

Sorting out the spinning of autism: heavy metals and the question of incidence

Mary Catherine DeSoto* and Robert T. Hitlan

Department of Psychology, University of Northern Iowa, Cedar Falls, Iowa, USA; *Email: cathy.desoto@uni.edu

The reasons for the rise in autism prevalence are a subject of heated professional debate. Featuring a critical appraisal of some research used to question whether there is a rise in cases and if rising levels of autism are related to environmental exposure to toxins (Soden et al. 2007, Thompson et al. 2007, Barbaresi et al. 2009) we aim to evaluate the actual state of scientific knowledge. In addition, we surveyed the empirical research on the topic of autism and heavy metal toxins. Overall, the various causes that have led to the increase in autism diagnosis are likely multi-faceted, and understanding the causes is one of the most important health topics today. We argue that scientific research does not support rejecting the link between the neurodevelopmental disorder of autism and toxic exposures.

Key words: autism, autism prevalence, heavy metals, mercury, toxins

INTRODUCTION

In this paper, we argue that increasingly over the past decade, positions that deny a link to environmental toxins and autism are based on relatively weak science and are disregarding the bulk of scientific literature. In this paper, we are not focusing on vaccines, which is but one exposure pathway, but on exposure to toxic heavy metals as a broader class, of which a vaccine containing a heavy metal preservative would be but one possibility of exposure. It should be clear that any link between toxins and autism is almost certainly mediated by one's genetic makeup, and that other toxins, such as organophosphates (Eskenazi et al. 2007) likely play a role as well. In this conceptualization, the gene pool did not change, but exposure to substances that directly affect gene functioning is changing. Therefore, the reason why one five year old has developed autism and another has not, is indeed in large part a function of the individual's genes. But the question is still why more children are being diagnosed as autistic today

than 30 years ago. Many factors are different today than a generation ago: autism awareness, exercise, diet, use of sunscreen and outdoor play, the amount of toxins in the environment – to name just a few. It is the authors' opinion that all of these things matter. Nevertheless, our interest is in the exposure to toxins, and in this paper to toxic heavy metals. Some prominent researchers still deny that there has been any actual increase in the cluster of behaviors that fall under the umbrella of autism spectrum disorders (ASD). For example, Roy Grinker in his top selling book on autism (2007) denies an actual increase has occurred, maintaining that it is all due to increased awareness and broadening of the diagnosis. Our opinion is not only that the increase is real, but that the increase in various contaminants is a major factor responsible for that increase.

QUESTION OF THE RISE IN AUTISM INCIDENCE

Before further discussion, we wish to make clear the following: there is evidence for changes in diagnostic practice to have played a role in the autism prevalence rate. To our knowledge, there is no one who denies that diagnostic changes have occurred. When adherents to

Correspondence should be addressed to M.C. DeSoto
Email: cathy.desoto@uni.edu

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the no-true-increase-view use the fact that diagnostic change has occurred as evidence that the other side is wrong and they are right, they are attacking a straw man because no one denies that some changes in diagnostic practices have occurred. For example, it is clear that many children receiving an ASD diagnosis today would not have been diagnosed as having archetypal Autism prior to the change in diagnostic criteria. Professional understanding of ASD has evolved from a yes-or-no diagnosis to a spectrum of severity.

Second, we wish to make clear that we agree that many of the studies that existed a few years ago left open the possibility that there was no increase in the actual incidence of Autism. But we believe that recent studies and recently available data sets are providing convergent evidence for a secular increase across numerous countries. We also think the within-time prevalence patterns differing as a function of where the pregnancy occurred (Windham et al. 2006) in tandem with CDC and IDEA state-by-state differences suggest the actual differences in current prevalence rates are occurring. Furthermore, such differences dovetail with recent findings that Autism rates are predicted by distance to and exposure to toxins (reviewed below). In sum, we fundamentally agree with Eric Fombonne about the state of things a few years ago (2003a):

“Whereas evidence suggests that a substantial part of the increase in prevalence is due to methodological factors, the additional possibility of a secular increase can not be ruled out. Unfortunately, most available epidemiological data (were) derived from surveys, and the few studies that provide incidence rates have not been adequate to test the hypothesis. In addition, no strong environmental exposures have been identified.” (Fombonne et al. 2003a, p. 88)

Several things have changed since his writing. One, environmental exposure to toxins has been identified, tested and supported (see Table I). Independent lab groups have now shown that autism rate at the level of school districts is not random but appears related to the amount of and distance to toxic emissions within states (Palmer et al. 2008, DeSoto 2009). Exposure to toxins during pregnancy or early infancy predict later ASD symptoms (Eskenazi et al. 2007). Second, the ability of low levels of mercury (levels that 8% of American women have in their blood streams) to cause specific damage to developing human brain cells have quite clearly been demonstrated (e.g., Tamm and Duckworth 2006). Moreover, the level of some heavy metals such as mer-

cury in the unborn child may be as much as 70% higher than the mother’s circulating supply (Stern and Smith 2003, Morrissette et al. 2004) further increasing the plausibility of such an environmental candidate. Third, the only study that had directly tested blood levels of mercury among autistic children and control subjects at the time of Dr. Fombonne’s quote was Ip and coauthors (2007) which was based on a series of now acknowledged errors (see below). Again, these studies have all been published in the last few years, and our point is that the weight of scientific evidence has shifted markedly.

As a noteworthy example, Atladottir and coworkers (2007) reported the change in incidence for children born in Denmark between the years 1990 to 1999 (n=669,995). Atladottir used standardized case ascertainment and standardized diagnostic procedures to document an increase in both Autism Spectrum Disorders (ASD) and Childhood Autism in Denmark. The increase was documented separately for two classifications: both ASD and Childhood Autism per se, and the increase was even more pronounced when the stricter diagnostic definition of Childhood Autism was used. It is important to realize that this recent study is employing strong methodology, consistent with the strictest Fombonne-approved methodology for “repeated surveys using the same methodology” across time (Fombonne 2003b, p. 375) with very large studies being preferred to access actual incidence increases (Fombonne 2003b, p. 376): this large study counts ASD and autism separately, the same way, across time in a circumscribed location.

Unlike Atladottir’s methods, which considered both the broad ASD diagnosis as well as classic Autism across time using standardized assessment techniques, United States school districts generally classify ASD as one category (whether mild, Asperger’s type symptoms or full blown classic autism) for purposes of counting cases. Because of this, the increase in the number of children classified by school systems as having an autism related disorder, while suggesting that there may be some real increase occurring, cannot be taken as strong evidence that there has been a 700% increase in incidence of classic autism, even if the rate of diagnoses did increase by 700%. It is important that this does not serve to confuse the issue; there are now other lines of evidence for an increase that better control for differences in diagnostic practice across time and on a large scale (e.g., Atladottir et al. 2007). There are however still few such carefully-controlled studies in existence that measure recent

changes in prevalence (separating ASD versus classic autism, similar ascertainment methods across time, circumscribed region, focusing on the last decade, etc), and it is possible to scrutinize the few individual studies' methods when results are contradictory.

There are some studies that reportedly do not show an increase. Latif and Williams (2007) report the lead author's diagnoses of ASD across time (1988 to 2004) in an economically depressed area of England and conclude that classic Kanner's autism has not increased. Like Altadottir and colleagues (2007), this study used essentially the same diagnostic criteria over time, a crucial control when addressing the question of increases over time. However, the study may have been limited in that determination of the precise diagnoses (ASD; "classic Kanners" autism; "other" autism), did not employ any of the guidelines or standardized tools recommended for diagnosing and classifying autism (Cicchetti et al. 2007, Ozonoff et al. 2007), but relied on clinical judgment. The lack of objective confirmation of diagnosis was appropriately acknowledged by the authors, and of course does not negate the results. It is important to note, however, that the decrease in "classic Kanner's" autism reported by Latif and Williams occurred concurrently with their report of a more than four fold increase in "other forms" of childhood Autism, and a more than doubling of ASD cases. Thus, the reported lack of increase in Kanner's autism is based on the judgment for classification of approximately two children per year to other forms of autism—occurring in the context of a dramatic increase in total autism cases across the years of study.¹ Total autism cases were documented as increasing.

Researchers (Fombonne 2001, 2003a, Wazana et al. 2007) have correctly noted that attempts to separate increased autism prevalence caused by changes in diagnostic criteria as opposed to an increase in actual incidence must use the same area, the same case ascertainment and the same diagnostic criteria across time as a minimum. An additional recent study that appears to meet the criteria is by Barbaresi and coauthors

(2009). They report that relying on clinical diagnosis alone results in an increased estimate of the change in incidence. Because we feel this has the potential to be cited as supporting the no-real-increase camp (which would not be the fault of the authors who do not formally put forth this conclusion), we think it is important to address. Barbaresi and coworkers report that while the increase in clinical diagnosis was 22 fold, utilizing a more careful research-based methodology (reviewing information from all health care and school sources for the entire population), showed increase in incidence closer to an eight fold (from 5.5 cases per 100 000 in the early 1980's to 45 cases per 100 000 for the mid 1990's). The authors' main conclusion was focused on the discrepancy between clinical and research-based ascertainment methodology (which was perfectly appropriate). However, it should also be noted that using either clinical diagnosis or a more careful research based incidence approach, a sharp increase is observed in a circumscribed region. Such results may be interpreted as further documenting that an increase has occurred, also including using the more stringent research-based incidence measure. Certainly finding a range of increase between 800% and 2200% using very different methods does not lend credence to the idea that no increase has occurred. Furthermore, the discrepancy in the size of the increase is perhaps best seen as an artifact of selecting the specific interval of time (grouping of the years 1980-1983 and using this time period as the starting point). Had there been one (just one) more clinically-diagnosed case in that specific time period, or had the years before or after this interval been selected as the starting point, it would not have resulted in the clinical-case-incidence increase being significantly higher than the research-based increase. For example, if one compares the years of the sharply increasing prevalence (1988 across 1997), one method shows a seven-fold increase and the other a nine-fold increase in autism. The data provided by Barbaresi and others (2009), appear to demonstrate an increase in incidence regardless of the method employed to count cases. Although it is necessary to refer to the original article for full clarity – it shows a clear, in fact almost parallel, increase in autism for the circumscribed region of Olmstead County, Minnesota across time using two distinctly different approaches to case ascertainment.

Overall, there are some studies that suggest at least some true increase in incidence has occurred (e.g.,

¹ The three person team that made all the diagnoses employed Kanner and Eisenberg's 1956 diagnostic criteria for diagnosing autism thorough out the study, but in 1993 the criteria used for ASD and Aspergers were updated to the new editions of ICD and DSM. It is not stated why the DSM was not used for classic Autism. The designation of "autism : other form" refers to autism that was not in keeping with the 1956 Kanners criteria and was also a separate category from either ASD or Aspergers; "other form" included children who had atypical autism or mental retardation associated with autism. The wording made it somewhat unclear to the current authors if the diagnostic criteria for "other form" was officially modified by the diagnostic team in 1993, but we assume it was not.

Atladdottir et al. 2007), and some that do not (e.g., Latif and Williams 2007). Professionals should judge the merits of the methods, and make sure that the full results are clearly understood as they weigh the evidence on this question. Indeed, a survey of Ph.D. level psychologists has suggested that the majority hold the opinion that at least some real increase in incidence has occurred (Osborn 2008).

We were interested in a survey of the empirical literature on the question of whether there is a link between heavy metals and autism. A PubMed search of “autism AND heavy metals OR autism AND mercury” yielded 163 articles. The majority of these articles were review, commentaries or letters. Some were of a novel type (e.g., a brief editor introduction, a note on unusual chelation side effect, comment on a movie portrayal, etc). An empirical data article was defined as using a relevant data set collected by or analyzed by the authors. Articles were coded as empirical data supporting a link, empirical data rejecting a link, or as not applicable (not relevant, review, commentary, etc). An inter-rater reliability check was performed on the ratings and suggested that the classification was straightforward ($r = 0.95$). Of these 163 articles, 58 were research articles with empirical data relevant to the question of a link between autism and one or more toxic heavy metals. Fifteen were offered as evidence against a link between exposure to these metals and autism. In contrast, a sum of 43 papers were supporting a link between autism and exposure to those metals. Case controlled data are particularly important to this question. It is worth noting that there have been only three empirical articles directly comparing those with and without an ASD on mercury levels in the body to a control group of normally developing matched controls that report that report no link (Ip et al. 2004, Soden et al. 2007, Hertz-Picciotto et al. 2010). While, the most recent article appears to be the strongest, lacking any obvious errors or flaws (we think that this recent article does provide at least some legitimate evidence contradicting the hypothesis that autism and heavy metals are linked), the other two are seriously flawed. In fact the flaws are of such magnitude that both studies actually should be interpreted as offering support for a link between autism and heavy metals, contrary to their conclusions but in keeping with the majority of studies that provide evidence for a connection.

Soden and coworkers published a report in 2007 concluding that chelation should not be used as a treatment for autism. While chelation for autism may well be ques-

tionable, there are many problems with Soden and coauthors methods of analysis. Of concern, the study appears to deny a link between heavy metals and autism and has been cited as proving that in subsequent review articles: “One other study concluded that generally no heavy metals were in any way involved (in autism)” (Hughes 2008, p. 426 - citing Soden et al. 2007). Soden and others data – if they show anything – actually show the opposite.

Problems with the original article included treating a continuous variable as dichotomous, taking the liberty of defining non-zero numbers as actual zeros, and employing methods that resulted in having 95% of samples returned by the lab as too low to detect. Nonetheless, there is something of interest in the data that was not noted by Soden and colleagues and which prompted a letter to the editor (DeSoto 2008) which will be elaborated upon here. Soden and coworkers concern that the binomial distribution approach, specifically the Clopper-Pearson Exact Confidence Interval, is the best way to analyze the data is questionable, and is addressed in a footnote.²

We begin by describing some serious problems with the Soden and coauthors (2007) methodology. Next, we provide statistical analyses of the data using categorical statistical techniques. This is important because it makes it very clear that no matter the approach, the data (faulty as they may be) support the contention that those with autism had higher levels of heavy metals. We then conclude by clarifying the procedure Soden and others used to arrive at their conclusion, as it may not be fully clear to all readers.

CHELATION METHODOLOGY

Soden states the purpose of her research in the introduction, “the DMSA provoked excretion test is used to

¹ Soden employed a binomial proportion confidence interval. This type of test does exactly what the name implies: it is used for the purpose of estimating how confident one can be that, for a sample where the presence of some condition has been observed, the proportion in the sample that have the condition is reflective for the larger population. It is based on the binomial distribution, and is designed for variables that are inherently categorical. The level of a heavy metal is not inherently categorical. The problems associated with treating a continuous variable as a categorical one are well known and a large body of literature exists about the dangers associated with this practice (see for example, Maxwell and Delaney 1993, MacCallum et al. 2002). I believe that what Soden et al. might say is that their outcome of interest was indeed categorical: the number of persons who would be helped by chelation as defined by having toxic levels of heavy metals excreted post chelation. Although this would be categorical way to define the outcome, this justification (the category represents an underlying feature of the individuals) and many others have been covered in statistical literature and are generally frowned upon as a way to defend treating a continuous variable as categorical when testing a hypothesis, “dichotomization is rarely defensible and often will yield misleading results.” (MacCallum et al. 2002, p.19). Further, if employing the binomial approach, Clopper-Pearson is not the ideal approach, the adjusted Wald would probably be more appropriate for the this data set (Agresti and Coull 1998, Yoo and David 2002).

challenge the assertion that a substantial proportion of autistic children have an excess chelatable body burden of heavy metals” (Soden et al. 2007, p. 477). Soden and coworkers report excretion amounts for 15 autistic persons and four normally developing controls. There was a specific protocol for the chelation and urine collection that required the parents to give set amounts of chelating agent at specific time intervals and to collect all urine for 24 hours. Some children were not toilet trained, which required the use of special diapers and collection procedures. There is not a direct way of measuring adherence to the dosing and collection protocol, but it was possible to check for one parameter: completing the chelation within 7 days of baseline. Approximately 27% of the autistic group and 25% of the control group did not comply. Although Soden appropriately acknowledges that “accurate results from a DMSA challenge depend upon ingestion of all DMSA doses, appropriate fluid intake, and collection of the entire urine sample” and that parental compliance with the protocol is a source of potential inaccuracy of results (Soden et al. 2007, p. 480). This potential is very real, and the published results are not consistent with a successful chelation methodology.

The ability of DMSA to increase the amount of mercury and heavy metals excreted in urine is well documented and the chemistry of the process is well understood. DMSA binds with heavy metals of the opposite charge and results in an increase in urinary excretion of heavy metals from the body. Even in studies with similar aims as that of Soden – and which lead to the conclusion that chelation for symptom improvement in sub-toxic cases is probably not effective – DMSA increasing excretion of heavy metals post chelation is still found (Sandborgh-Englund et al. 1994, Frumkin et al. 2001). For example, Sandborgh-Englund et al. administered DMSA to 10 participants and also had a placebo group of 10 participants. Like Soden et al. Sandborgh-Englund used 24 hour urine collection. The lab’s detection limit was lower and unlike Soden et al. all participants had detectable levels of mercury; and, second, 90% showed an increase in 24 hour content of heavy metals in urine after chelation. The fact that only 3 of 20 in Soden’s study showed an increase in even one of the measured metals may be seen as a red flag that something about the methods used fundamentally did not work. To wit: two participants excreted LESS heavy metals during chelation than before chelation and only three excreted more. This is not in keeping with research that shows DMSA results in an increase in excretion of heavy met-

als (Graziano 1986, Fournier et al. 1988, Roels et al. 1991, including those with non-toxic levels: Sandborgh-Englund et al. 1994, Frumkin et al. 2001).

For example, participant 1’s baseline urine excretion arsenic level is 25 micrograms. But, of concern, chelation does not increase the excretion, it appears to lessen it. This same participant has an elevated cadmium level at baseline, but instead of increasing, after DMSA chelation, none is detected. Given what is known about chelation, these results are hard to understand. The parentally-monitored chelation protocol efficacy is also called into question from the results of Participant 12. Chelation by the parents produced no measurable arsenic, no cadmium, and no lead. But when chelation was repeated after a month (most probably under some medical supervision and strict adherence to protocol), each of the heavy metals (that were measured at zero level under the original DMSA protocol) was now detected. As stated, this particular child had no seafood for a month prior to the second chelation, which probably rules out organic arsenic as a transient source. Soden admits that the reasons for these facts are not explained, but some reasonable explanations for these unusual results are required if any conclusion is to be made based on the results of the chelation process as employed. Authors offered no explanation, therefore there is a real question as to whether the chelation procedure as employed worked at all.

However, there is an interesting result regarding heavy metals. We believe Soden and colleagues (2007) erred in not bringing these results to the readers’ attention as it is a question of some theoretical interest (and very hot debate). Specifically, there are lab results for heavy metals in the urine of known autistic children along with a sample of normally developing control children. These results are of interest and they should not be lost.

ANALYZING THE NUMBERS REPORTED BY SODEN AND COWORKERS

Based on the lab results, Soden and his team (2007) classified all autistic participants (including those whose heavy metal values were either “non-detectable” as well as those who had detectable amounts but were not defined as being in the “toxic” range) as zero. Not surprisingly, the obtained confidence interval included zero (0-22%). It should also be mentioned that because all values were coded as zero, the confidence interval is one-tailed, by definition. Concerned about this statistical approach, the first author used the *t*-test to

assess whether autistics reported in the original article differed from controls, overall, in the proportion of individuals exhibiting heavy metal excretion (DeSoto 2008). In Soden’s rebuttal, she argued in support of her original conclusion on the grounds that the approach used in the reanalysis violated statistical assumptions associated with a *t*-test analysis. Therefore, Soden and others appeared to claim that conclusion that autistics had more heavy metals was dependent on the use of the *t*-test approach. It was not.

BINOMIAL CONFIDENCE INTERVAL APPROACH

For the sake of argument, let us assume that the binomial confidence interval is the correct way to analyze the data set. Again using the results as reported (Soden et al. 2007, Table I), four heavy metals were tested in 15 participants. The provoked urine samples resulted in seven detectable measures in 60 autistic samples (11.7%). Soden and others Table II illustrates that no metals were detected at any point in any of the normally developing controls either pre or post DMSA provocation (all are coded as zero). Using the Soden-approved Exact Confidence Interval, the 95% confidence interval for the autistic sample proportion is 0.0482 to 0.2257, and it does not include zero. We also calculated the confidence intervals using a 99% confidence interval. Again, none of the confidence intervals included zero.

To avoid potential to deflect these results and to make it very clear that the result of heavy metals in the autistic group is not a matter of cherry picking which exact way to statistically test, the results using multiple techniques are provided. Here are the 95% confidence intervals for the proportion of 24 hour urine samples in the autistic group that detected heavy metals after the (supposed) chelation (7 out of 60 samples) using different methods of confidence intervals for a proportion:

Wald Method	0.0354 – 0.1979
Adjusted Wald Method	0.0547 – 0.2248
Score Method	0.0577 – 0.2218
Clopper Pearson Exact Method	0.0482 – 0.2257

If one wishes to conceptualize the question as the number of autistic persons who had at least one heavy metal excreted post chelation, here are the 95% confidence intervals for the proportion of autistic patients whose 24 hour urine samples detected any of the heavy metals after the (supposed) chelation (27%, 4 out of 15 persons):

Wald Method	0.0035 – 0.4905
Adjusted Wald Method	0.0216 – 0.1639
Score Method	0.0262 – 0.1593
Clopper Pearson Exact Method	0.0185 – 0.1620

If one wishes to conceptualize the question as the number of autistic persons who had at least one heavy metal increase post chelation (We think this is relevant as this concern represents one of Soden’s criticisms of the first author’s reply to the original article), here are the 95% confidence intervals for the proportion of autistic patients whose 24 hour urine samples increased after the (supposed) chelation (3 out of 15 persons):

Wald Method	0.0051 – 0.1051
Adjusted Wald Method	0.0117 – 0.1425
Score Method	0.0171 – 0.1370
Clopper Pearson Exact Method	0.0104 – 0.1392

To be clear, there is often more than one way to correctly test a hypothesis, but some ways are clearly wrong. Some would even argue none of the statistical analyses are appropriate given all of the problems associated with the data and study itself.

SODEN AND COWORKERS (2007) DUBIOUS STEP

What Soden and coworkers have done is questionable. Soden has taken the liberty of defining anything below the lab’s toxic level as a non-case, a zero. Here is the key quote: “Based on the laboratory’s reference range for unprovoked 24-hour urine collections, the

proportion of autistic patients in this pilot study whose DMSA provoked excretion of AS, Cd, Pb or Hg rose to the potentially toxic range is zero.” (Soden et al. 2007, p. 479). If readers refer again to Table I, what is being done is to consider every number in that table as a zero (the actual numbers and the non-detects are all counted as zeros). The difference between Soden’s results and those above is that we have used the data that came back from the lab as reported in Table I, similar to what other researchers have done when they have studied the results of chelation (even those cited by Soden et al. 2007 such as Frumkin et al. 2001). In contrast, Soden and coauthors simply defined all of the numbers including non-detects and detected values alike as non-cases: a zero. Besides the disregard for the extensive body of literature documenting there are neurological effects that occur well below “toxic” levels (see for example Bellinger et al. 2008), this is a questionable approach. As noted above, Sandborgh-Englund and others (1994) did a somewhat similar study and did not define low levels as zero – their actual measured results were used, even when they were less than 1 microgram for 24 hour urine excretion. Frumkin and colleagues (2001) also used 24 hour urine levels for mercury. The mean in the control group was far below levels considered “toxic” (2.89 micrograms) and the scores themselves were used and analyzed as continual measures. In the end, the statistical test conducted by Soden and coworkers is meaningless and distracting from the essentials of what was done. The authors measured metal levels, then (based on the lab definition of toxicity) all values were defined as zero, then – they tested this actual zero statistically and found that one could not rule out zero.

An important limitation of the study as a whole is the manner of sample collection which is highly sug-

gestive that chelation was not successfully performed (refer again to Subjects 1 and 12), but may also call into question the heavy metal results that were obtained and analyzed. Usually, 24 hour urine collection requires the specimen be collected in an acid-washed container with a specific nitric acid content, with refrigeration of the sample. It is not at all clear how closely this was followed by the parents, and there is no discussion of what effect this might have. For example, in the Frumkin and coworkers (2001), the study reviewed by Soden, the urine was collected in “plastic containers provided by the Centers for Disease Control” all urine samples “were kept refrigerated”. It is noted that the volume was measured and if a 24 hour urine collection was less than 500 mL, it was “considered incomplete and was excluded” (p. 168). Such controls are not mentioned by Soden in his paper. Given the diapering protocol used, information about potentially incomplete sample would have been helpful. But let readers be clear about this central point: if one is willing to consider the actual numbers reported and test those numbers, the results are clear - a larger proportion of autistics had heavy metals excreted as the result of chelation. An additional study that has been frequently cited as evidence that autism is not related to heavy metals is Ip and others (2004). The original article has been cited a total of 83 times.

IP AND COWORKERS (2004)

In 2004, the first case controlled study of circulating levels of mercury was published in the *Journal of Child Neurology*, a journal whose editorial board included one of the lead authors of the paper. In 2004, Ip and coworkers wrote: “The mean blood mercury levels of

Table I

Replicated findings by four or more lab groups on autism prevalence and neurotoxins

Those with ASD have higher levels of toxins.	Pockets of higher prevalence exist (within-study).	Increased rates associated with sources of contaminants.	Those with ASD have decreased detoxifying ability.
Edelson 2000	Hoshino et al. 1982	Windham et al. 2006	Serajee et al. 2004
Nataf et al. 2006	Oliviera et al. 2007	Roberts et al. 2007	James et al. 2004
Eskenazi et al. 2007	Kamer et al. 2004	Palmer et al. 2008	Poling et al. 2006
Geier and Geier 2006	Barnevik-Olsson et al. 2008	DeSoto 2009	Pasca et al. 2008
DeSoto and Hitlan 2007			

the autistic and control groups were 19.53 and 17.68 nmol/L.” and described their results as showing, “there is no causal relationship between mercury as an environmental neurotoxin and autism.” (Ip et al. 2004, p. 431). These results have been cited as supporting that autism is not related to mercury, or to vaccines: “By and large, biological studies of ethylmercury exposure have also failed to support the thimerosal hypothesis” (Fombonne et al. 2006, citing Ip et al. 2004 - as evidence). However, the means reported in 2004 actually are significantly different ($p < 0.05$, two-tailed). To be clear, the numbers published in the 2004 paper result in a significantly higher mean blood mercury level among autistics, but through a series of typographical errors and a miscalculation of a *t*-test, inaccurate results were reported. The author of record has publicly acknowledged that these numbers and the statistical calculation were in error in an erratum (Ip et al. 2007) and the journal editor notes the reason given was a series of typographical errors (Brumback 2007). Furthermore, a careful and correct analysis of the full data set results in a statistically significant difference (Brumback 2007, DeSoto and Hitlan 2007, DeSoto 2008) with autistic children having higher mean levels of mercury.

As can be seen by comparing the erratum to the original article, the standard deviations were wrong for both groups, the stated statistical significance in 2004 was not even close: their original stated level of statistical probability was off by almost 10 fold.

GENERAL DISCUSSION

We analyzed the data reported in some articles that have been, or might be, taken to support the view of no-real-increase or no-environmental-connection. Overall, we have offered a critical view of some of the literature from the perspective of research scientists who have become interested in the topic within the past five years and sought to gauge the actual state of scientific knowledge regarding autism etiology. To our knowledge, there have been only three empirical, case-control studies of those with and without ASD compared on a measure of actual mercury levels in the body that purportedly fail to find any link (Ip et al. 2004, Soden et al. 2007, Hertz-Picciotto 2010). Two of these data sets actually show that those with an ASD appear to have more metals (Ip et al. 2004, Soden et al. 2007), contrary to what the original authors say about the data. A very recent study reported in 2010 by

Hertz-Picciotto offers evidence against an association without any major flaws. On the other hand, there are numerous scientists that have investigated autistic persons and compared measured levels of heavy metals, reported differences, and concluded a difference does exist. Recent examples include Yorbik and coworkers (2010) and Adams and others (2007). Other researchers have used indices of heavy metal exposures such as the zinc/copper ratio (Faber et al. 2009), urinary porphyrins (Geier et al. 2009), or measured toxins combined with genetic expression (Stamova et al. 2009). To summarize, of the 58 empirical reports on autism and heavy metal toxins, 43 suggest some link may be present, while 13 reports found no link. Even with the tendency for null results not to be reported, it cannot be said there is no evidence for a link between heavy metal toxins and autism: although the question may still be open-in sum, the evidence favors a link.

This particular controversy does have truly high stakes for many reasons. Pharmaceutical companies have been known to offer significant monetary support to the research-related endeavors of scientists whose research findings do not support a link between vaccination and autism or which might have had relevance for evaluation of their products (vaccines). In as much as this controversy has become heated, it may have at times been tainted with opinions that are too strong to be called objective, and science may be a bit thwarted. When we published our reanalysis of the data of the Ip et al. data, we were a bit surprised to receive many highly emotional responses, and some threats. We think the question of toxins and autism should be seen as broad, perhaps including but also transcending any link to vaccines. A recent empirical article against a link between vaccines and various developmental outcomes in the *New England Journal of Medicine* included a disclosure statement noting that seven of the authors had received fees from Merck, Kaiser Permanente and other pharmaceutical companies that may have or had an interest in disproving any link to thimerosal and/or mercury exposure and developmental disorders (Thompson et al. 2007). First, it is important to be very clear that we do not believe that authors would purposefully change their data, or consciously misstate conclusions. Not only would this be unethical, but the stakes are very high. But this does not mean there is no bias; the bias would be subtle and far less nefarious than any sort of purposeful altering of data. If a person has publicly staked his/her career on a cer-

tain position being right, it may become harder to keep a truly open mind, even when new data become available and even when the original intent was to be objective. A way this bias might manifest itself is an overstatement or slight misstatement of results. We feel that both sides have been guilty of this, and this happens when a person becomes so confident in the correctness of his/her own view that he/she no longer reviews evidence to the contrary. Unconscious bias may exist even in the best scientists.

For example, Paul Offit concludes that Thompson and others (2007) study “found no evidence of neurological problems in children exposed to mercury-containing vaccines” (Offit 2007, p. 1979). But is this really true? According to the article’s authors, they detected only a “few significant associations with exposure to mercury” (Thompson et al. 2007, p. 1281). Of some interest to the question of early exposure and autism, “Increasing mercury exposure (in the first month of life) was associated with poorer performance of a measure of speech articulation.” (Thompson et al. 2007, p. 1281), although this finding is in need of replication, it is of interest since poor articulation occurs in those with autism (Shriberg et al. 2001). Among boys, higher mercury exposure during the first month was associated with an increase in performance IQ. This is again interesting because children with autism are known for having an uneven IQ performance such that their performance IQ is often higher than their verbal IQ (Ehlers et al. 1997). To be sure, overall, the results are not overwhelming and the inclusion of so many measures (42 different outcomes) makes it plausible to write off the few significant results as chance occurrences. But if the aim of the study was meant to see if thimerosal might relate to autism, future research may want to target specific measures based on the autism literature and make specific predictions. If the aim was to see if thimerosal relates to general cognitive skills, it would have been wise to select tests previously shown to relate to mercury exposure. For example, past research (Weil et al. 2005) has shown that higher blood levels of mercury are associated with lower scores on visual memory (not tested by Thompson et al. 2007). There is, in fact, a significant amount of literature on mercury and cognitive function for both young children (Lederman et al. 2008) and adults (Yokoo et al. 2003, Zachi et al. 2007). In general, higher levels of mercury are associated with reductions in certain psychomotor tests and prenatal exposure to mercury often

results in reduced working memory in humans and in animals (Goulet et al. 2003). The most recent research suggests that prenatal exposure specifically affects a type of learning sometimes referred to as “perseveration” especially in reversal learning (such as the Wisconsin Card Sorting Task). It has been suggested that contradictory results (and even lack of results) might relate to whether an outcome taps this precise domain (see Newland et al. 2008 for a review).

This is the sort of bias, whether conscious or unconscious, that occurs. Because some of the authors of the Thompson study have publicly aligned with opposing a mercury-autism link (by taking consulting fees), they may be unconsciously more prone to review studies that support their view, less likely to review opposing viewpoints, and may eventually become unaware of relevant research (e.g., Newland et al. 2008). By using 42 measures and finding only a small handful of effects, it is easy to say the obtained relations are chance occurrences. Then, another scholar summarizes the study and slightly changes the results based on a world view that there is no effect of thimerosal, “found no evidence of neurological problems in children exposed to mercury-containing vaccines” (Offit 2007, p. 1279). Then this assessment gets quoted by those who do not bother to look carefully at the original study, and scientific advancement becomes stifled.

The question about toxic exposure and autism is open, with the weight of evidence favoring a connection that is not well understood. Although it is not possible to say with certainty, it seems likely that the connection would be mediated by genetic susceptibility and ability to detoxify. That is, some people have genotypes that confer higher susceptibility to toxic exposures. If so, then 50 years ago few people would have had enough toxic exposure to have the neurological changes that result in autism. Today, because many rather than few children are exposed to all sorts of neurotoxins with lesser resources to detoxify the body (environment, diet, lifestyle) those that are vulnerable, may develop autism. Although to say so is perhaps cliché, it could not be more true in the case of autism and neurotoxins: more research is needed.

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