury vapor (Hg^0) , inorganic mercury (Hg^{2+}), or organic mercury (methyl and ethylmercury) were considered. The Expert Panel and LSRO were not asked by the Sponsor to review studies that evaluated the dental or material properties of dental amalgam, compared the risks and benefits of alternative replacement materials *versus* dental amalgam, or considered the environmental consequences of dental amalgam disposal. Papers dealing with these topics were, therefore, excluded because they were beyond the scope of this project. Letters, comments, news articles, editorials, lectures, and other non peer-reviewed documents were also excluded. Approximately 961 articles were identified as broadly meeting these inclusion criteria (Report Supplement).

The Expert Panel and LSRO adopted the U.S. EPA's General Assessment Factors (2003) to select relevant articles of significant scientific merit from the initial pool of 961 articles. These assessment factors provided a framework for evaluating the soundness, applicability and utility, clarity and completeness, uncertainty and variability, and evaluation and review of the published information.

As part of the process of evaluating the studies, the Expert Panel and LSRO made several decisions about how they would approach the available literature. Although other literature reviews of dental amalgam have placed emphasis on animal and *in vitro* studies (Berlin, 2002), human studies of mercury vapor or dental amalgam exposure provided the primary basis of the current review. In contrast to data generated by other study designs, data obtained from human studies are directly applicable to an assessment of adverse

human health effects attributed to dental amalgam. Case reports (*i.e.*, adverse health effects reported for one or a few individuals) present interesting information that may be used to generate hypotheses. Because single reports do not contribute to empirical research, case reports were evaluated individually and as a group.

A majority of animal studies published since the beginning of 1996 evaluated oral or parenteral exposures to $HgCl_2$. Because Hg^{2+} is not inhaled and is unable to cross the blood-brain barrier, only animal studies evaluating mercury vapor exposures (including dental amalgam) were considered as relevant to human dental amalgam exposures by the Expert Panel and LSRO. Studies of animal exposures to mercury vapor were evaluated for the toxicokinetic information and threshold values for observed effects that they provided. Numerous *in vitro* cell-based studies were published since the beginning of 1996. The Expert Panel and LSRO agreed that while *in vitro* studies may aid in an understanding of the mechanism of action, they are an unreliable determinant of human health risks.

The organic mercury literature was scrutinized. However, the Expert Panel and LSRO concluded that the organic mercury literature contributed little to the understanding of Hg^0 exposure from dental amalgam because of the dissimilar metabolism and toxicokinetics of organic mercury and mercury vapor in the human body. This decision was also supported by human studies that have failed to demonstrate that quantifiable amounts of either Hg^0 or Hg^{2+} are converted to organic mercury by the human body (Agency for Toxic Substances and Disease Registry, 1999; Barregard *et al.*, 1994a). Studies evaluating co-exposure to methylmercury and mercury vapor were considered by the Expert Panel and LSRO.

Approximately 300 of the initial pool of 961 studies met the criteria set by the Expert Panel and LSRO for scientific merit, appropriate study design, and relevant mercury exposure. Urine mercury was adopted as the most appropriate, widely-used biomarker to evaluate human exposure to mercury vapor. Urine mercury reflects cumulative exposure to mercury vapor over time and unlike other biomarkers is not subject to significant confounding by methylmercury. Some of the 300 studies that were selected reported biomarkers that were not appropriate for the measurement of mercury vapor exposure (*i.e.*, hair, nails, and unspeciated blood mercury) or biomarkers that lack standardized sample analysis (*i.e.*, saliva). Studies reporting conclusions based on these measures were accorded little weight in the weight of evidence deliberations. Generally, studies that evaluated occupational exposures to mercury vapor were better controlled and the exposures were better defined than studies evaluating exposures to dental amalgam (with only a few notable exceptions). As a result, the data reported by occupational studies were accorded more weight than the data reported by dental amalgam exposure studies.

The remaining studies were summarized and grouped by the type of exposure or adverse outcome that was evaluated. Criteria commonly used to assess biologic plausibility were used by the Expert Panel and LSRO to guide its discussions of whether the scientific evidence supported a causal relationship between dental amalgam exposure and adverse human health effects (Hill, 1965). The evidence regarding adverse human outcomes was evaluated from the perspective of epidemiological studies, secular trend data, animal toxicity studies, dose-response relationships, and the plausibility of biological mechanisms.

Conclusions

Based on a weight of evidence evaluation of the literature published between January 1, 1996 and December 31, 2003, the Expert Panel and LSRO came to the following conclusions:

Experimental evidence consistently demonstrated that mercury vapor (Hg^0) is released from dental amalgam restorations and absorbed by the human body. Numerous studies have demonstrated a positive correlation between the number of dental amalgam restorations or surfaces and urine mercury concentrations in non-occupationally exposed individuals. Mean urine mercury concentrations (HgU) were < 2 µg Hg/L in most surveys of the general population that were published since the beginning of 1996. Furthermore, approximately 95% of the study participants had HgU at or below the pre-1996 WHO estimate of approximately 4-5 µg Hg/L.

The long-term use of nicotine chewing gum (> 24 months) combined with intense chewing and > 20 dental amalgam surfaces presents the greatest chance that the HgU of non-occupationally exposed individuals may significantly exceed the mean HgU values measured for the general population and approach levels observed in workers exposed to Hg^0 (24.8 µg Hg/L highest reported HgU value for a nicotine gum-chewer). Adverse health effects for long-term nicotine gum chewers due to Hg^0 exposure were not evaluated in the literature. Bruxism and dental amalgam placement and removal appear to have less impact on exposure levels than the use of nicotine chewing gum.

Studies of occupationally-exposed individuals have yielded information that is directly applicable to assessing the likelihood of renal or other injury posed by dental amalgam. Hg^0-exposed workers serve as sentinels, as they are usually exposed to substantially higher Hg^0 levels than persons with dental amalgam restorations, although it is recognized that occupational exposures are typically 8 hours/day, 5 days/week for 20-30 years whereas persons with dental amalgam restorations experience lifelong exposures of 24 hours/day.

Current occupational exposure guidelines recommend that the HgU of workers not exceed the Biological Exposure Index of 35 µg Hg/L (reflecting the current Threshold

Limit Value Time Weighted Average of 0.025 mg/m^3). This limit was established to guard against the adverse neurological and renal effects that have been observed in some workers with HgU that chronically exceeds 50 µg Hg/L. Studies of chloralkali, natural gas, mercury production, thermometer, and fluorescent lamp factory employees were reviewed. Urinary N-acetyl-ß-D-glucosaminidase (NAG) activity, an early and sensitive, but nonspecific biomarker of kidney effect, consistently exhibited a modest, reversible increase in workers with urinary HgU = 25 to 35 µg Hg/L. There were two reports of slight decreases in tumor necrosis factor a (TNF-a) in workers, but the biological significance of reduced TNF-a levels has not been established. Thus, on the basis of a number of occupational exposure studies, there appears to be a substantial margin of safety between Hg^0 exposure of persons with dental amalgam restorations and occupational Hg^0 exposures that produce slight alterations in sensitive biochemical indices.

Case reports and studies of immune function consistently demonstrated that dental amalgam is capable of producing delayed hypersensitivity reactions in some individuals. These reactions usually present with dermatological or oral symptoms. For individuals exhibiting positive patch tests, the removal of dental amalgam restorations and their replacement with composite materials may promote the resolution of the observed symptoms. While there is evidence that a small portion of the human population demonstrates this allergic sensitivity, there is insufficient evidence for other types of sensitivity, such as genetic susceptibility. Insufficient research was done to support or refute the hypotheses that dental amalgam causes antibiotic-resistance in human gut or oral flora or is an etiologic agent in any autoimmune disease, including multiple sclerosis.

Studies in the area of neuropsychological function were primarily negative or reported conflicting findings. Some raised concerns regarding experimental control of relevant confounding variables. In total, these studies failed to support the hypothesis that Hg^0 exposure, at the levels released by dental amalgam, interferes with human neuropsychological function or acts as an etiologic factor for the neurodegenerative diseases - Parkinson's disease and Alzheimer's disease.

Insufficient evidence was published since the beginning of 1996 to support or refute the hypothesis that mercury exposure from dental amalgam restorations contributes to adverse pregnancy outcomes. Studies of human fertility suggest that occupational exposure to Hg^0 has little adverse effect on male fertility, but may increase the prevalence of dysmenorrhea in females.

The majority of the human reproductive and developmental literature focused on exposure measures. Inorganic mercury in the placenta, maternal blood, and cord blood correlate with maternal dental amalgam load. Both methylmercury and inorganic

mercury can be measured in breast milk. The relative proportions of these species depend on the frequency of fish consumption, dental amalgam status, and occupational exposures. High level exposures of pregnant rats (1.8 mg/m^3 for 1 hour/day on gestational days 14-19) and monkeys (0.5-1.0 mg/m^3 for 4-7 hours/day, 5 days/week) to mercury vapor induce behavioral deficits in the offspring, but no data are available to judge whether low level exposures also lead to such effects. Co-exposure of rats to high levels of both mercury vapor and methylmercury during gestation induced adverse behavioral effects greater than exposure to the vapor alone, but similar human assessments have not been performed for co-exposure to the two species of mercury.

The current data are insufficient to support an association between mercury release from dental amalgam and the various complaints that have been attributed to this restoration material. These complaints are broad and nonspecific compared to the well-defined set of effects that have been documented for occupational and accidental Hg^0 exposures. Individuals with dental amalgam-attributed complaints had neither elevated HgU nor increased prevalence of hypersensitivity to dental amalgam or mercury when compared with controls. The findings of these studies suggested that individuals with complaints self-attributed to dental amalgam should be screened for underlying dental, physical, and psychiatric conditions. In particular, these data indicate that many individuals presenting with dental amalgam-attributed complaints may suffer from affective symptoms independent of mercury exposure.

Although some individuals undergo chelation therapy to treat neurological, neurobehavioral, or mood complaints attributed to Hg^0 exposure from dental amalgam, animal studies evaluating the toxicokinetics of mercury removal from the kidneys and brain by the chelators, *meso*-2,3-dimercaptosuccinic acid (DMSA) and 2,3-dimercaptopropane-1-sulfonate (DMPS), demonstrated that these agents mobilize mercury from the kidneys, but not the brain. Chelators may cause adverse health effects, such as headache, dizziness, nausea, and the loss of essential metals. In addition, DMSA has been reported to be teratogenic and fetotoxic in animal models.

Research gaps

In general, many of the low level mercury vapor (Hg^0) or dental amalgam exposure studies that were evaluated by the Expert Panel and LSRO provided insufficient information to enable definitive conclusions. Many of the published studies did not report well-defined exposures, appropriate biomarkers of exposure (HgU, not hair, nail, or total blood mercury), or the duration and cumulative Hg^0 exposure of workers. In addition, many studies were conducted using too small groups of subjects without matched controls and did not account for potential confounding factors.

The Expert Panel and LSRO were not charged with making research recommendations. Various important research gaps, however, were identified during the course of the

review that when filled may definitively support or refute the hypothesis that dental amalgam causes adverse health effects. These research gaps include:

- Well-controlled studies using standardized measures that evaluate whether low level Hg^0 exposures (air levels < 0.025 mg/m^3 or HgU < 35 µg/L) produce neurotoxic and/or neuropsychological effects and, if identified, provide dose-response relationships for those effects.

- Studies that determine the effects of co-exposure to Hg^0 and methylmercury. There is no pharmacokinetic basis for assessing whether co-exposure to methyl mercury and Hg^0 result in additive nephrotoxicity or neurotoxicity. Blood, brain, kidney and urinary Hg^{2+} time-course data are needed for animals administered a range of doses of methylmercury, Hg^0, and methylmercury plus Hg^0. Target organ Hg^{2+} concentrations should be correlated with adverse renal and neurological effects.

- Studies that investigate whether low level *in utero* exposure to Hg^0 (air levels < 0.025 mg/m^3 or HgU < 35 µg /L) produces effects on the developing brain.

- Occupational studies that evaluate reproductive and pregnancy outcomes in large groups of workers with well-defined Hg^0 exposures.

- Studies that can be used to determine the amount of Hg^{2+} that is absorbed by the human neonatal gut from breast milk and what, if any, effect this exposure has on the brain development of infants.

- Well-controlled studies using standardized measures that investigate whether dental professionals have increased incidences of kidney disease, emotional instability, erethrism, pulmonary dysfunction, or other characteristics of occupational Hg^0 exposure.

- Studies that evaluate whether there is a genetic basis for sensitivity to mercury exposure and whether potential gender differences exist in the pharmacokinetics and toxicity of mercury.