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Review

Acetaminophen Causes Neurodevelopmental Injury in Susceptible Babies and Children: No Valid Rationale for Controversy

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Abstract: Evidence that early life exposure to acetaminophen causes neurodevelopmental injury in susceptible children has mounted for more than a decade. Evidence is diverse, including extensive work with laboratory animals, otherwise unexplained associations, factors associated with the metabolism of acetaminophen, and some limited studies in humans. Although evidence has reached an overwhelming level and has been reviewed in detail recently, some controversy remains. In this narrative review, some of those controversies are evaluated. First, the associations through time between acetaminophen use and the prevalence of neurodevelopmental disorders are considered. A systematic review reveals that the use of acetaminophen in the pediatric population was never tracked carefully, but historical events that affected use of the drug were documented and are sufficient to establish apparent correlations with changes in the prevalence of neurodevelopmental disorders. Second, problems with exclusive reliance on results from meta-analyses of large datasets and from studies involving small time frames of drug exposure are reviewed. Third, the potential bias in a study designed to separate the role of vaccines and acetaminophen in the induction of autism spectrum disorder (*Autism* 2008;12:293-307) is carefully evaluated. Finally, evidence demonstrating why some children are susceptible to acetaminophen-induced neurodevelopmental injury is examined. It is concluded that, at least among the factors considered, there is no valid rationale for controversy regarding the conclusion that early life exposure to acetaminophen causes neurodevelopmental injury in susceptible babies and small children.

Keywords: acetaminophen; autism; paracetamol; neurodevelopment

1. Introduction: Mounting evidence for the induction of neurodevelopmental injury by early life exposure to acetaminophen

Evidence that acetaminophen exposure during early development is a primary inducer of neurodevelopment injury has been mounting for more than a decade. Although evidence is largely circumstantial or based on studies in animal models, the preponderance of evidence weighs so heavily that a causal relationship can be inferred with no reasonable doubts remaining (1). Evidence demonstrates that, while most children are relatively unharmed by exposure to the drug, some children are at risk due to the presence of oxidative stress (1, 2). Evidence points in particular toward induction of autism spectrum disorder (ASD), with possible connections to both developmental delays and attention deficits (1). Further, evidence points toward exposure between birth and approximately 5 years of age as being the period of highest risk, with prenatal exposure being significant but less consequential (1, 2). This evidence has been reviewed in detail recently (1, 2), and will not be reviewed in detail again here.

A recent, exhaustive review of the literature, compete with citation tracking, demonstrated that, within the medical profession, acetaminophen is widely assumed to be safe

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when use as directed in the pediatric population (3). Unfortunately, the widely held belief that the drug is safe for pediatric use is based on numerous clinical studies that assume the liver will be the target of the drug's toxicity (3). Indeed, in adults, the liver was found to be the target of acetaminophen toxicity during the 1960s (4-6). At that time, however, the view that babies metabolize drugs in a manner identical to adults was already known to be a dangerous assumption (7); this knowledge has yet to be applied to the toxicity of acetaminophen in human children (3). More than a decade later, a study using laboratory animals demonstrated that this assumption should not be applied to the metabolism of acetaminophen (8). Although the target organ of acetaminophen toxicity in newborn rats was not identified in that study, the target organ was demonstrated to *not* be the liver (8), a finding that has been recently verified (9). It was only within last decade that the brain was identified as a target organ for acetaminophen toxicity in newborn laboratory mice based on profound, long-term losses of cognitive function following exposure to relatively low doses of the drug (10). In support of the view that acetaminophen is neurotoxic, a 2010 study in *adult* rats demonstrated that acetaminophen induces death of cortical neurons at concentrations lower than those required to induce acute liver failure (11). This evidence was reviewed recently (1), and will not be reviewed in detail again here.

The preponderance of evidence pointing toward the conclusion that early life exposure to acetaminophen causes neurodevelopment injury in susceptible babies and children by has not been directly challenged. Nevertheless, the conclusion that the drug is hazardous for neurodevelopment might be considered controversial by some, and objections have been voiced. Those objections center around one or at most a few of the numerous lines of evidence, and should be considered in light of the entire body of evidence. With this approach in mind, herein we review several issues that may be considered controversial in the field. In particular, issues associated with studies in humans, studies in animal models, and factors associated with the metabolism of acetaminophen are considered.

2. The use of acetaminophen in babies and small children was not monitored as practice changed

To establish any associations between the use of acetaminophen in the pediatric population and the incidence of neurodevelopmental disorders, it is most convenient to establish the prevalence of both factors through time with some degree of certainty. To evaluate what is known about the prevalence of use of acetaminophen in the pediatric population at different points in time and in different locations, a systematic review was conducted as described in **Figure 1**. Although 48 studies were identified that evaluated the extent of acetaminophen use in babies and in children under 6 years of age, it is difficult to establish a coherent picture of exactly how much acetaminophen was used historically, and when or where exactly practice changed. Data were found for 38 countries, but data from 14 of those countries were limited to the "International Study of Asthma and Allergies in Childhood" (ISAAC) study (12) during 2000-2003. Further, in four countries where the ISAAC study was not the only study conducted, the results from the ISAAC study deviated by an average of 26.5% from other studies in that country. In Hungary and Portugal, the ISAAC results were higher than results from other sources, whereas in New Zealand and Spain, results were lower than from other sources. In addition, results from the Danish National Birth Cohort (DNBC) (13) were also in disagreement with independently conducted work, with approximately 10% using acetaminophen during the first 18 months of life as reported in the DNBC (14), contrasting with 65% using the drug within a three month period in an independent study evaluating a subset of the population covered by the DNBC (15). Further, data from the "Avon Longitudinal Study of Parents and Children" (ALSPAC) study in England (16) were not reported consistently, with use of acetaminophen in babies from 0-6 months during 1991-1992 described as 6% (17) and 84% (18), depending on the report. Moreover, data from more than a single, independent study was found only for 11 of the 38 countries, and three or more studies were

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found in only 5 countries. **Figure 2** shows the results from the five countries (United States, Italy, New Zealand, Norway, and Spain) in which at least three independent studies have evaluated the use of acetaminophen in babies and children under 6 years of age. Although numerous studies were conducted in various countries starting in the late 1990s, trends through time are not evident, and results varied considerably. This situation creates difficulty in correlating changes in neurodevelopmental disorders with changes in medical practice. However, as discussed in the next section, key historical events that affected acetaminophen use in the pediatric population are documented, and these can be used as surrogate markers for purposes of identifying correlations with neurodevelopmental disorders.

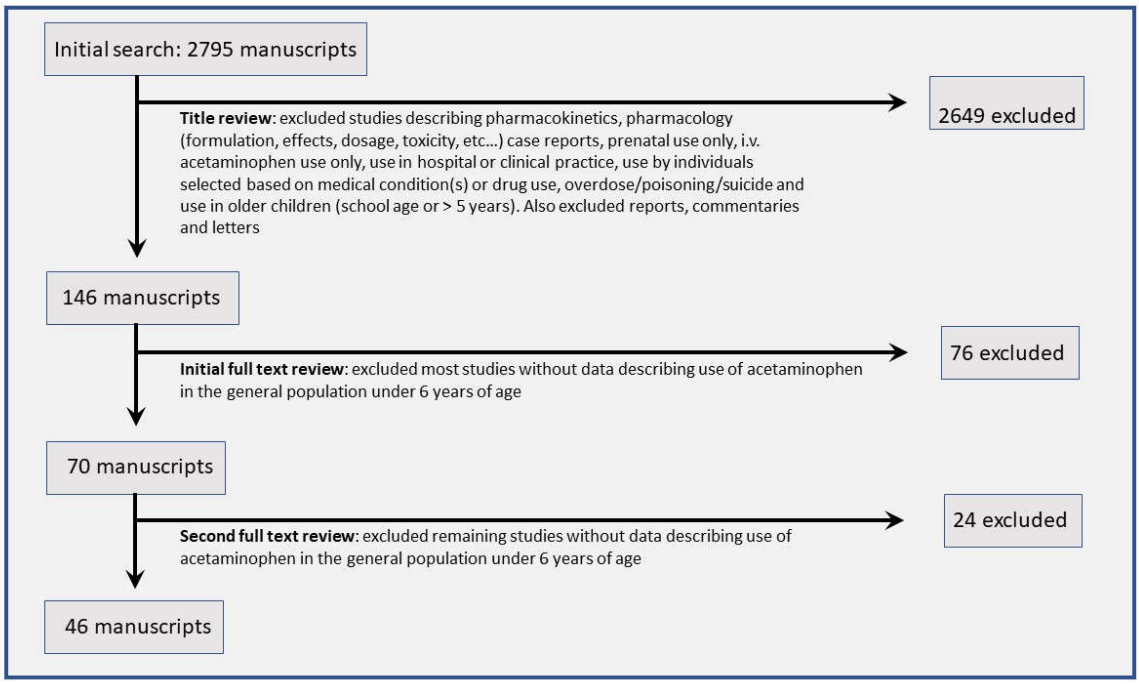

Figure 1. Systematic search for studies tracking acetaminophen use in the general population 5 years of age or less. The initial search was conducted on PubMed on August 25, 2022 without any restrictions on time frame. The search terms used were (acetaminophen or paracetamol) + (use or administration) + (infant or child or postnatal or pediatric or neonate or newborn or baby) - (review or mouse or mice or rat). The initial title review was conducted by co-author WP. The initial full text review was conducted by co-author LZ, and the second and final full text review was conducted by co-authors LZ and WP.

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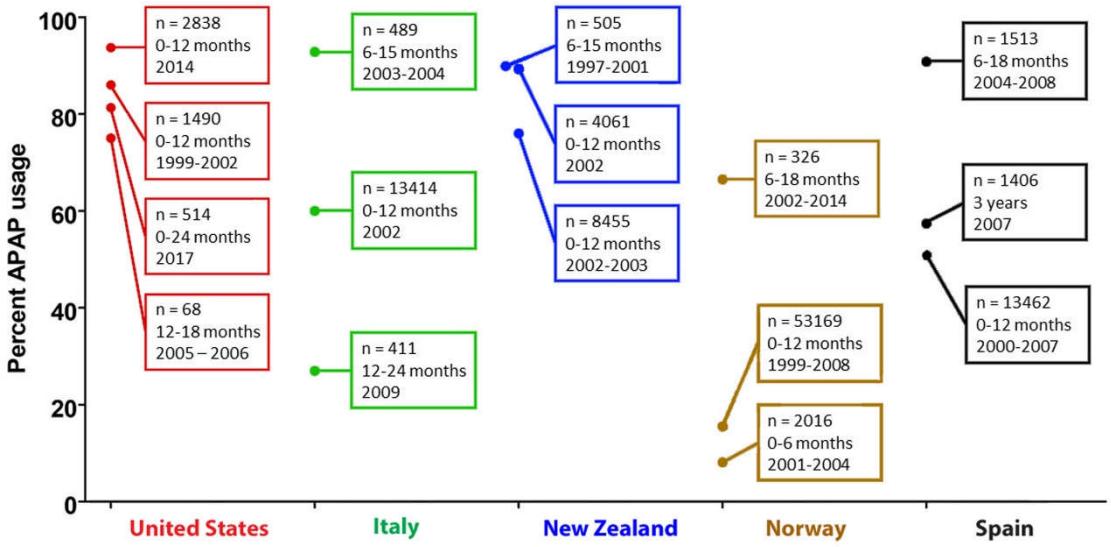

Figure 2. Variation in studies probing the use of acetaminophen (APAP) in babies and children under 6 years old. Results are shown for all 5 countries in which at least 3 studies using independent data sets have been conducted evaluating the use of acetaminophen in babies and children under 6 years of age. In cases where two studies used the same data set, the results are presented together. The number of babies/children in the study, the age of the babies/children at the time of acetaminophen use, and the years in which acetaminophen use was measured are shown in the box attached to each data point. The lowest value shown for the country of Spain is the average of three similar values (49.1%, 51.4%, and 52.0%) from three studies using the ISAAC in Spain data, two evaluating data from 2000-2003 (68, 69) and one evaluating data from 2006-2007 (70).

3. Associations between the incidence of ASD and early life exposure to acetaminophen.

At least three of the numerous circumstantial lines of evidence (1) pointing toward a causal role of acetaminophen in the induction of ASD involve the association through time between factors affecting use of acetaminophen in the pediatric population and changing prevalence of ASD (**Figure 3**). One of the three temporal relationships shown in Figure 3 entails an increase in the ratio of regressive versus infantile ASD beginning with children born after 1980 (19), coinciding with time that aspirin use in babies and children was being replaced by use of acetaminophen due to increasing awareness of the connection between aspirin and Reye Syndrome (20-22). This shifting ratio indicates that some factor was introduced into the population such that ASD could be induced even after brain development had proceeded on a relatively normal trajectory for a period of years.

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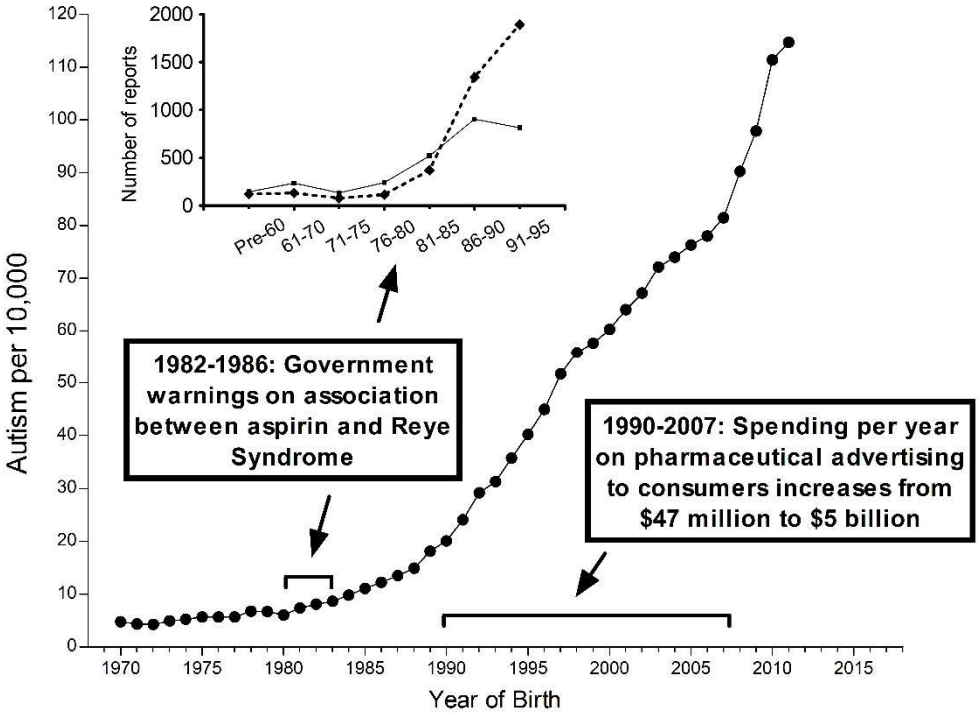

Figure 3. Temporal associations between the reported incidence of ASD (autism) in California and factors affecting the use of acetaminophen. The prevalence of ASD in California as compiled by Nevison (26) is shown in the graph. Data is a composite of ''snapshot'' data (information collected at one point in time) from the California Department of Developmental Services (covering birth years 1970–2011) (26). From 1982 to 1986, government warnings on using aspirin due to the association with Reye Syndrome were issued from the Centers for Disease Control and Prevention and the Food and Drug Administration (71). From 1990 to 2007, total spending on direct-to-consumer pharmaceutical advertising (DTCPA) underwent great increases, going from $47 million dollars in 1990 to $5 billion in 2007 (72). In the inset, previously published survey data (19) from the Autism Research Institute and the Autism Society of America are shown (19). The number of surveys that were collected within a given time frame are shown, and reports are separated into reports describing infantile (non-regressive or early-onset) ASD (solid line) and those describing regressive ASD (dashed line). The information in this diagram does not take into account increases in use of glutathione-depleting compounds such as pesticides and plastic-associated chemicals that have occurred during the time frame shown. Given that oxidative stress is a co-factor in the induction of acetaminophen-induced neurodevelopmental issues (1, 2, 32), such factors are expected to influence the incidence of ASD (32).

A second and distinct temporal relationship described in Figure 3 involves the beginning of the rise of the incidence of ASD in the early 1980s, coinciding again with replacement of aspirin in babies and in children with acetaminophen due to concerns over Reye Syndrome. Although it has been argued that aspirin was replaced by ibuprofen rather than acetaminophen in the US in the early 1980s (23), this argument is contradicted by available data, which demonstrates that acetaminophen was a drug of choice in the US when the pediatric use of aspirin was dramatically reduced (see, e.g., Rahwan and Rahwan (20), Arrowsmith et al. (21), and results of the systematic review shown in Figure 2). Further, as pointed out by Saugstad (24), ibuprofen for children was not approved as a prescription drug in the US until 1989, more than 30 years after a children's formulation of acetaminophen was marketed. Finally, children's ibuprofen was not approved for over-the-counter use until 1995 (25), long after the measured incidence of ASD began to rise (**Figure 3**).

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A third temporal correlation is shown in **Figure 3**: The rate of ASD has continued to climb as direct-to-consumer advertising in the US increased dramatically and then became a part of the US culture. Here, however, it should be noted that the actual use of acetaminophen in the pediatric population was poorly tracked, as discussed above. Trends in use through time are complicated by multiple means of acquiring the drug: through administration by physicians in clinics and hospitals and by caregivers using over-the-counter formulations at home. Thus, while changes in the quantity and qualitative nature of ASD coincide with major events affecting the pediatric use of acetaminophen (Figure 3), the exact pattern of change in acetaminophen use through time cannot be accurately ascertained from the literature. Nevertheless, it is apparent that use of acetaminophen in babies and in young children, a relatively uncommon occurrence half a century ago, is now more common than not.

One potential argument that acetaminophen cannot cause ASD maintains that rising rates of ASD through time associated with acetaminophen use are, at least in part, a consequence of changing diagnostic criteria, increased awareness, and other factors (discussed by co-author CDN and colleagues (26)). Based on this argument, it has been concluded by some that no chemical can conceivably account for the increased rates of ASD (27, 28). However, a careful analysis of epidemiologic evidence strongly suggests that the perceived rise in ASD since 1980 is real, at least in part, and not due entirely to artificial inflation (26). Further, the view that actual increases in the incidence of ASD are not real cannot readily account for the changing ratio of regressive to infantile ASD observed in the early 1980s (**Figure 3**). Perhaps more importantly, disparities in the prevalence of ASD measured in side-by-side cohorts (29, 30) demonstrate that some environmental factor or factors do, at least under some circumstances, play a pivotal role in the induction of ASD (1). Finally, epidemiologic evidence is only one factor among others which points toward a causal role of early life exposure to acetaminophen in the induction of neurodevelopmental disorders (1, 2).

Other objections to the conclusion that early life exposure to acetaminophen causes ASD in susceptible children include the fact that associations between rates of neurodevelopmental disorders and increased exposures to acetaminophen do not prove causation (31). While the fact that association does not prove causation is both undeniable and widely appreciated, it is also true that (a) multiple associations coupled with additional, independent evidence support causation, and (b) causation cannot exist without association. However, temporal associations in this case are complicated by several facts. For example, as pointed out above, the actual use of acetaminophen in the pediatric population was not tracked well through time. In addition, factors affecting oxidative stress, the necessary co-factor in acetaminophen-induced neurological injury discussed in detail below, may be changing through time (32). Further, the idea that the medical establishment and society in general might need to recalibrate diagnosis and awareness for a rapidly increasing incidence of cognitive disfunction seems reasonable if not expected. Such recalibration could account for short term shifts in data concerning the incidence of ASD. However, it seems implausible to attribute a dramatic, steady, 40-year climb in incidence to such factors. Indeed, ASD, although known by other labels through time (33), has consistently been distinguished by a deficit in social awareness (34), and was viewed as rare by the very knowledgeable individuals in the US and in Europe who discovered the condition more than 80 years ago (35, 36).

4. Studies in humans probing the association between early-life exposure to acetaminophen and ASD.

A very limited number of studies have attempted to ascertain the association between early life (post-partum) exposure to acetaminophen and ASD in humans. Notably, the recent study by Alemany and colleagues (17) observed an increase in ASD associated with use of acetaminophen in the DNBC. The analysis showed an unacceptably large odds ratio (1.30) for a commonplace occurrence (prenatal acetaminophen exposure), indicating

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that prenatal exposure to acetaminophen reported in the study accounts for a substantial quantity of cases of ASD. But, despite having a database with more than 60,000 children, the degree of uncertainty ranged from an odds ratio of 1.02 (clinically insignificant) to 1.66 (intolerable by any standard). Thus, it is not possible to draw any firm conclusions from Alemany's study on the importance of acetaminophen in the pathogenesis of ASD. We have previously demonstrated that the common use of the drug in babies and children without oxidative stress (and thus not at risk for acetaminophen-associated neurodevelopmental problems) will interfere with multivariate analyses such as the one performed by Alemany and colleagues, resulting in (a) underestimation of the impact of acetaminophen on the incidence of ASD, and (b) a lack of statistical power, leading to confidence intervals that are too large to draw conclusions (1). Since the lack of reliability of the multivariate analysis in this context has been dealt with in some detail previously (1), it will not be discussed here. An additional problem with analysis of data obtained from databases such as the DNBC is evident from the systematic review described above. That review casts doubt on the reliability of information pertaining to acetaminophen use in large databases, a factor that could adversely affect the reliability of the results obtained from analysis of the data. Thus, results from multivariate analyses of large data sets do not provide any valid basis for asserting that early life exposure to acetaminophen might be safe for neurodevelopment.

The first study to indicate that pediatric use of acetaminophen is associated with ASD was a survey-based, case-controlled study published by Stephen Schultz (37), a physician who saw his son regress into ASD following a vaccination (38). In that study, Schultz and colleagues noted that acetaminophen use with vaccination was associated with ASD if caregivers administered acetaminophen. In cases where the caregivers did not administer acetaminophen, no significant association with ASD was found. The odds ratios for ASD diagnosis following acetaminophen exposure were quite striking, depending on the comparisons made, with ratios exceeding a factor of 20-fold in some cases (37). Although the study by Schultz and colleagues was small, the results were persuasive, and comprise one piece of evidence in the case identifying early life exposure to acetaminophen as a cause of ASD (1).

Several criticisms of the Schultz study have been published, some of which are easily dismissed. For example, one objection was that Schultz and colleagues did not "estimate a sample size required for a study of this nature (a survey study)" (39). As pointed out correctly by Schultz in response (40), given that calculations of the sample size for a study require some foreknowledge of the size of the expected effect, the sample size required could not have possibly been calculated. The fact that comparisons were statistically significant does in fact demonstrate that the sample size was adequate, of course.

The most common objection to the Schultz study is that the selection of subjects from internet groups produced a "biased sample" (31, 39). The supposition that Schultz's study is undermined by bias among the participants may be why the study never affected clinical practice, never stimulated follow-up studies, and has been omitted more than once when critically considering the role of acetaminophen in neurodevelopmental outcomes following acetaminophen exposure (23, 41). Given the potential importance of the Schultz study, it is worth close examination of potential bias in the cohort he examined. The cohort was recruited from two internet-based groups in 2005 and 2006, after both Wakefield (42) and Rimland (19) had suggested that vaccines might cause ASD. Further, bias that vaccines cause ASD has persisted in parents of children with ASD (43, 44), so it seems highly likely that the parents in the Schultz study were biased in favor of the view that vaccines can induce ASD.

In contrast to biases related to vaccines, a review of the literature at the time suggests that bias probably did not exist favoring the view that early life exposure to acetaminophen causes ASD in susceptible children; A PubMed search for the terms "paracetamol or acetaminophen" and "autism" reveals only 4 papers prior to 2006. None of the 4 papers suggested that acetaminophen might cause ASD. For example, the initial study by Antonino Alberti's group in Italy showing profound impairment of acetaminophen

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metabolism in children with ASD (45) had been published in 1999, several years prior to the Schultz study. However, Alberti's study did not in any way suggest that exposure to acetaminophen might cause ASD. Further, Alberti's study was cited in PubMed indexed journals only three times prior to 2006 (46-48), all within the context of understanding the physiology of ASD, not the cause. Alberti's paper was cited by the Alternative Medicine Review (not PubMed indexed) in 2002 (49), and acetaminophen was listed by the author as a potentially neurotoxic compound in children with oxidative stress. However, the concern regarding acetaminophen occupied only one line in a 25-page report that included a pages-long discussion of the potential role of vaccines and vaccine components in the induction of ASD. Then, in 2003, Anthony Torres at Utah State University suggested that use of antipyretics in general may lead to ASD (50), but the hypothesis was that the absence of fever rather than the presence of acetaminophen might be the problem. That paper was not cited in the literature until 2009, and was, interestingly, cited in the context of the potential importance of vaccines, not acetaminophen, in the etiology of ASD (51). Further, coauthor WP has been actively engaged with the community of parents of children with ASD, and has observed that few parents, even in the past 5 years, have been aware of the view that early life exposure to acetaminophen can cause ASD in susceptible babies and children.

Thus, it seems highly likely that the parents surveyed in the Schultz study (37) were indeed biased in favor of the idea that vaccines cause ASD, but is seems exceedingly unlikely that they had a similar bias with acetaminophen. Indeed, as Schultz explained, "*The hypothesis that APAP (acetaminophen) causes ASD was completely unknown to the parents being surveyed. In fact, my study conducted in 2005 and 2006 was the first to explore this hypothesis*." (Personal communication to co-author WP, used with written permission.) It has been suggested that parents with ASD might try harder to recall information while in search for answers (31), but studies probing this issue have not found that parents of children with adverse outcomes have better recall (52). Perhaps more importantly, the data provided by Schultz do not suggest that parents of children with ASD had better recall than parents of neurotypical children. The Schultz study used yes or no questions, and the response rate to particular questions could be taken, at least in part, as a surrogate indicator of recall. Using response rate as a metric, Schultz's data reveal no evidence that parents of children with ASD have better recall than parents of neurotypical controls (37). For example, the answer rate was 100.0% for cases and controls asked about their child's acetaminophen use in conjunction with vaccines, 83.1% and 85% for cases and controls, respectively, when asked about their child's acetaminophen use between 12 and 18 months of age, and 59.0% and 72.5% for cases and controls, respectively, when asked about their child's exposure to ibuprofen between 12 and 18 months of age. Further, Schultz specifically addressed the issue of recall, independently analyzing surveys with greater time lapse since the events in question (37). As correctly described by Schultz (40), the results were robust and did not indicate that time had affected the outcome.

With the above discussion in mind, the conclusions of the Schultz study can be amended: In cases where the parents are likely biased toward the view that vaccines cause ASD, exposure to acetaminophen rather than vaccines was likely a factor in the induction of ASD in their child. Further, it is apparent that dismissal of the study due to bias is unwarranted and not supported by any available information. Thus, the Schultz study (37) contributes to the body of abundant circumstantial evidence pointing toward use of acetaminophen as one cause of neurodevelopmental injury in susceptible babies and children (1).**5. Clues from the metabolism of acetaminophen pointing toward problems with acetaminophen toxicity in children with ASD.**

The metabolism of acetaminophen is extremely well characterized, and provides considerable insight into how acetaminophen can cause neurodevelopmental injury (32, 53). The human body processes acetaminophen via three pathways (**Figure 4**). Two of these pathways involve the addition of highly water-soluble structures, either glucuronate via the glucuronide pathway, or sulfate via the sulfation pathway. In adults, the addition of

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glucuronate predominates over the addition of sulfate (54), whereas in babies and in children under the age of 9 years, the addition of a sulfate predominates over the addition of glucuronate (54, 55). The third pathway also involves addition of a highly water-soluble molecule (glutathione), but the first step in this pathway involves production of a highly toxic substance, N-acetyl-p-benzoquinone imine (NAPQI). Fortunately, in healthy individuals, NAPQI is rapidly neutralized by glutathione (**Figure 4**). Unfortunately, children with ASD tend to have an impaired ability to utilize the sulfate pathway (45, 56, 57). In addition, children with ASD tend to have oxidative stress (2, 58), a factor that depletes glutathione (56). Further, acetaminophen exposure itself depletes glutathione to a very significant degree (59), suggesting that repeated exposure to the drug is potentially more hazardous than a single exposure. Another factor that makes matters worse is that almost three quarters of children with ASD have autoantibodies that block folate transport to the brain (60). Since folate is necessary for the synthesis of glutathione, an impaired ability to detoxify NAPQI is expected in those children. Thus, many children with ASD have two impaired pathways for clearance of acetaminophen. It has been speculated that the third pathway, glucuronidation, might compensate for the problem (31). However, the glucuronidation pathway is not apparently upregulated by repeated exposure to acetaminophen (61), and is a minor pathway in babies and children, as discussed above (54, 55). Further, since some NAPQI is created regardless of the function of the other two pathways, failure in the glutathione-dependent pathway is expected to result in accumulation of NAPQI and subsequent toxicity even if the other two pathways are functional.

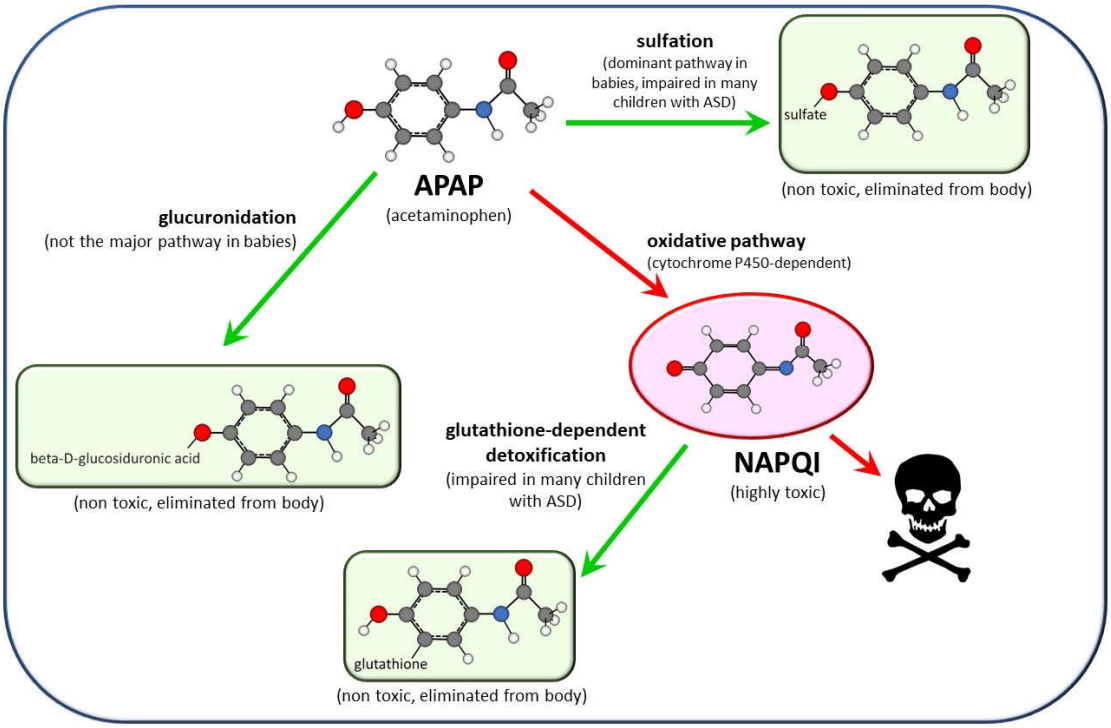

Figure 4. Metabolism of acetaminophen in humans. The three pathways, (a) glucuronidation, (b) sulfation, and (c) oxidation followed by reaction with glutathione are shown. The major pathway in babies and in children, sulfation, tends to be impaired in children with ASD. This is expected to shunt more of the drug through the oxidative pathway, resulting in production of excess NAPQI, the toxic compound shown in the diagram. Unfortunately, children with ASD also tend to have a reduced ability to detoxify NAPQI, resulting in an increase in the toxicity of acetaminophen due to excess NAPQI.

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It is probably not surprising that both sulfation and glutathione-dependent pathways are aberrant in the same population, since these pathways are metabolically connected (32, 62, 63). Alterations in both of these pathways enhance oxidative stress, increasing the toxicity of acetaminophen. Unfortunately, even at levels of acetaminophen that are currently considered acceptable, this situation will result in exposure of some babies and children to levels of acetaminophen toxicity that are much greater than would be seen in typical, non-susceptible individuals or in laboratory animals (**Figure 5**).

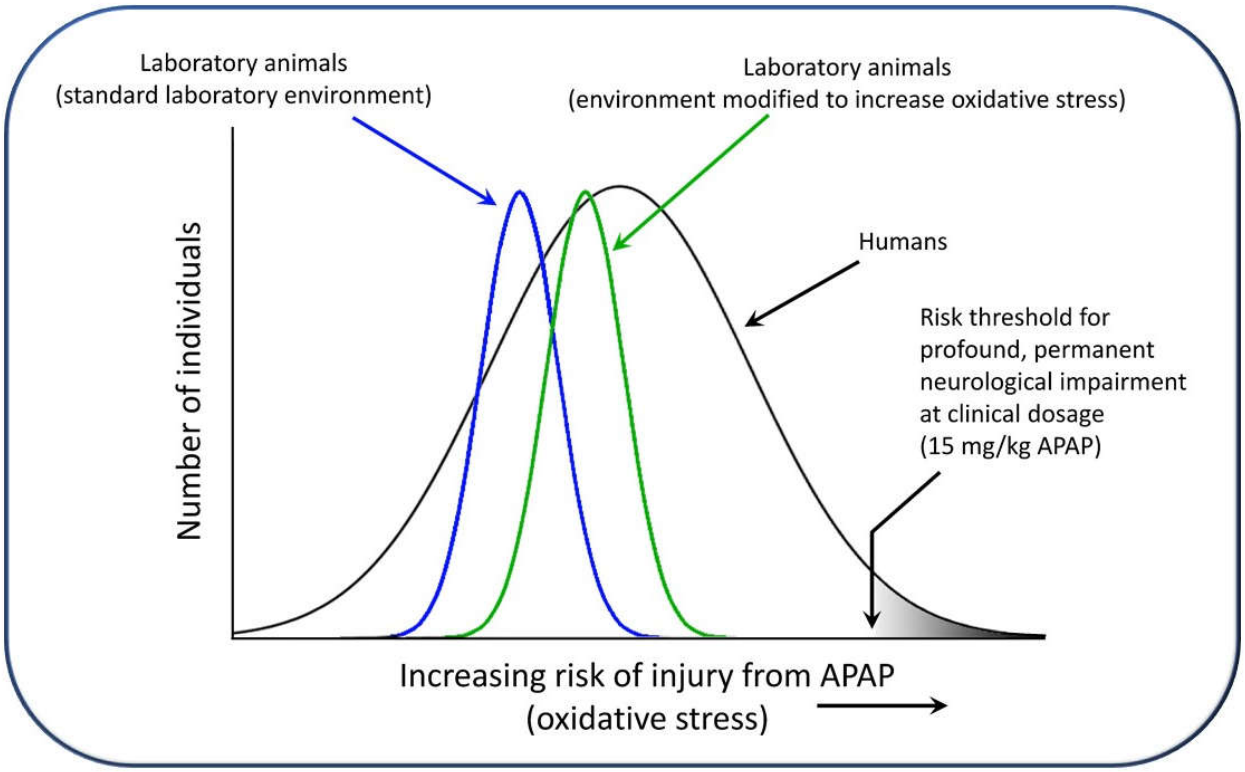

Figure 5. Schematic diagram illustrating relative sensitivities to laboratory animal pups and of human infants and children to acetaminophen-induced neurodevelopmental injury. The diagram illustrates how laboratory conditions can be modified to enhance oxidative stress, thus increasing the sensitivity of the animals to acetaminophen-induced neurodevelopmental injury. The schematic diagram illustrates that the sensitivity of healthy laboratory pups to acetaminophen-induced neurodevelopmental injury is relatively homogenous and less broadly distributed than that of human babies and children. Further, the diagram illustrates that the sensitivity of healthy laboratory animal pups to acetaminophen-induced neurodevelopmental injury is less in magnitude than that of at-risk human babies and children. In this model, exposures of laboratory animals can be made comparable to exposures in at-risk human babies and children by either (a) increasing the dose of acetaminophen in the laboratory pups, or (b) increasing oxidative stress in the laboratory pups. Quantitative estimates of the difference in the risks between laboratory animal pups and human babies and children have not been made, and the schematic diagram is not meant to indicate quantitative values.

6. Conclusions and future directions

In this narrative review and in our previous narrative reviews on the safety of pediatric acetaminophen use, we address several lines of evidence that might be considered controversial. We believe that considering multiple lives of evidence is necessary given complexities particular to this topic. For example, a recent systematic review and meta-analysis by Tan and colleagues at the University of Auckland, considering almost 20 studies and quarter of a million children less than 2 years old, raised no substantial flags concerning safety of early life exposure to acetaminophen (64). Unfortunately, based on the approach used in the study by Tan and colleagues, the results obtained are to be expected

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regardless of whether early life exposure to acetaminophen is responsible for most cases of ASD. Tan and colleagues note that exposure rates to acetaminophen in the pediatric population now approach 95%, a factor which will preclude identification of acetaminophen as a causative agent in neurodevelopmental disorders using a multivariate analysis of large data sets (1). In addition, consistent with our recent results (3), Tan and colleagues note that measures of adverse outcomes were limited to acute events rather than neurodevelopmental outcomes. As previously discussed, other factors impede the usefulness of such analyses, including the need for long term monitoring of exposures from the time of conception, an inability to separate confounding factors from oxidative stress-inducing cofactors, and the use of intravenous formulations of acetaminophen containing the antidote for acetaminophen toxicity in some studies. Indeed, an evaluation of the effect of early life exposure to acetaminophen on neurodevelopment outcomes would require a substantial effort that is unlikely to occur in the near future, as previously discussed (3).

Studies in animal models are, at present, sufficient to conclude that early life exposure to acetaminophen causes neurodevelopmental problems (3). The observation is robust, encompassing both laboratory rats and mice and a variety of study designs (see review by Patel and colleagues (1) and recent studies from the University of New Orleans (65, 66)). However, studies have yet to recapitulate symptoms of ASD, and this remains a highly laudable goal of research in the field. Although it has been argued that "clinically relevant" doses of acetaminophen should be used in such studies, it is expected that recapitulating conditions in susceptible humans using healthy laboratory animals will necessarily require higher doses of drug than those commonly encountered by humans (**Figure 5**). To summarize, it is expected that laboratory rats under ideal laboratory conditions will be more resistant to acetaminophen-induced neurodevelopmental injury than are humans with significant problems in metabolizing the drug. Not only are laboratory rats bred to be healthy under standard laboratory conditions, thus potentially reducing genetic factors that might make them susceptible to disease, but they are fed an exceedingly healthy diet (1) and are often largely free of the infections, environmental toxins, and other oxidative stress factors associated with ASD in humans. Indeed, current regulations take this into account, stipulating that preclinical testing should include higher doses of drug than those expected to be encountered by patients (67).

The failure of the medical community to accurately track the use of acetaminophen in the pediatric population through time as well as the almost ubiquitous use of the drug found in some studies (Figure 2) reflect a high degree of acceptance of the drug. The incorrect assumption that babies react to acetaminophen in a manner similar to adults was a key factor in the current level of acceptance of the drug (3). However, other factors undoubtedly contributed. For examples, (a) critical studies in laboratory animals were conducted only recently, (b) most babies and children suffer no apparent, serious adverse neurodevelopmental effects from acetaminophen, (c) severe adverse neurodevelopmental effects may not be diagnosed until long after drug exposure, (d) the litany of oxidative stress-inducing co-factors in the induction of acetaminophen-induced neurodevelopmental injury creates a large and potentially confusing number of associations with the neurodevelopmental injury, and (e) any severe, adverse neurodevelopmental effects might be attributed to the reason for taking the drug.

At the present time, most clinicians and caregivers have not been informed of available knowledge concerning the apparent adverse reactions to early life acetaminophen exposure in susceptible children. The conduct of large, long-term studies in human children may not be feasible, as discussed above. This point, however, may be irrelevant given that the preponderance of available evidence renders such a study unnecessarily risky and thus unethical. With this in mind, regulatory agencies and professional medical societies should move forward with currently available information, first acknowledging and then promoting awareness of the problem. Changes to medical practice should be implemented which effectively weight the risks with the benefits of the drug. Failure to move forward with changes to medical practice at the present time constitutes a disregard for ample evidence of harm despite the absence of any valid rationale for the view that

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acetaminophen might be safe for neurodevelopment. Finally, the ability of antidotes for acetaminophen toxicity such as N-acetylcysteine to prevent acetaminophen-induced neurodevelopmental injury could be probed.

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