Respondent’s Exhibit G
February 25, 2008

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Torts Branch, Civil Division
U.S. Dept. of Justice
P.O. Box 146
Ben Franklin Station
Washington, D.C. 20044

RE: Claims for vaccine injuries resulting in autism spectrum disorders, or similar neurodevelopmental disorder caused by thimerosal-containing vaccines

Dear Mr. Matanoski,

Thank you for asking for my evaluation of the above matter. This report encapsulates my opinions regarding both the general and specific causation issues regarding the question of whether thimerosal-containing vaccines could have played any role whatsoever in the etiology of the petitioners’ autism.

Because of the length of my report I have included a table of contents below:
My Background .................................................................................................................. 4
Summary of opinions ........................................................................................................ 6
Scientific basis for opinions ........................................................................................... 8
  Mercury poisoning does not manifest as autistic spectrum disorders 8
The petitioners do not have any known or accepted form of mercury toxicity.

12

Scientific Methodology In The Assessment Of Causation

12

*In vitro* studies do not provide sufficient scientific basis to conclude that a particular substance is a potential cause of any specific toxic effect in humans.

16

The *in vitro* studies of Deth.

19

It is not scientifically valid to draw conclusions regarding the toxicity of ethyl mercury or thimerosal from data on the toxicity of methyl mercury or any other form of mercury.

21

The reference dose for methyl mercury provides no support for the assertion that thimerosal from vaccines can cause autism.

24

The data from studies on inorganic mercury are both inapplicable and do not support an increased risk of ASD.

30

There is reliable scientific evidence that thimerosal is not toxic to humans, including infants and children, at doses delivered either individually or cumulatively, by thimerosal-containing vaccines.

32

There is no reliable scientific evidence that antibiotic therapy alters mercury levels after receiving thimerosal-containing vaccines.

35

Dr. Aposhian’s calculation of brain mercury concentrations after vaccination is misleading and fails to provide all of the relevant data.

36

The study of Hornig, upon which Drs Deth and Aposhian rely, has been conclusively shown to be incorrect and non-reproducible.

39

There is no reliable scientific evidence that chelation therapy improves the clinical manifestations of ASD, nor is it plausible.

40

There is no scientific evidence of a sub-population of children who are particularly vulnerable to mercury toxicity based on the inability to excrete mercury from thimerosal-containing vaccines or any other factor.

41
There is no vulnerable sub-population of children who could not excrete mercury and thus retain and accumulate it in their tissues, including their brains. 41

Studies on hair do not support any difference in mercury concentrations between autistic children and normal controls. 41

There is no reliable evidence that there is a quantitative difference in mercury in baby teeth of children with autism versus controls. 46

Chelation studies do not support a role for mercury in ASD. 47

There is no recognized disease in medicine known as a "mercury efflux disorder." 50

There is no sub-population of children with increased susceptibility to ethyl mercury or thimerosal that has ever been described. 50

Acrodynia is neither related to ASD nor can it be considered that it is an example of a susceptible population. 51

Polymorphism of the coproporphyrinogen oxidase gene has not been shown to be a cause of ASD and does not define a population susceptible to developing this condition. 52

The lack of association between thimerosal-containing vaccines and ASD is universally accepted in the mainstream medical and scientific communities. 54
MY BACKGROUND

I am a physician and hold the rank of Clinical Professor of both Pediatrics and Internal Medicine at the University of Colorado Health Sciences Center (UCHSC). I am board-certified in medical toxicology – a sub-specialty recognized by the American Board of Medical Specialties. I maintain a private medical practice in that field through my work at Toxicology Associates. Toxicology Associates is one of the largest medical practices in the United States devoted to the discipline of clinical toxicology. As a medical toxicologist, I specialize in the assessment, diagnosis, and treatment of adverse effects of pharmaceuticals and other agents on humans. At present, there are approximately 300 physicians in the United States holding sub-specialty certification in medical toxicology.

In addition to my M.D., I have earned a Ph.D. in biochemistry. My doctoral research focused on potential toxic effects of medicinal agents. Upon completion of my Ph.D. program at Mount Sinai School of Medicine, I served as a post-doctoral Fellow at the Institute of Cancer Research at Columbia University, College of Physicians and Surgeons.

Following completion of this Fellowship, I attended medical school at the State University of New York, School of Medicine at Buffalo, and was awarded an M.D. I then completed an internship in general surgery at the Harvard 5th Surgical Service in Boston, Massachusetts, followed by a primary residency in emergency medicine at Emory University School of Medicine in Atlanta, Georgia. After completion of my residency, I became a Fellow in medical toxicology at the UCHSC and the Rocky Mountain Poison Center. Upon completion of this 2-year fellowship, I was invited to join the faculty of both institutions and ultimately became the Director of the medical toxicology fellowship program, a position I held until 1993. Since my initial appointment in 1987, I have continuously remained on the UCHSC faculty, rising from Instructor, to Assistant, then
Associate, and finally to full Professor, the highest achievable academic rank at my institution. At UCHSC, and at my private practice at Toxicology Associates, I devote most of my time to patient care, research, writing, editorial activities, activities in professional medical societies, and teaching in the area of medical toxicology.

During the course of my career as a medical toxicologist, I have had the unique opportunity to evaluate, consult on, diagnose, and treat hundreds of patients for heavy metal poisoning, including mercury toxicity. I have published in the area of mercury toxicity and have given invited lectures on this topic both at national meetings in the U.S. and international congresses.

I have been active in the research area throughout my career. My research focuses on the adverse effects of, or poisoning by, pharmaceuticals and environmental or other toxins. I have been an author on over 200 peer-reviewed publications, invited articles, book chapters, abstracts, and other publications.

I have served as Editor-In-Chief of Toxicological Reviews and I am currently a Senior Editor of Clinical Toxicology, which is the largest circulation peer-reviewed journal in the world devoted to this discipline. I was recently appointed to the founding editorial board of the new Open Toxicology Journal. I serve as a peer-reviewer for a number of medical journals including the New England Journal of Medicine, the Journal of the American Medical Association, Journal of Emergency Medicine, Annals of Emergency Medicine, Academic Emergency Medicine, American Journal of Emergency Medicine, Archives of Internal Medicine, Clinical Toxicology, Critical Care Medicine, Journal of Medical Toxicology, and the American Journal of Kidney Diseases.
I have been Section Editor for environmental toxicology of one toxicology text (Emergency Toxicology, 2nd Ed., Little Brown) and I am Senior Editor of a major text entitled Critical Care Toxicology – The Diagnosis and Management of the Critically Poisoned Patient.

I am Past-President of the American Academy of Clinical Toxicology, which is the largest professional society in the world devoted to this discipline. Its membership includes virtually all medical toxicologists and Poison Control Center directors in the U.S. and Canada and a large proportion of those worldwide. Presently, I am a member of the Board of Directors of the American College of Medical Toxicology, which is the professional organization of physicians who are sub-specialty qualified in medical toxicology.

I have acted as a consultant to various federal agencies, the World Health Organization, multiple universities, the American College of Physicians, and various district attorney offices throughout the country. I have provided consultative services to pharmaceutical companies regarding vaccine safety and have provided an expert deposition on this topic in one case. As listed in my Curriculum vitae, I have received numerous national and international honors and awards in the area of clinical toxicology and have lectured both nationally and internationally on mercury and other topics in the field of clinical toxicology.

A more detailed recitation of my experience and qualifications can be found in my Curriculum vitae, which is attached to this report as Exhibit H.

**SUMMARY OF OPINIONS**

I have been asked to render an opinion regarding the existence of a causal link between the administration of thimerosal-containing vaccines to infants and children and the subsequent development of either autism or mercury toxicity.
In addition, I have been asked to review the medical records of the petitioners and to express an opinion regarding the claim that their receipt of thimerosal-containing vaccines contributed in any way to their abnormal development.

After a thorough and careful review of the existing body of scientific literature bearing on the subject and the application of the basic principles of toxicology, I have unambiguously concluded that there is no reliable scientific evidence to support the claim that exposure to thimerosal-containing vaccines administered in accordance with the recommended childhood immunization schedule is causally associated with the development of either mercury toxicity or autism spectrum disorders (ASD). It is thus my opinion that, to a reasonable degree of medical and scientific probability, thimerosal-containing vaccines are not implicated in the etiology of autism or ASD in general, and did not cause and were not contributory to the petitioners’ conditions.

Based on my review of the relevant, reliable, scientific evidence, and on my knowledge, experience, and training as a medical toxicologist, I have concluded that the dose of mercury to which the petitioners were exposed through their thimerosal-containing vaccines was enormously lower than that which could have resulted in toxicity. The petitioners did not display, and there is no indication that they ever displayed, any of the characteristic signs or symptoms of mercury toxicity. It is therefore my opinion that, to a reasonable degree of medical and scientific probability, the thimerosal in vaccines administered to the petitioners did not lead to mercury toxicity or adversely affect their health and well being in any way.

I have carefully reviewed and considered the medical records and laboratory reports pertaining to the petitioners; the 2001 and 2004 U.S. National Academy of Sciences, Institute of Medicine reports addressing and refuting the hypothesis that exposure to thimerosal-containing vaccines can cause autism or other
neurodevelopmental disorders, transcripts of these proceedings, and related slide presentations; a very large body of peer-reviewed studies of the effects of mercury; and the reports of Drs. Aposhian, Mumper, Deth, and Greenland.

As will be reviewed in this report, the studies relied upon by Dr. Deth bear no scientifically supportable relationship to regressive autism or any form of ASD.

Dr. Aposhian’s report is internally contradictory and fails to provide a full and objective assessment of scientific knowledge on the issue of thimerosal, or mercury in any form, and ASD. His theory of the relationship between thimerosal administration and ASD is based on 6 pillars, 5 dealing with mercury. NONE of the five pillars that deal with mercury are scientifically supportable. These 5 pillars are:

1. That there is increased mercury in baby teeth in autistic children;
2. That some autistics have a hypothesized “mercury efflux disorder,” manifested by deceased hair mercury;
3. That studies involving the administration of chelating agents support a role for mercury in ASD;
4. That chelation therapy is efficacious treatment for ASD; and
5. That autoimmune sensitive mice exposed to mercury develop an autism-like syndrome.

Lastly, the views expressed in this report will be demonstrated to reflect not only my own view, but – unlike those of Drs. Aposhian, Deth, Greenland, and Mumper – are well and broadly accepted in contemporary medical and scientific thought.

**SCIENTIFIC BASIS FOR OPINIONS**

**Mercury poisoning does not manifest as autistic spectrum disorders**
In my practice as a medical toxicologist, I have consulted on, and personally treated, adults and children suffering from mercury poisoning, and I am very familiar with the clinical presentation of mercury toxicity. In addition, I have experience with a number of children with autistic spectrum disorders who have been brought to me for toxicological evaluation and treatment. Based on this clinical experience, I can say with a very high degree of medical probability that ASD and mercury toxicity are two very different and distinct clinical syndromes, each with its own characteristic signs, symptoms, and laboratory findings.

Exposure to toxic doses of ethyl mercury (the form of mercury to which individuals are exposed after receiving a vaccine containing thimerosal) causes characteristic signs and symptoms that include visual field constriction, ataxia (i.e., uncoordination of movement), dysarthria (slurred speech due to poor motor control of speech related muscles), peripheral neuropathy (i.e., abnormal sensation due to damage of peripheral nerves, particularly in the lower extremities), characteristic abnormalities of renal function, and tremor. None of these is characteristic of autism or ASDs. (Nelson and Bauman 2003) While not specializing in the diagnosis of ASDs, I have seen, evaluated, and treated a number of autistic children in my medical toxicology clinical practice, and I am familiar with the characteristic signs and symptoms of this disorder, which include lack of eye contact, impairments in social reciprocity, speech delays, speech/language deficits, and repetitive and stereotypical behaviors. The fact that some autistic children may also exhibit a variety of other non-specific neurological signs and symptoms that occur in any number of disorders, in the population in general, and even in some people who have been exposed to a mercury-containing substance, is neither surprising nor indicative that the child is suffering from something other than an ASD. Given the clear differences between the core signs, symptoms, and laboratory findings of the two disorders (see table below), no informed physician can reasonably confuse mercury poisoning with ASDs, or vice versa.
Ethyl mercury poisoning and ASD are completely different diseases

<table>
<thead>
<tr>
<th>ASD</th>
<th>Ethyl mercury poisoning</th>
</tr>
</thead>
<tbody>
<tr>
<td>Large head size</td>
<td>Small head size</td>
</tr>
<tr>
<td>Lack of social reciprocity</td>
<td>No lack of reciprocity</td>
</tr>
<tr>
<td>Stereotypical behavior</td>
<td>No stereotypical behavior</td>
</tr>
<tr>
<td>Purkinje cells affected</td>
<td>Granular cells affected</td>
</tr>
<tr>
<td>Normal renal function</td>
<td>Abnormal renal function</td>
</tr>
<tr>
<td>No tunnel vision</td>
<td>Tunnel vision</td>
</tr>
<tr>
<td>No tremor</td>
<td>Tremor</td>
</tr>
<tr>
<td>No paresthesias</td>
<td>Paresthesias</td>
</tr>
<tr>
<td>No ataxia</td>
<td>Ataxia</td>
</tr>
<tr>
<td>No dysarthria</td>
<td>Dysarthria</td>
</tr>
<tr>
<td>Associated with relatively normal blood mercury levels (normal approx. &lt; 10 ug/L)</td>
<td>Associated with high blood mercury levels (&gt; 500 ug/L)</td>
</tr>
</tbody>
</table>

I have reviewed the article by Bernard et al. (2001), reporting to find substantial similarities and overlap between signs and symptoms of mercury poisoning and those of ASDs and concluding, therefore, that autism is a "novel" form of mercury poisoning. This conclusion is wholly insupportable.

In making their comparison between the two outcomes, Bernard et al. apparently reviewed the entire body of published literature addressing the issues of mercury exposure and ASDs and extracted from these articles every sign or symptom that has ever been reported by anyone who was exposed to mercury in any form and in any dose, and every sign or symptom ever reported to have been exhibited by anyone with ASD. Such an approach to the issue is wholly unscientific and cannot support the authors' ultimate conclusion. Moreover, most of the symptoms of mercury toxicity used by Bernard et al. are from cases of exposure to elemental or methylmercury, not ethylmercury. The differences between these two compounds and their respective potential toxic effects on the body are discussed below.
In addition, a number of articles referenced by Bernard et al. do not support their characterizations of the symptoms reported. For example, for “social deficits, shyness, social withdrawal” Bernard et al. cites Vroom and Greer (1972), which is a case series of mine workers exposed to elemental mercury vapor; no symptoms of social deficits, shyness, or withdrawal are documented. Most problematic, however, is that the “traits of mercurialism” identified by the authors go far beyond what is generally accepted as being the characteristic signs and symptoms of mercury poisoning. As documented in the medical literature, mostly in the form of case reports and case series, a person with mercury poisoning may exhibit a variety of signs and symptoms, whether related to mercury exposure or not, that are neither characteristic of, nor specific to, a diagnosis of mercury poisoning.\(^1\) Despite this variation, however, virtually all patients with mercury toxicity present with some, or all, of the core set of signs and symptoms set forth above.

Bernard et al.’s claim of overlapping or similar signs and symptoms in both mercury poisoning and ASD is blatantly incorrect – an observation that has been made independently by investigators from the U.S. National Institute of Neurological Diseases and Stroke and the Children’s Neurology Service at Harvard Medical School. (Nelson and Bauman 2003) The clinical characteristics of mercury poisoning are so different from ASDs that it is inconceivable that any competent physician could confuse the two.

\(^1\) Case reports and case series are simply published reports of a physician’s clinical experience in the treatment of a single patient or a series of individual patients. Because of the constraints inherent in the way in which they arise, case reports and case series are not controlled in any way. Nor are they studies that can test a causation hypothesis because there is no basis on which to distinguish whether the observed outcomes/symptoms are attributable to the background rate of occurrence in the general population, the exposure in question, and underlying disease or disorder, or concomitant medication. It is thus generally accepted in the medical and scientific communities that case reports and case series are anecdotal and do not constitute proof of a hypothetical causal relationship between an exposure and a subsequent adverse effect.
The petitioners do not have any known or accepted form of mercury toxicity

As noted in this report, there is a characteristic constellation of signs, symptoms, and laboratory findings that are used in the diagnosis of ethyl mercury poisoning. My review of petitioners' medical records revealed no evidence of mercury poisoning.

It is thus my opinion as a medical toxicologist with a great deal of experience in mercury poisoning that, to a reasonable degree of medical probability, the petitioners are not, and were not at any time, suffering from mercury toxicity and their exposure to thimerosal-containing vaccines did not cause or contribute to their current medical conditions.

SCIENTIFIC METHODOLOGY IN THE ASSESSMENT OF CAUSATION

As a medical toxicologist, one of the things that I am repeatedly required to do is to apply basic scientific and epidemiologic principles to the assessment of whether a medication or other chemical substance is capable of causing a particular adverse outcome. It is not until this fundamental question has been answered in the positive that it is scientifically or medically appropriate to move to the next step and to ask whether the agent in question actually caused this outcome in any particular patient.

In applying such principles, the first step in assessing whether the patient's presenting signs or symptoms were caused by an exposure to a particular chemical is to determine if there is sufficient evidence to allow me to assess if that substance is capable of causing the outcome in question, and if so, whether it is likely to have done so at the dose and circumstances to which the patient was exposed. The latter is a reflection of the fundamental toxicological concept
embodied in the adage: “The dose makes the poison.” At the heart of this principle is the recognition that every chemical substance – including those that are vital to the substance of life and those that we may come in contact with on a daily basis such as water or air – can be toxic. What makes any substance toxic, benign, or beneficial is the dose that is delivered to the site of action. For example, low to moderate doses of the chemical N-(4-hydroxyphenyl) acetamide – commonly known as acetaminophen (Tylenol®) – is an effective and safe pain reliever; however, at high doses, it is very toxic to the liver. It is therefore grossly inappropriate and misleading to say, categorically, that any substance is “toxic” per se and to conclude that exposure to that substance at any dose whatsoever can result in toxicity.

Dr. Aposhian, petitioners’ toxicology expert, eschews the fundamental toxicological concept of dose in his views on causation. This is apparent in his report and in his previous testimony, which was so demonstrably at odds with basic toxicology that I took particular note of it. In a radical departure from currently accepted scientific thought, when asked about the fundamental concept that substances are toxic or non-toxic based upon the dose, plaintiffs’ toxicology expert, Dr. Aposhian, testified in Cedillo that “This is an ancient form of quotation that until recently we taught in medical schools, and in undergraduate school, and in graduate school” (Cedillo transcript, p. 129). He further testified: “We no longer believe that the dose determines the poison” (Cedillo transcript, p. 129). When questioned further about this very unconventional position, he further stated “I don’t agree that only dose makes a poison. I mean, that is an antiquated belief today. If I had a graduate student here answering your question he would laugh. He would laugh because students are more up to date than many of us” (Cedillo transcript, p. 130).
All standard toxicology textbooks of which I am aware embrace the fundamental
significance of dose, and I know of none that express the sentiment embodied in
the above quotation from Dr. Aposhian's testimony.²

Speculating about the existence of a "hypersusceptible" population, as Dr.
Aposhian and other plaintiffs' experts do, allows the speculator to evoke this
explanation for virtually any medical phenomenon that cannot be explained by
true science. One can speculate, for example, that a person's stroke was due to
the fact that three days ago they drank a Coca Cola®. When confronted with the
inevitable, rational, and scientific skepticism pointing out that there is no
evidence in human experience that would even suggest that there may be a
causal relationship between having a stroke and drinking a Coca Cola®, nor is it
even plausible, the proponent of the causal view need only to say that the
answer is simple - "this person was not like everybody else, they were
hypersusceptible." Such a blind acceptance of unsupportable explanations based
on "hypersusceptibilities" would allow individuals to evoke a causal nexus

² To list every statement in every toxicology text demonstrating the lack of acceptance of Dr. Aposhian's
view of dose would be exhaustive both to the reader and the writer. However, I will quote from several
salient examples.

Casarett and Doull's Toxicology- The Basic Science of Poisons (5th Ed) is the most widely read and sold
toxicology text in the Western world. In the very first chapter (History and Scope of Toxicology) it states
"The mid-1950s witnessed the strengthening of the U.S. Food and Drug Administration's commitment to
toxicology under the guidance of Arnold Lehman. Lehman's tutelage and influence are still felt today.
The adage "You too can be a toxicologist" is as important a summation of toxicology as the often quoted
statement of Paracelsus: "The dose makes the poison." (p.9)

In the second chapter of Casarett and Doull (Principles of Toxicology), this sentiment is once again
emphasized. "The characteristics of exposure and the spectrum of effects come together in a correlative
relationship customarily referred to as the dose-response relationship. This relationship is the most
fundamental and pervasive concept in toxicology. Indeed, an understanding of this relationship is essential
for the study of toxic materials." (p.18)

In another particularly relevant example, Dr. I. Glenn Sipes, the Chairman of the Department of
Pharmacology at the University of Arizona, a department in which Dr. Aposhian tells us he is a Professor,
states in Clinical Environmental Health And Toxic Exposures (Sullivan and Krieger Eds., 2nd Ed.) in the
chapter entitled Principles of Toxicology: "One of the most important concepts in toxicology is the dose-
response relationship. The underlying premise is that any compound can be toxic if it is encountered in
large enough doses." (p. 50). It should be noted that Dr. Sullivan, the Senior Editor of this book, is the
Associate Dean for Clinical Affairs at Dr. Aposhian's own institution.
between any two random and unrelated events. This includes receiving a vaccination and developing autism.

Where hypersusceptible populations are known to exist, they are very well characterized and subject to formal scientific scrutiny before acceptance. For example, there are known and well characterized populations that are known to be susceptible to sun-induced skin cancers based on an enzymatic defect. Another example is that children with iron deficiency are known to be susceptible to lead poisoning because of increased absorption of lead through the gastrointestinal tract. These and many other highly characterized examples of susceptible populations are well accepted within the scientific community. Their characteristics have been well worked out. There is no ambiguity about who fits into these hypersusceptible populations and who does not. The relevant individuals can be identified by objective criteria. . The same cannot be said of the proposed population that is purported to be uniquely susceptible to thimerosal as the causative agent for their autism. This hypothesized population that is hypersusceptible to develop ASD from thimerosal remains conjecture without scientific support.

Neither Dr. Aposhian, nor any of the petitioners’ other experts, can define their so-called hypersusceptible population beyond simply telling us that if a child has been vaccinated and developed ASD, the ASD must have been vaccine-caused because the patient was “hypersusceptible” to the vaccination. They tell us no uniquely defining characteristic of this so-called definable population that would allow us to test whether this implausible hypothesis has any weight. Rather, Dr. Aposhian and Deth list multiple characteristics which they speculate may make a child more vulnerable to thimerosal-induced ASD. Any one of these characteristics could be easily evaluated by controlled human studies. However, neither Dr. Aposhian nor Deth point to a single controlled study showing that any of their hypothesized characteristics make a child vulnerable to ASD from
thimerosal-containing vaccines. The reason for this failure to reference such studies is clear – there are none.

The fundamental principles described above and their application to the questions presented in this case – i.e., whether exposure to thimerosal-containing vaccines can contribute to autism and whether such exposure actually was a contributing cause of the petitioners’ conditions – are discussed below. Although the toxicology of mercury is quite complex, this discussion will focus primarily on mercury exposure from vaccines, namely that from thimerosal or its metabolic productethylmercury.3

**In vitro studies do not provide sufficient scientific basis to conclude that a particular substance is a potential cause of any specific toxic effect in humans**

An *in vitro* study is one that is carried out in isolation from the complex environment of the intact living organism. Thus, this kind of study consists of experiments performed on cells or tissue grown in a petri dish, in an artificial culture medium, or on a cell or tissue homogenate – i.e., a slurry produced by mechanical disruption (e.g., grinding) that destroys the cell/tissue membrane structure. Such studies are not sufficient to reach conclusions regarding the toxicity in the human body. However, they can be useful for generating specific hypotheses to be tested or for studying potential mechanisms by which an established cause and effect relationship might occur.

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3 Different forms of mercury (Hg), including elemental (Hg⁰) ionic (e.g., Hg²⁺), and organic (e.g., dimethyl mercury, methyl mercury, and ethyl mercury) have different toxicological properties and affect different organs. For example, at sufficient doses, exposure to mercury vapor (Hg⁰) affects both kidneys and the central nervous system, while toxic exposures to ionic mercury/mercuric salts (e.g., Hg²⁺) primarily causes kidney damage. Even within these broad classes, there are toxicological differences between specific substances. The primary target organ for the organic mercurial methyl mercury is the brain (i.e., the central nervous system) while the chemically similar ethyl mercury can have, at sufficient dose, both renal- and neurotoxic consequences. Thimerosal is the sodium salt of ethyl mercury thiosalicylic acid. In the body, thimerosal is rapidly metabolized to ethyl mercury. (Clarkson 2002)
Before a causal relationship between an exposure and a particular outcome can be inferred, there must first be reliable scientific evidence of an association between exposure at the dose of concern and the particular outcome of interest. In the absence of an established association, discussion of a biological mechanism is a non sequitur. In vitro studies have multiple inherent limitations that severely diminish their utility in the assessment of whether a causal relationship exists between human exposure to a substance tested and the subsequent development of a clinical condition. If what happens in a petri dish were, in fact, predictive of a human response to the tested substance, then there would be no need for clinical trials or epidemiologic studies to establish the efficacy or deleterious effects of drugs or other substances. Clearly, this is not the case.

Due to the artificiality of the in vitro environment, the response of tissue or cells to an exposure in vitro will very often differ from the response in the intact organism. For example, when one is investigating the potential effect of a substance on brain structure and/or function, it is of limited utility to know that the substance investigated will kill petri dish-cultured neurons at a particular concentration, unless you know the dose that must be administered to a human before the toxic concentration can be achieved at the neurons in the brain. There is no substance known that will not kill neurons or any other cells in vitro at sufficient concentrations. This includes such benign chemicals as water and oxygen. Moreover, most tissue cultures are composed of a single cell type, perhaps with some impurities; as such, the reductionist environment of tissue culture negates the “cross-talk” and homeostatic control that is of fundamental importance in vivo. As an example of “cross-talk”, in the brain there are neurons and cells known as glial cells that act as nutritional and biochemical support cells for the neurons. Astrocytes are a type of glial cell that take up and inactivates mercury. In fact, a recent study demonstrates that the addition of astrocytes to
neuronal cell cultures protects against methyl mercury-induced neurotoxicity. (Morken, 2005) Thus, just having neurons alone in culture – for example in the studies relied upon by Dr. Deth – creates a markedly unnatural condition that renders the neurons to be unusually vulnerable.

In addition to this difference in effective dose, the form in which the substance is present \textit{in vivo} (in the intact living organism) may be markedly different from that which exists \textit{in vitro}. For example, \textit{in vivo}, many substances (including mercury compounds) are bound to extracellular proteins and other molecules and thus cannot be taken up by target cells. If these binding substances are not included in the \textit{in vitro} incubation mixture, and they usually are not, the \textit{in vivo} exposure will not be replicated. One important example of this concept is that mercurials are bound and inactivated by a protector molecule known as metallothionein. (Leiv/a-Presa, 2004) If the appropriate concentrations of metallothionein are not added in \textit{in vitro} experiments, cells will be artificially rendered extremely vulnerable. Yet, metallothionein is virtually never included in these kinds of studies.

Cells are further protected from potentially toxic substances by a number of other biochemical and physiological defense mechanisms. These natural detoxification processes are an essential part of how our bodies deal with many potentially toxic substances to which we are exposed on a daily basis. Because \textit{in vitro} models cannot duplicate, and do not account for, these complex and highly efficient defense systems, cell responses \textit{in vitro} may be markedly exaggerated and thus inaccurate representations for what actually occurs \textit{in vivo}.

Because of all of these considerations, the U.S. National Academy of Sciences, Institute of Medicine, Immunization Safety Review: Vaccines and Autism concluded: “Demonstrating an adverse effect of mercury \textit{in vitro} does not readily translate into a physiologic argument.” (2004 IOM Report, p. 140)
The in vitro studies of Dr. Deth

Dr. Deth bases most of his opinions on a 2004 in vitro study done in his laboratory. (Waly et al., 2004) This study assessed the effects of thimerosal and other substances on the activity of an enzyme known as methionine synthase from cells known as SH-SY5Y cells. There were no other cell types (e.g., astrocytes) present, and the medium in which the incubation occurred was simply a standard incubation medium with antibiotics added. The latter are typically added to prevent bacterial growth of the cultures. SH-SY5Y cells are derived from cells that were initially removed from a metastatic bone tumor in 1970 and have been genetically manipulated since. They are chromosomally abnormal and clearly have little similarity to human neurons. Importantly, the very enzyme studied in this experiment, methionine synthase, is defective in this particular line. (Deth, 2006) Thus, the results from this cell line cannot be generalized. Despite this, most of the theories put forth by Deth in his report in this matter are based on the results of his 2004 study with this experimental model.

The particular experiment done by Dr. Deth was to assess the activity of methionine synthase from SH-SY5Y cells. Data from the Waly (and Deth) 2004 publication purported to show that thimerosal blocks methionine synthase activity. However, this only occurs in an artificial copper-free environment. When copper was added back to the reaction mixture at a concentration of 1 micromolar, there was little to no inhibition of methionine synthase until concentrations of thimerosal were so high as to have no possible relevance to any exposure of neurons after thimerosal-containing vaccine. The importance of this is highlighted by the fact that the normal intracellular concentration of copper is in the range of 15 micromolar! (Merk Manual, 2008) These various
issues contribute to the irrelevance of the Waly data to anything that could be reasonably expected to occur physiologically.

Waly and Deth then speculate that if the results from their *in vitro* experiment were generalizable to individuals with autism. These results would affect a process known as DNA methylation. In his report Dr. Deth tells us that abnormal patterns of DNA methylation are, in his opinion, “an important contributor to developmental disorders.” Yet he cites no data supporting abnormal DNA methylation as a cause of autism. The two references he does cite for this proposition deal with Prader-Willi, Angelman, and Rett’s syndrome.

In the Waly 2004 paper, Dr. Deth supplies data on DNA methylation in SH5-SY5Y cells. Curiously, although they report the effects of several substances on DNA methylation, there are no data provided for thimerosal or ethyl mercury on this process. This is baffling since the focus of their paper was on thimerosal.

To explain the gaps with his theory of causation, Dr. Deth, like Dr. Aposhian, relies upon speculation involving "genetic vulnerability." These discussions are, however, mere conjecture, and Dr. Deth cannot identify the specific genetic profile that can be used access the validity of his theory. Instead, he speculates about several genes that might be involved in the hypersusceptible population. Without reliable scientific evidence to support these claims, the only distinguishing criteria of this subpopulation is the fact that they developed autism after receiving a thimerosal-containing vaccination. As the 2004 National Academy of Sciences, Institute of Medicine Immunization Safety Review – Vaccines and Autism concluded when discussing the Waly (and Deth) 2004 study (p. 137): “The authors hypothesize that disruption of this pathway [methionine synthase] by thimerosal leads to autism, ADHD, and other developmental disorders. However, the committee is aware of no evidence that autism is caused by alterations in this biochemical pathway. In addition, the evidence that
several important toxicants disrupt this pathway and that it is involved in many physiological effects weakens the argument that thimerosal might cause autism through this mechanism.”

**IT IS NOT SCIENTIFICALLY VALID TO DRAW CONCLUSIONS REGARDING THE TOXICITY OF ETHYL MERCURY OR THIMEROSAL FROM DATA ON THE TOXICITY OF METHYL MERCURY OR ANY OTHER FORM OF MERCURY**

It is not scientifically justifiable to assume that the toxicological properties of any specific compound are the same as those of a different compound, even if the two are chemically similar. For example, methyl alcohol (methanol, or wood alcohol) and ethyl alcohol (ethanol, or the alcohol in spirits), which are chemically very similar, have dramatically different toxicological properties. At moderate to high doses, ethanol, the alcohol in spirits, causes acute alcohol intoxication (i.e., drunkenness), but very low doses of methanol can cause acute multisystem dysfunction, often death, and in survivors, blindness.

The minor difference in chemical structure between methanol and ethanol is precisely the same as the chemical difference between methyl mercury and ethyl mercury. There are many studies that show that, in the body, methyl mercury and ethyl mercury behave very differently (e.g. Magos et al., 1985; Harry et al., 2004; Burbacher et al., 2005).

The recognition that that which is applicable to one form of mercury does not necessarily apply to other forms is well accepted and broadly articulated in the medical and scientific literature. For example, in Casarett and Doull’s widely read book *Toxicology – The Basic Science of Poisons* (2001; Klaassen, Ed.) it is stated that: “No other metal better illustrates the diversity of effects caused by different chemical species than does mercury” (p. 834). Similarly, the United States
Centers for Disease Control and Prevention (CDC) makes this point several times. For example, the Agency for Toxic Substances and Disease Registry (ATSDR), a component of the CDC, states in its toxicological profile on mercury (1999): “Exposure to mercury however does not necessarily mean that adverse health effects will result. Health effects depend on the amount of exposure, the form of mercury, and the route of exposure.” (Italics added).

Dr. Aposhian recognizes the differences between ethyl mercury, methyl mercury, and inorganic mercury in his report related to this case. For example, beginning on page 15 and continuing to page 17, Dr. Aposhian discusses the different forms of mercury. In much of his report he discusses the toxicological issues regarding methyl mercury. This is similar to the presentation that he gave to the Court on the *Cedillo* issue, which relied heavily on methyl mercury studies. For example, in *Cedillo*, Dr. Aposhian testified “The chemical forms of species of mercury are different chemically and have different toxicological properties.” (p. 114); he also testified “We don’t like to use the term mercury without specifying what form of mercury we’re talking about.” (p. 128); and yet again on page 127 he agreed with the statement about the diversity of effects of different species of mercury from Casarett and Doull’s book described above. In addition, on pp. 143-144 there is a dialogue concerning mercury in which Dr. Aposhian stated that “everyone knows” the different forms of mercury have different toxicological properties. It should be noted that petitioners have not implicated inorganic or methyl mercury in the causation of autism.

The effects of methyl mercury at doses typically encountered in the United States appear to have no adverse effect on childhood development. Methyl mercury exposure in humans is primarily through consumption of seafood. A recent publication by the Avon Longitudinal Study of Parents and Children found that mothers with the highest seafood intake (>340 grams/week which is the current EPA and FDA recommended limit) had children with better IQ, prosocial
behavior, social development, and communication than did children of mothers who ate lower amounts of seafood during pregnancy. (Hibbeln, 2007) Similarly, as with thimerosal/ethyl mercury, it is clear that there is no relationship between methyl mercury intake and autism. An epidemiological investigation from the Faroe Islands, where methyl mercury intake is far in excess of that in the United States, found a prevalence of autism of approximately 0.56%, which is not higher than that which is seen, for example, in the United States. (Ellefson, 2007) Similarly, children in the Seychelles have much higher methyl mercury exposure through seafood than do U.S. children. Despite the intensity with which the Seychelles population has been studied, there have been no reports of increased rates of autism in that population.

In his discussion of the data from the Faroe Island studies and the Seychelles Child Development Study, Dr. Aposhian profoundly misrepresents the data from the Seychelles. On page 23 of his report, Dr. Aposhian tells us that: “Once some method changes were made, intelligence deficits were also found subsequently in some Seychelles Island children (Myers et al., 2003).” This is entirely incorrect. The Myers publication was a report of the results of the Seychelles study assessing the effects of prenatal methyl mercury exposure on the cohort that had been followed, at that time, to age nine. That publication reported no adverse effects related to prenatal methyl mercury exposure.

Dr. Aposhian also points to the study by Hightower (2003) as evidence of adverse effects of methyl mercury. However, as with his description of the Myers data, his report grossly mischaracterizes the Hightower study. He describes the patients as “a group of wealthy persons (CEOs, attorneys, and physicians) who presented to their physicians with central nervous system complaints. On page 605 of the Hightower report, they describe the individuals studied; while there were attorneys and physicians noted, there were also individuals with many other affluent and less affluent positions, including retirees
and homemakers. There were no CEOs. The group of patients was not individuals “who presented to their physicians with central nervous system complaints.” Rather, the group consisted of individuals who ate a significant amount of seafood or had a variety of non-specific symptoms. Dr. Aposhian indicates that they “ate expensive fish, such as shark, swordfish, and tuna, almost exclusively as their protein source.” This too is not true. Some of the individuals were heavy seafood eaters, but certainly there is no indication in the paper that this was their almost exclusive protein source. Further, the most common form of seafood that was eaten was canned tuna fish, which 78% of the individuals consumed. Dr. Aposhian tells us that after being on a seafood-free diet “the patients returned to normal health.” However, no health assessments were done in this study. The only parameter that was assessed while being on a seafood-free diet was blood mercury concentrations.

A more relevant study, that actually did assess neurological endpoints, was the Baltimore Memory Study, which was a longitudinal study of cognitive function involving over 1,100 individuals between 50-70 years old. That study measured total blood mercury concentrations as a surrogate marker of methyl mercury exposure from seafood and assessed 20 neurobehavioral scores from 12 separate neurocognitive tests. That study concluded: “Overall, the data do not provide strong evidence that blood mercury levels are associated with worse neurobehavioral performance in this population of older urban adults.” (Weil 2005)

**The reference dose for methyl mercury provides no support for the assertion that thimerosal from vaccines can cause autism**

Petitioners’ hypothesis that post-natal exposure to ethyl mercury from thimerosal-containing vaccines can cause ASD in children is based, in part, on the proposition that an infant who was vaccinated in accordance with the
recommended childhood immunization schedule could be exposed to quantities of ethyl mercury that exceed the U.S. Environmental Protection Agency’s (EPA’s) reference dose (RfD) for methyl mercury.\textsuperscript{4}

The EPA RfD of 0.1 ug/kg/day is not a threshold exposure dose above which toxicity is reasonably likely to occur. Rather, it is an estimate of the average daily oral exposure that is likely to be without an appreciable risk of adverse effects if continued over a person’s lifetime. In calculating the RfD, the EPA chose as its starting point the level of \textit{in utero} exposure to methylmercury associated with a predefined level of risk of subtle, sub-clinical, neurodevelopmental deficits;\textsuperscript{5} ASD was not an endpoint in this study from which the RfD was derived. The EPA’s recommended safe intake level is thus

\textsuperscript{4}Like the EPA, the FDA, the ATSDR, and the World Health Organization (WHO) have all issued safety guidelines for methylmercury consumption. Exposure to mercury (in the form of ethylmercury) from thimerosal-containing vaccines does not exceed any of these other guidelines – with a single exception of an infant in the 5\textsuperscript{th} percentile for body weight, whose cumulative mercury dose would exceed the ATSDR guidelines (Ball et al., 2001).

\textsuperscript{5}The RfD for methylmercury was derived from data gathered during the course of a long-term study on the risk of harm posed by \textit{in utero} exposure to methylmercury via mother’s regular consumption of contaminated seafood and episodic consumption of whale meat, which is known to have a very high methylmercury content. In this Faroe Island Study, such prenatal methylmercury exposure was reported to be associated with subtle, sub-clinical, deficits in the area of learning, memory, and language, as measured by various neuropsychological tests. Using the Faroe Island data (Rice et al., 2000), EPA determined the concentration of mercury in cord blood that, for each test, was associated with the doubling of the number of children with a response at the 5\textsuperscript{th} percentile. This is known as the Benchmark Dose. The lower limit of the 95\textsuperscript{th} confidence interval surrounding this Benchmark Dose – known as the Benchmark Dose Lower Limit (BMDL) was then chosen as a so-called point of departure dose. Then, to account for the fact that some unrecognized adverse event might occur at a dose lower than the BMDL, and recognizing that, due to differences in pharmacokinetics (e.g., impaired mercury elimination) and pharmacodynamics, some people in the population might be more susceptible than others to the adverse effects of mercury, the BMDL of 58 parts per billion (ppb) were divided by a factor of 10. Thus, the level of continuous mercury exposure considered safe to the unborn fetus was determined to be 5.8 ppb (which is the same as 5.8 ug/L). It was then determined that to reach this steady state concentration of mercury in the umbilical blood, a pregnant woman would have to consume an average of 0.1 ug/Hg/kg body weight/day and throughout her pregnancy. This calculation is based, in part, on the half-life of mercury in the blood following dietary methylmercury exposure. It should be noted that in a similar study in another similarly exposed fish-eating population (Seychelles Island) no neurodevelopmental effects were found following fetal exposure to methylmercury (Myers et al., 2003; Davidson et al., 1998). While it has been suggested that the difference between the two studies might be attributed to the so-called “bolus dose” exposure to the fetus as a result of the mother’s periodic consumption of whale meat, which contained extremely high levels of methylmercury and resulted in repeated spikes in exposure, the Faroe Island investigators tested this hypothesis and found no support for such a bolus effect (Grandjean et al., 2003).
inherently designed to protect those who are arguably the most sensitive to methylmercury — developing fetuses. In addition, once the dose that was observed to create this increased risk of harm was determined, the dose at the lower limit of the 95% confidence interval around that dose was further reduced by a factor of 10 to account for the possibility that some people might be even more susceptible to methylmercury neurotoxicity and to account for the possibility that other unrecognized effects might occur at a lower dose. The RfD, which was calculated to protect sensitive populations from the first instance, thus incorporates an additional substantial margin of safety for both childhood and adult exposures to methylmercury. There is thus a substantial difference from a presumptively safe daily dose described by the RfD and the dose believed to give rise to toxicity.

Moreover, because the RfD is expressed as the average daily exposure that is presumptively safe if continued over a lifetime, short-term exceedances of the RfD are anticipated and are not expected to result in adverse consequences; the key is that the RfD defines a chronic exposure that results in a steady state blood level. If the RfD were a toxicity threshold, then one would expect that a child’s or nursing mother’s periodic consumption of fish or seafood would give rise to neurotoxicity in the child. This is clearly not the case. Consequently, even when considering exposures to methylmercury, the fact that the RfD is exceeded for a brief period is not indicative of toxicity.

Furthermore, in establishing the RfD, EPA evaluated only dietary exposure; i.e., oral exposure through the ingestion of contaminated fish and seafood. As demonstrated in a recent study conducted by the National Institute of Environmental Health Sciences (Harry et al., 2004), the concentration of mercury in the blood and brain following oral exposure to methylmercury is considerably higher than the concentrations resulting from intramuscular exposure to either methylmercury or thimerosal. Given this difference in absorption and distribution
between oral and intramuscular exposures, it is reasonable to conclude that the RfD actually overestimates the risk potentially associated with intramuscular exposure. In other words, if an RfD were to be calculated for intramuscular exposure to methylmercury, it would likely be higher than the present RfD for dietary exposure.

In addition to the fact that the RfD was derived from data generated as a result of a route of exposure (ingestion) that was not at issue with the administration of thimerosal-containing vaccines (intramuscular), the RfD is also reflective of continuous, long-term exposure to methylmercury and essentially steady state blood concentrations. Vaccines, on the other hand, are administered approximately once every two months over a six-month period, and then again, approximately one year later. Therefore, because the ethylmercury from episodic vaccinations is rapidly eliminated, the exposure is not continuous nor is it cumulative. Thus, the cumulative dose of fetal exposure would be significantly higher than an infant’s post-natal exposure due to bimonthly inoculation with thimerosal-containing vaccines. This difference in dose between cumulative, chronic exposure to methylmercury from seafood in the Faroese Cohort and the infrequent ethylmercury exposures from vaccines renders the comparison between the two situations meaningless. Clearly there is a much greater dose of methylmercury from seafood in the Faroese Cohort than there is in ethylmercury/thimerosal from the vaccination of children.

Second, even if the RfD could be viewed as a toxicity threshold for intramuscular administration of methylmercury, application of that threshold to thimerosal/ethylmercury exposures would be valid only if the neurotoxicity of ethylmercury were quantitatively the same as that of methylmercury. It is not.

Animal studies have shown, for example, that with equivalent exposures – *i.e.*, equal doses of mercury delivered by the same route of administration –
thimerosal/ethylmercury-exposed animals have less mercury in their brain than those exposed to methylmercury (Magos et al., 1985; Harry et al., 2004). It has further been demonstrated that mercury clears from both blood and brain much more quickly following intramuscular thimerosal exposure than it does following dietary methylmercury exposure (Pichichero et al., 2002; Suzuki et al., 1973; Burbacher et al., 2005). Because the neurotoxic potential of any substance will depend on how much of the substance reaches the brain in toxic form, these studies indicate that thimerosal/ethylmercury is quantitatively less neurotoxic than methylmercury.

The study by Burbacher et al. (2005) was designed to evaluate whether the RfD for methylmercury provides an accurate assessment to the risk of children from administration of thimerosal-containing vaccines. In this study, newborn macaque monkeys were administered equivalent doses of thimerosal-containing vaccines via intramuscular injection and methylmercury via dietary exposure on a schedule designed to mimic the childhood vaccination schedule. Each dose was 20 ug Hg/kg body weight, which is substantially higher than the dose administered at any point in the recommended childhood immunization schedule.6

Blood mercury levels were measured 2, 7, and 14 days after the initial dose; 7 and 14 days after each subsequent dose; and 28 days after administration of the last dose. Brain mercury levels were measured only during the 28-day period after administration of the last dose.

The results of this investigation demonstrate that in macaque monkeys: 1. the half life of mercury in the blood is nearly 3 times shorter following the intramuscular thimerosal exposure than it is following dietary exposure to

6 The dose was chosen for technical experimental reasons and was not meant to reproduce the dose received by children at the time of vaccination. (Sager, oral presentation, February 9, 2004)
methylmercury;\(^7\) 2. although initial blood mercury levels were approximately the same in the two treatment groups following the first dose, mercury accumulated in blood between successive doses of methylmercury, but not between successive doses of thimerosal-containing vaccines, so that at the end of the dosing period, the thimerosal-treated group had blood mercury levels that were approximately 1/4 the level observed in the methylmercury-treated group; 3. brain levels of mercury were approximately 3 times higher following dietary methylmercury exposure, as compared to intramuscular exposure to thimerosal-containing vaccines; and 4. mercury cleared from the brain approximately 2 times faster in the thimerosal-treated monkeys.

One observation in the Burbacher et al. study emphasized by Dr. Aposhian in his report is that the rate of demethylation of mercury in the brain is faster for ethyl than for methylmercury. However, this is of no known toxicologic consequence. It is well known that all humans and animals accumulate small amounts of mercury in the brain that increase over time. This can come from multiple sources including diet, infant breast-feeding, and, because mercury is present in the air we breath, simply the act of breathing. Because of this natural mercury accumulation, organs of the body, including the brain, have multiple well-developed and highly effective protective mechanisms to detoxify and inactivate mercury, rendering it biologically innocuous. (Clarkson and Magos, 2006) It is only under circumstances of excessive exposure, where the natural detoxifying systems are overwhelmed, that adverse effects from mercury accumulation may occur. This occurs only at doses of mercury vastly exceeding those that would be associated with vaccination. For example, the cumulative dose of mercury to infants from vaccinations in approximately half as much as that received from breast-feeding. (Marques et al, 2007)

\(^7\) This short blood half-life is consistent with the findings of Pichichero et al. (2002), who calculated a blood mercury half-life of approximately 7 days in infants administered thimerosal-containing vaccines. This is in contrast to the 50-70 day half-life associated with dietary exposure to methyl mercury in humans.
Burbacher et al. concluded, based on these differences in the pharmacokinetic properties of oral methylmercury administration and intramuscular thimerosal administration, that applying the RfD for methylmercury would overestimate the potential for toxicity following the administration of thimerosal-containing vaccines (Burbacher et al., 2005).

For all the reasons stated above, it is not scientifically valid to use the methylmercury RfD as the basis for predicting the potential neurotoxicity of the ethylmercury exposure resulting from the administration of thimerosal-containing vaccines. In fact, the demonstrated differences in the route of administration and the relative pharmacokinetic properties of the two compounds indicate that ethylmercury is a less potent neurotoxin than methylmercury; the safe intake level for ethylmercury would be expected to be higher than the current methylmercury RfD. Even if the RfD for methylmercury were applicable for thimerosal/ethylmercury, exceeding this dose, either cumulatively or on any given day, would not be considered a risk factor for any of the subtle neurological endpoints considered in the Faroe Island study and certainly not for ASD.

Based on all of the scientific literature reviewed above, it is scientifically unreasonable to claim that thimerosal is toxic at doses that meet or exceed the RfD for methylmercury. What can be said with confidence, however, is that there is no scientific basis for the claim that thimerosal causes mercury toxicity or ASDs at the doses delivered by thimerosal-containing vaccines.

The data from studies on inorganic mercury are both inapplicable and do not support an increased risk of ASD.
While Dr. Aposhian implicates thimerosal exposure as the cause of autism/ASD, he makes the following statement on pages 7 of his report: "The more mercury in the air the greater the incidence of autism (Palmer et al., 2006)."

The Palmer (2006) study is an ecological study that attempted to correlate inorganic mercury release into the environment, based on EPA data, with the prevalence of autism in various counties and school districts, concluding that there is a higher prevalence of autism in urban school districts. They also concluded that the release of inorganic mercury into the air from industrial emissions in these districts fully accounted for the increased rate of ASD seen in Texas. Being an ecological study, Palmer et al. simply looked at population trends and not at individual cases. Because the study is cross-sectional, it assessed quantities of mercury released and autism prevalence at one snapshot in time. Thus, this study quantitates mercury releases after the diagnosis of autism was already made. Therefore, no conclusions can be made about inorganic mercury exposure and the subsequent development of ASD. More importantly, the Palmer study could not correlate exposure (i.e., dose) with release of mercury and fails to account for the important ecological drift that would be expected in families with ASD.

The release of inorganic mercury from point sources in Texas, as reflected in EPA records, bears no relationship to population exposure. Overwhelmingly, the exposure to the population in Texas from airborne mercury sources comes from outside of the United States, particularly Asia. (Lewandowski, 2006) Airborne releases originating in Texas are carried in the normal west to east jet stream and are primarily deposited in areas that are hundreds to thousands of miles east of Texas. (Lewandowski, 2006) Detailed studies on population exposure assessment from point sources of industrial emissions of mercury have shown that these sources do not contribute significantly to ground level mercury concentrations and using such sources will result in significant exposure
misclassification. (Hodgson, 2007) No correlation between point source mercury release, as Palmer et al. have evaluated, and mercury exposure in the population can be reasonably gleaned.

A more likely explanation for the results of the Palmer study is that families with children with ASD may migrate to urban areas where there are more sophisticated special education programs. These urban areas are associated with higher reported levels of mercury release.

Furthermore, it is ironic that Dr. Aposhian relies on this ecological study purporting to show an association between inorganic mercury exposure and the development of autism. Petitioners’ epidemiology expert, Dr. Greenland, dismisses ecological studies as being non-informative. Were ecological studies of such little value, they would not be published; they are published, however, and when well designed are published in the most prestigious medical and scientific journals, such as the New England Journal of Medicine.

**THERE IS RELIABLE SCIENTIFIC EVIDENCE THAT THIMEROSAL IS NOT TOXIC TO HUMANS, INCLUDING INFANTS AND CHILDREN, AT DOSES DELIVERED EITHER INDIVIDUALLY OR CUMULATIVELY, BY THIMEROSAL-CONTAINING VACCINES**

As indicated above, one of the most basic and well-accepted tenets of toxicology is that there are no such things as toxic substances – there are only toxic doses. Therefore, the fact that high dose exposure to a particular substance may lead to toxicity does not imply that toxicity will result in doses that are substantially lower.

As will be discussed in this section, the highest mercury concentration measured in term infants after vaccination in three studies was 8 ug/L. (Stajich, 2000;
Pichichero, 2002 & 2008) This is well within the range of blood concentrations in the general population. There has never been any adverse effect associated with blood mercury concentrations in this range.

The available evidence regarding human exposure to thimerosal indicates that the doses of ethyl mercury administered as a result of routine administration of thimerosal-containing vaccines are orders of magnitude less than those doses reported to be associated with neurotoxicity in either adults or children. Even under conditions where the dose is sufficient to cause neurotoxicity, the outcome is not ASD.

For example, in a study by Haeney et al. (1979), twenty-six patients, including young children, were treated with thimerosal-preserved immunoglobulin administered intramuscularly, typically on a weekly basis, for up to 17 years. The youngest patients received these injections for a minimum of 6 months and up to 4 years. The amount of thimerosal-derived mercury ranged from 4-734 mg (i.e., 4,000-734,000 ug) as compared to the maximum of 187.5 ug delivered by thimerosal-containing vaccines over 6 months, or 237.5 ug over 18 months. Yet, the Haeney et al. investigators reported that not one of their 26 patients exhibited clinical evidence of mercury toxicity.

The literature also contains a report of a 6-week old infant administered intramuscular injections of thimerosal-containing antibiotic who received a minimum of 12,750 ug of mercury (nearly 70 times the typical maximum amount delivered by thimerosal-containing vaccines over six months). This infant showed no signs of mercury toxicity during a 2-month followup. (Axton, 1972)

While there are reports in the peer-reviewed scientific literature of thimerosal-related toxicity in humans, these uniformly involved doses far in excess of those delivered by thimerosal-containing vaccines and in some, the diagnosis or
implication of mercury poisoning was questionable. There has not been a single case report of even high dose methyl mercury or ethyl mercury exposures resulting in ASD.

For example, there is a report in the literature of an 18-month old infant who had been administered a thimerosal solution for treatment of otitis media. (Rohyans, 1984) Over a 1-month period, she received 600,000 ug of mercury which, due to the way in which the thimerosal solution was administered, was ultimately swallowed. This is more than 2,500 times the maximum cumulative exposure of mercury delivered by vaccines over an 18-month period and approximately 3,200 times more than the 6-month vaccination schedule. Signs of neurotoxicity quickly developed; she also developed signs of renal toxicity, which is characteristic of high dose ethyl mercury exposure. Her plasma mercury concentration was 1,630 ug/L. This child’s whole blood mercury level would have been much greater than her plasma level, yet the latter is over 203 times higher than the highest blood mercury level ever observed following administration of thimerosal-containing vaccines. In addition, her exposure continued over a prolonged period of time compared to that of vaccines.

Suzuki (1973) reported on a 13-year old boy with a serious condition known as protein-losing enteropathy in which plasma proteins are lost from the blood.

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8 Because ethyl mercury in the body is primarily found in hemoglobin in red blood cells, plasma mercury concentrations would be expected to be significantly lower than whole blood concentrations.
9 There are three studies in infants that look at blood mercury concentrations following the administration of thimerosal-containing vaccines. The first is Stajich et al. (2000), which measured the level of mercury in blood of 5 full-term and 15 pre-term infants following the administration of a birth dose of Hepatitis B vaccine. The vaccine administered contained 12.5 ug of mercury. The term infants had an average post-vaccination blood mercury concentration of 2.24 ug/L (range: 1.4-2.9); in pre-term infants the mean mercury blood concentration was 7.36 ug/L (range: 1.3-23.6). Pichichero et al. (2002) measured blood mercury concentrations in 2- and 6-month old infants following administration of thimerosal-containing vaccines. The highest mercury concentration in the blood of the 2-month olds was 4.13 ug/L, resulting from a cumulative mercury exposure of 37.5-63.5 ug. In 6-month olds the highest measured blood mercury concentration was 1.39 ug/L. In the latter children, the exposures ranged from 87.5-175.0 ug.

Pichichero (2008) measured blood mercury concentrations in 216 infants (72 newborns, 72 infants at 2 months old, and 72 infants at 6-months old). The highest mercury concentration detected was 8 ug/L in a newborn 12 hours after receiving a dose of Hepatitis B vaccine containing 32.5 ug of mercury.
enter the intestines, and are excreted. The treatment for his illness consisted of frequent plasma transfusions, often using thimerosal-preserved human plasma. During the last 3 months of his life, the child was administered a total of 283,500 ug mercury — over 15,000 times more than a child could receive in 6 months from thimerosal-containing vaccines. Although the authors did not contend that the boy died of mercury poisoning and did not note any signs of neurotoxicity, they did note that his blood mercury concentrations 3-4 days prior to his death was over 7,000 ug/L. This apparently non-neurotoxic blood concentration is 875 times greater than the highest blood level ever observed following administration of thimerosal-containing vaccines to term infants.

**There is no reliable scientific evidence that antibiotic therapy alters mercury levels after receiving thimerosal-containing vaccines**

Dr. Aposhian opined on page 6 of his report that if antibiotics are used with mercury exposure, these can inhibit mercury excretion “and thus potentially increase its toxicity. (Rowland et al., 1984).” The Rowland study was one in which mice were fed methyl mercury; half of whom were treated orally with a mixture of bacitracin, neomycin, and streptomycin. This oral regimen delivered directly to the gastrointestinal tract would be expected to decrease normal intestinal bacteria. In the antibiotic treated mice, there was a decrease in methyl mercury excretion, presumably due to the loss of ability to demethylate methyl mercury to inorganic mercury. The latter would not be reabsorbed from the intestinal tract but rather excreted fecally. Whether such an effect would occur following more conventional antibiotics is unlikely given that contemporary regimens tend not to affect intestinal bacteria nearly as much as the regimen used by Rowland. More importantly, however, is that unlike methyl mercury, ethyl mercury is rapidly and spontaneously broken down to inorganic mercury and hence the influence of any changes to gut flora is likely to be minimal to nil. These profound differences in the rate of spontaneous formation of inorganic
mercury from methyl mercury compared to that from ethyl mercury is yet another example of how data from one of these compounds would not be expected to be applicable to the other.

**Dr. Aposhian’s calculation of brain mercury concentrations after vaccination is misleading and fails to provide all of the relevant data**

In his report, Dr. Aposhian does a calculation based on his synthesis of the data from Pichichero (2002) and Burbacher (2005). Dr. Aposhian multiplied the mean blood mercury concentration post-vaccination of 8.2 nM (1.6 ug/L) in two-month old children by a factor of 3.5 to come out with a brain mercury concentration of 28.7 nM. He then noted that the highest mercury concentration in the two-month old group in Pichichero was 20.6 nM (4.1 ug/L) which, after multiplying by 3.5, led him to conclude that a two-month old infant can have brain mercury concentrations as high as 72.1 nM. The factor 3.5 used by Dr. Aposhian is grossly inaccurate based on the Burbacher study. Burbacher administered sequential doses of thimerosal such that the animals received four doses of 20 ug/kg at 7-day intervals.10 This dosing regimen, unlike that from vaccines, caused accumulation of mercury to progressively higher blood concentrations. This occurred because, unlike the situation with human vaccination, the mercury was administered at such short dosing intervals that at each successive dose there was still a substantial portion of the mercury from the prior dose present, causing higher and higher mercury levels to appear. This is illustrated in the following graph from the Burbacher paper showing blood mercury concentrations from thimerosal injections during the experimental period (the last thimerosal injection was on day 28):

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10 At no point would a child vaccinated in accordance with the recommended schedule ever have been exposed to 20 ug/kg of mercury from receipt of thimerosal-containing vaccines. In addition, while the dosing interval in the Burbacher study was approximately equivalent to the half-life of mercury following thimerosal administration, the time between successive vaccinations in children is more than five half-lives.
In the Burbacher experiment, sequential blood and brain mercury concentrations were determined after the 4 weekly immunizations were administered. That data showed that the brain ethyl mercury concentrations fell more slowly than those in the blood. Thus the ratio of the blood to brain concentration was constantly increasing. This is shown in the diagram below from the Burbacher paper:
The factor of 3.5 used by Dr. Aposhian represents an arbitrary use of a single time point. In reality, the blood brain/mercury concentration is a constantly changing ratio. More importantly, as can be seen in the figure above, brain mercury concentrations decrease over time. Dr. Aposhian then tells us on page 14 that the Pichichero data upon which he bases his calculation is based on normal and not autistic children and: “If there is a subset of autistic children with a mercury efflux disorder, they would be expected to have even greater concentrations of mercury in their tissues.” This is an unsupported statement. Neither Dr. Aposhian nor anyone else knows whether any of the children in this study ultimately had an ASD diagnosis. More importantly, as will be described below, the concept of a mercury efflux disorder is one uniquely used by petitioners’ experts and others advocating a connection between thimerosal exposure and ASD. A “mercury efflux disorder” is not a condition that is accepted in medicine or used outside of these contexts.

Even accepting for the sake of illustration Dr. Aposhian’s calculation of 72.1 nM in the brain, that amount is equivalent to 0.072 μM or 14 ug/kg. In an almost
identical model as the Burbacher one from which Dr. Aposhian derived his data, brain mercury concentrations in untreated animals averaged 55 ug/kg. (Blair, 1975) Clinical mercury neurotoxicity in humans has not been documented to occur at brain mercury concentrations less than 300 ug/kg. (Lapham et al., 1995)

By way of comparison, and not mentioned in Dr. Aposhian’s report, brain mercury concentrations were over 3 times higher in the Burbacher experiment when monkeys were given an oral dose of methyl mercury with equivalent mercury content to the injected thimerosal.

The study of Hornig et al., upon which Drs. Deth and Aposhian rely, has conclusively been shown to be incorrect and non-reproducible

Both Drs. Aposhian and Deth rely on the study by Hornig (2004). In Dr. Aposhian’s report, beginning on page 24, he lays out his six-point theory that he believes justifies his beliefs that exposure to thimerosal-containing vaccines gives rise to ASD. The fifth cornerstone of Dr. Aposhian’s theory is that: “autoimmune disease-sensitive mice exposed to mercury after birth develop enlarged brains and autistic-like symptoms (Hornig et al., 2004).” A close reading of the Hornig study does not support this interpretation. However, a detailed discussion of this is moot because a recent study attempting to reproduce the results of Hornig, using more detailed and sophisticated techniques, could not reproduce most of the Hornig data. (Berman et al. 2007) That study was specifically developed to further study the data presented by Hornig. It concluded (p.2): “Considered together the present results do not indicate pervasive development neurotoxicity following vaccine-level thimerosal injections in SJL mice, and provide little if any support for the hypothesis that thimerosal exposure contributes to the etiology of neurodevelopmental disorders.”
There is no reliable scientific evidence that chelation therapy improves the clinical manifestations of ASD, nor is it plausible.

The fourth of Dr. Aposhian’s points upon which he bases his belief that ethyl mercury or thimerosal are harmful in doses received by vaccination is that “...the most beneficial treatment for autism as reported by the parent’s of autistic children was chelation therapy....” There has never been a published scientific study supporting this nor is such a result plausible. Dr. Aposhian implicates the inorganic mercury formed from ethyl mercury that remains in the brain as causing neurological injury. However he himself has published that chelators, and in particular DMSA, do not remove inorganic mercury from the brain. (Aposhian, 2003).11 Thus it is implausible that chelation could be beneficial in treating autism according to Dr. Aposhian’s theories. It is similarly implausible that chelation could improve neurological function if it were the parent ethyl mercury molecule that caused the neurotoxicity. That molecule is rapidly removed from the brain with a half-life in the Burbacher study of 14 days. (Burbacher, 2005). Thus, there is no residual ethyl mercury in the brain by 10 weeks post vaccination. Therefore, it is implausible that chelation therapy beyond this time frame could be beneficial for removing ethyl mercury.

11 In addition to DMSA, Dr. Aposhian’s paper also studied the effects of DMPS, vitamin C, and glutathione, and concluded that “none of these regiments reduced the mercury content in the brain” (p. 339).
THERE IS NO SCIENTIFIC EVIDENCE OF A SUB-POPULATION OF CHILDREN WHO ARE PARTICULARLY VULNERABLE TO MERCURY TOXICITY BASED ON THE INABILITY TO EXCRETE MERCURY FROM THIMEROSAL-CONTAINING VACCINES OR ANY OTHER FACTOR

There is no vulnerable sub-population of children who could not excrete mercury and thus retain and accumulate it in their tissues, including their brains.

Dr. Aposhian has evoked the concept of a “mercury efflux disorder” to hypothesize that autism/ASD is caused by a build up of mercury in a certain sub-population of children who cannot effectively excrete it. Such claims are scientifically untenable. To support his theory of a “mercury efflux disorder” he cites data from hair studies, a study on teeth, and chelation data. These make up the first three of the six points on which Dr. Aposhian’s hypothesis is built.

Studies on hair do not support any difference in mercury concentrations between autistic children and normal controls.

There have been six published studies on mercury concentrations in hair in autistic children. Dr. Aposhian only cites one such study and an unpublished, uninterpretable abstract. A more comprehensive examination of the hair study data is given below.

The study cited by Dr. Aposhian is that of Holmes (2003), which compared first baby haircut samples from autistic children with those of non-autistic “controls.” The hair samples were cut when children were between one and two years old. The authors claimed to have found a statistically significant lower mercury concentration in the hair of autistic children, indicating a decreased ability of these children to “excrete” mercury into their hair and hypothesized that these
children thus retain mercury in their tissues, including their brain. This study, however, is full of methodological and analytical flaws including myriad problems with the recruitment and selection of controls. Not surprisingly, the Holmes results are markedly at variance with the five other published studies not cited by Dr. Aposhian on this topic. The following are a few of the more egregious problems with the Holmes study.

In 2004, the National Center for Health Statistics of the U.S. CDC published the results of its National Health and Nutrition Examination Survey (NHANES) in which they assessed levels of mercury in U.S. children and women of childbearing age. (McDowell, 2004) This study reported that the mean mercury concentration in the hair of children 1-5 years old is 0.22 ppm; children in the 95th percentile have a hair mercury concentration of 0.64 ppm. The sample used in NHANES was the centimeter of hair closest to the scalp, thus reflecting methyl mercury intake over the last 30 days. In most children who were reported to have consumed fish or seafood three or more times during the preceding 30 days, their mean hair concentration was 0.40 ppm (with a 95% confidence interval of 0.24 – 0.55); fish consuming children in the 95th percentile were found to have a mean hair mercury of 2.00 ppm (with a 95% confidence interval of 0.39-3.62).

In the Holmes et al. study, the mean concentration of mercury in baby hair of autistic children was 0.47 ppm while that of controls was 3.63 ppm. One thing is immediately obvious from these results. The concentration of mercury in the baby hair of autistic children in this sample was exactly in the range of normal children in the U.S. On the other hand, the “normal” controls selected to participate in the Holmes study had a mean hair mercury concentration that was more than 15 times the national average and nearly 10 times higher than the mean observed in children with high fish consumption. These data therefore provide no support whatsoever to the proposition that “excretion” of mercury
into hair is somehow impaired in autistic children. In fact, the only thing the Holmes study suggests is that there was either a bias in the selection of the control population, such that subjects with unusually high hair mercury concentration were preferentially selected, or there were other methodological or analytical errors.

The Holmes study is the only fully published study cited by Dr. Aposhian. While completely ignoring the other studies on this topic, he tells us that the report of Holmes et al. has been confirmed by Hu et al., 2003. The latter is an abstract from a 2003 meeting.

In the Hu abstract, there were three autistic subjects, of unreported age, two of whom were “under treatment for heavy metal detoxification.” The treatment protocol required complete exclusion of seafood from the diet. Not surprisingly, the third autistic individual had a normal hair mercury concentration (0.43 ppm) that was higher than the hair mercury concentration of those individuals on a seafood-free diet. This study also assessed control hair samples from healthy individuals (age range: 6 months to 40 years) and reference standards, but there was no comparison between them and the three autistic subjects. Thus, the Hu abstract shows one autistic individual with normal hair mercury concentrations and two others who were on a seafood free diet who had lower hair mercury concentrations. This result is hardly surprising and does not support the data of Holmes et al., or Dr. Aposhian’s hypothesis of impaired excretion of mercury via hair in autistic children.

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12 It should be pointed out that hair is not a mode of excretion of mercury and the total elimination from the mercury through the body by hair is trivial.
In contrast to the one study and one abstract cited by Dr. Aposhian, there are five other studies published in the peer-reviewed medical literature that were unable to replicate the study and abstract relied upon by Dr. Aposhian.

A study by Ip (2004) evaluated hair mercury concentrations of autistic children and found that they were the same as non-autistic controls with similar mercury exposures. While the mean age at the time of sampling was higher in the Ip study than the Holmes study, that should have no bearing on the outcome. Although there has been a re-analysis (of questionable significance) (DeSoto et al., 2007) of the blood data in the Ip et al. study, the hair data from the study was not challenged.

A study by Fido (2005) assessed hair mercury concentrations of autistic children and compared them with controls. That study found that autistic children have statistically higher mercury concentrations than controls.

The study by Adams (2006) assessed hair mercury concentrations of 51 children with ASD and compared them to 40 controls. There was no significant difference between the two groups.

Similarly, a study by Williams et al., 2007 compared autistic children with neurotypical control siblings and found no significant difference in hair mercury concentrations between the two groups. It is important to note in this study that when Dr. Aposhian was confronted by the Ip study in his testimony in Cedillo he criticized it because the samples were not sent to Doctor’s Data Laboratory, which he endorsed (Cedillo Transcript, p. 200). Doctor’s Data Laboratory is a company that operates out of the realm of mainstream medicine. It specializes in offering direct-to-consumer laboratory testing services and tests that appeal to alternative medicine practitioners. Its tests and results are widely considered to be unreliable by the medical toxicology community. It may be noted that
petitioners utilize the services of, and rely on results from, Doctor’s Data. However, what is significant about the Williams study is that they sent their samples to Doctor’s Data, as did Holmes, and yet the investigators were unable to reproduce the Holmes’ results. In addition, the aforementioned Adam’s study, which similarly could not reproduce the Holmes’ results, also utilized Doctor’s Data.

Similarly, a recent study by Kern (2007) assessed the concentration of several metals in the hair of children 1-6 years old with autism compared to non-autistic controls. This study, which also sent their samples to Doctor’s Data Laboratory, found no statistically significant difference in the hair mercury concentrations between the two groups.

If the hypothesis by Dr. Aposhian of differential incorporation of mercury into the hair of autistics were true, then there should be no difference between autistic and non-autistic children with similar exposures at all ages. In other words, whatever metabolic or genetic factors make autistics different from non-autistics regarding their ability to “excrete” mercury into hair would not be expected to change with age. Thus, the Holmes data is not only implausible, but five other studies, three of which used Doctor’s Data, have failed to replicate it.

Another serious problem with the Holmes study is that it did not evaluate anything that might actually signal a difference in children’s ability to excrete mercury. It is well established and generally accepted that the excretion of mercury following exposure to organic species such as ethyl mercury or thimerosal occurs primarily in feces and, to a lesser extent, to urine. In fact, Pichichero (2002 and 2008) demonstrated that infants rapidly excrete mercury into stool following administration of thimerosal-containing vaccines. Hair is not a significant route of mercury excretion. Bald people do not develop mercury toxicity; this includes infants with little early hair growth. There is simply too
little mercury in hair to possibly impact on body burden. Therefore, if one were truly interested in evaluating whether autistic children have impaired ability to excrete mercury, the medium that should be tested is feces. Holmes could have done this; they did not.

The analytical and methodological deficiencies discussed above are merely the tip of the iceberg. They do, however, provide ample basis for rejecting the conclusion of Holmes and Aposhian as they are clearly scientifically invalid, irreproducible, and insupportable. There are no reliable data to support the assertion that autistic children have a “mercury efflux disorder” manifested by inability to “excrete” mercury into hair.

**There is no reliable evidence that there is a quantitative difference in mercury in baby teeth of children with autism versus controls**

The first of the pillars of Dr. Aposhian’s theory of ethyl mercury-induced autism is a study by Adams (2007), which he says demonstrated that teeth from autistic children had higher mercury content than non-autistic controls. The Adams study assessed baby teeth from 16 children with autism and compared the mercury content with that from 11 control children. They reported that children with autism have a mean mercury concentration in their teeth of 0.15 µg/g (median 0.14) compared to controls that had 0.07 µg/g (median 0.05). They then applied a statistical test known as a “t test” and concluded that the difference is statistically significant. However, there are numerous problems with the Adams’ study. For one, the statistical test they used is dependent upon the assumption that the mercury concentration in the teeth is normally distributed. This is very unlikely given the small size of the study group and they have certainly not demonstrated this is the case. Thus the statistical test they used is inappropriate. Because they provided insufficient information in their paper (for example, are the means given ± standard deviation or standard error?), it is
impossible to check their statistical methodology. More importantly, they did not take into account a number of extremely important variables that are known to profoundly affect tooth mercury concentrations. (Tvinnereim, 2000) The lack of controlling for all of these important factors makes the Adams' study uninterpretable, even if they had used appropriate statistics. Finally, and critical to whether these results are reliable, the Adams' study has yet to be duplicated.

Chelation studies do not support a role for mercury in ASD

The third pillar upon which Dr. Aposhian builds his theory relates to a study by Bradstreet (2003) and what he describes as the confirmation of the results of the Bradstreet by the study of Nataf (2006). However, the Nataf and Bradstreet studies assessed completely different parameters, and the Bradstreet study, as will be described below, is completely uninformative. The Nataf study, as also will be described, says nothing about mercury and the etiology of ASD.

In the Bradstreet (2003) study, urinary mercury excretion following chelation with succimer (DMSA) was evaluated in 221 children with ASD and 18 unmatched controls. The authors report that the post-chelation-mercury concentrations in the urine of the autistic cases was approximately 3 times higher than in controls and that the difference was statistically significant. From this and other sub-analyses, the authors conclude that “children who develop autistic spectrum disorders had significantly greater accumulated mercury than controls,” and this “probably result[ed] from a decreased ability of children with autistic spectrum disorders to excrete mercury, resulting in the retention of potentially toxic mercury levels.” This conclusion neither follows from the data presented, even if they are valid, nor is supported by them. The Bradstreet study has so many methodological and conceptual errors that it provides no useful information whatsoever.
The first problem with jumping to the conclusions reached by Bradstreet is that urinary excretion following succimer chelation is not an accurate or reliable measure of mercury body burden. This has been demonstrated in multiple studies (e.g., Archbold, 2004). Moreover, the authors themselves acknowledge in the article that the higher observed urinary mercury excretion in the autistic cases could merely reflect nothing more than higher current exposure to mercury,\textsuperscript{13} rather than increased tissue retention. In fact, given how the controls were recruited, this is likely. The controls in the Bradstreet study were not randomly selected, but rather, were chosen from a population of children whose parents brought them to Dr. Bradstreet's clinic for elective determination of mercury levels. This was done because of concern about potential mercury toxicity. Bradstreet et al. never investigated whether concern caused the parents of the controls to restrict their child's exposure to mercury in the diet. It is likely that this concern caused them to restrict the child's exposure to mercury from seafood. Thus, bias in the selection of controls raises serious concerns regarding the validity of the results. No effort was made by Bradstreet to control for this factor, a confounding variable of considerable significance.

In addition, since in the Bradstreet study there were no pre-chelation levels reported, it is impossible to tell whether DMSA had any affect whatsoever on mercury excretion. It is absolutely the standard of care in both treatment and in research to assess both pre- and post-chelation levels before reaching any conclusions about the results of chelation. Moreover, the difference in the mean post-chelation urinary mercury excretion between the cases and controls is not dramatic. Although the authors claim that this difference is statistically significant, their statistical methodology is inadequately described and the authors clearly based their analysis on the premise that the new data is normally

\textsuperscript{13} The authors did not report any attempt whatsoever to evaluate either present or past level of mercury exposure in either the cases or the controls.
distributed. However, they did not demonstrate that the data was, indeed, normal. An inspection of the ranges and standard deviations of the data presented suggest a non-normal distribution. Therefore, the statistical tests used may not have been appropriate. Further, even using their described statistics and assuming a normal distribution, I could not calculate statistically significant results from their presented data using the methodology they said they employed.

The Bradstreet study provides no information regarding a potential association between thimerosal-containing vaccines – or mercury in any form – and the development of ASD. Nor does this study support the hypothesis that children with ASD excrete mercury any differently than normal controls. In fact, after considering the Bradstreet (and Holmes) studies, the 2004 IOM Panel concluded they "do not provide any evidence of a relationship between vaccines or thimerosal and autism." (2004 IOM Report pp. 132-134, 140-141)

Dr. Aposhian then tells us that the Nataf study confirmed the Bradstreet results. However, that study (Nataf, 2006) did not measure urinary mercury levels, or any mercury concentrations for that matter. The Nataf study assessed a completely different parameter – the patterns of excretion of molecules known as porphyrins. Porphyrins have not been shown to play a causative role in ASD. They are discussed in greater detail in a later section of this report.

Most importantly, an attempt to confirm and reproduce the Bradstreet results, using more appropriate and sophisticated methodology, found no difference in DMSA-induced urinary mercury excretion between autistic children and developmentally normal controls. (Soden, 2007) Unlike Bradstreet, that study used full 24-hr urinary mercury collections (as opposed to the less preferable “spot”, or single, urine collection in Bradstreet), eliminated seafood for 1 week prior to the study to avoid biasing the results, and compared pre- and post-
chelation results. In the Soden study, there was no demonstrable difference in DMSA-induced urinary mercury excretion between autistic children and neurotypically developing controls.

The fourth pillar of Dr. Aposhian’s theory is that he says that “The most beneficial treatment for autism as reported by the parent’s of autistic children was chelation therapy....” There has never been a study in the published medical literature that supports this claim. Chelation is a treatment reserved for patients with toxic metal poisoning and is medically inappropriate and without scientific support as a treatment for autism.

**There is no recognized disease in medicine known as a “mercury efflux disorder”**

Dr. Aposhian repeatedly evoked the concept of a “mercury efflux disorder” implying that a population of children exists with autism have an inability to excrete mercury. There is no such disorder recognized in legitimate medicine. A search of the National Library of Medicine database (PubMed) for the topic “mercury efflux disorder” fails to generate a single citation. I know of no textbook of medical toxicology, pharmacology, or medicine in general that describes a “mercury efflux disorder.” As described in this report, there are no reliable scientific studies demonstrating a “mercury efflux disorder.”

**THERE IS NO SUB-POPULATION OF CHILDREN WITH INCREASED SUSCEPTIBILITY TO ETHYL MERCURY OR THIMEROSAL THAT HAS EVER BEEN DESCRIBED**
Acrodyinia is neither related to ASD nor can it be considered that it is an example of a susceptible population

Acrodyinia is a medical condition caused almost exclusively from inorganic mercury. Its manifestations have nothing to do with ASD. It is characterized by bright pink or red skin, painful, raw and peeling hands and feet, occasionally gangrenous extremities, inflamed and tender gums with loosening of teeth and perfuse sweating. (Dally, 1997) It was described initially in the 1920s. (Bilderback, 1920; Dally, 1997). In that era, inorganic mercury poisoning was quite common and most clinicians were familiar with its clinical manifestations. This caused Zahorsky to observe as early as 1922 that mercury poisoning and acrodyinia appear to be the same condition. (Cheek, 1959; Dally, 1997) In fact, a textbook in 1938 by Davison gave mercury as the cause of acrodyinia. That mercury was indeed the cause of acrodyinia was ultimately shown by Warkany in 1948.

Many cases of acrodyinia were traced to the inclusion of calomel (mercurous chloride) in teething powders. It is ironic that inorganic mercury poisoning causes loose teeth and inflamed gums because these effects only served to cause parents to give children greater amounts of the teething powder. However, there have been a number of other inorganic mercury exposures that have been associated with acrodyinia including worming medications, a home contaminated with mercury, and mercury from broken thermometers in isolettes. (Davidson, 2004; Waffarn 1979) Some have expressed a view that since not all children exposed to inorganic mercury develop mercury poisoning or acrodyinia that there is a sub-population of susceptible children that get the disease. However, a close examination of the data shows that it appears to be a phenomenon associated with exposure to high doses of mercury and thus almost certainly dose-related. The work of Warkany described above that demonstrated a relationship between acrodyinia and exposure to inorganic mercury found high
levels of urinary mercury in children with acrodynia. Other investigators have also pointed out that almost all cases of acrodynia in which urinary mercury concentrations were assessed, there were high levels present. (Magos, 1997)

In Cedillo, Dr. Aposhian, while testifying about acrodynia, told the court: “I put this first because I want you to remember this, please. The medical establishment did not accept mercurous chloride as the cause, and we’ll say this again.” (Cedillo transcript, p. 90) As described above, this is clearly untrue.

The data on acrodynia clearly shows that it is a syndrome that is fairly typical of inorganic mercury toxicity and appears to occur in children with high exposures, explaining why not all exposed children developed this condition. In Dr. Aposhian’s report, he refers the reader to the Dally paper, which he refers to as “an excellent review.” The fact that acrodynia was described in approximately 1920, and by 1922 Zahorsky pointed out the similarity between mercury poisoning and this condition, is well documented in that article. Also documented in Dally is that acrodynia is associated with high urinary mercury concentrations, representing high dose exposure.

**Polymorphism of the coproporphyrinogen oxidase (CPOX) gene has not been shown to be a cause of ASD and does not define a population susceptible to developing this condition**

In Dr. Aposhian’s report, he tells us that a publication by Woods (2006) has shown a polymorphism in the enzyme known as CPOX. He then tells us that many toxicologists do not cite this in their review articles “because they are not cognizant or familiar with the importance of genetics and genetic toxicology as an area of human toxicology.”
The demonstration of the CPOX polymorphism is not new and has been known and written about since at least the mid-1990s. (Grandchamp, 1995) Porphyrins are molecules that are found in almost all cells and are involved in a variety of normal metabolic pathways. The synthesis and metabolism of these agents are controlled by numerous genes and, as would be expected, polymorphisms (different forms of these genes) exist. CPOX4 is one such polymorphism and is known to occur in approximately 15% of the general population. (Woods, 2005) Individuals with a CPOX4 profile are fully functional and this polymorphism has not been shown to be more common in individuals with ASD.

The various studies that have been done on porphyrin profiles, both in children with ASD and normal individuals, reflect porphyrin metabolism in kidneys. Polymorphisms of CPOX have not been shown to play any role in the genesis of ASD. Very importantly, and completely contrary to Dr. Aposhian’s theory, a recent controlled study has shown that the CPOX4 polymorphism occurs less frequently in autistic children than in unaffected controls. (Rose, 2008)

I see absolutely no justification for Dr. Aposhian’s conclusion that toxicologists are unfamiliar with the importance of genetics. As a medical toxicologist, I have no idea why he would make such a statement. The field of toxicogenetics is one of great interest in contemporary toxicology and is discussed routinely in the mainstream clinical toxicology literature as well as at clinical toxicology meetings. Although Dr. Aposhian cites a 2006 paper on CPOX4, this polymorphism has been known for decades.
THE LACK OF ASSOCIATION BETWEEN THIMEROSAL-CONTAINING VACCINES AND ASD IS UNIVERSALLY ACCEPTED IN THE MAINSTREAM MEDICAL AND SCIENTIFIC COMMUNITIES

The results expressed in this report reflect not only the views of the writer but those of every governmental and mainstream non-governmental agency that has assessed this matter. These include the following:

American Academy of Pediatrics
The American College of Medical Toxicology
The Cochrane Collaboration
The European Medicines Agency
Health Canada
The U.S. Centers for Disease Control and Prevention
The U.S. National Academy of Sciences/Institute of Medicine
The World Health Organization

There is no such agency that has concluded that there is a connection between thimerosal-containing vaccines and ASD.

Yours most sincerely,

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