Acetaminophen causes neurodevelopmental injury in susceptible babies and children: no valid rationale for controversy.

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Abstract: Despite worldwide acceptance of acetaminophen as a necessary medicine in the field of pediatrics, evidence that early life exposure to acetaminophen causes neurodevelopmental injury in susceptible babies and 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. Evidence from the prepartum and the postpartum period is considered, avoiding controversies raised by considering only the limited evidence pointing exclusively toward risks during the prepartum period. Among other issues, 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. In addition, problems with exclusive reliance on results from meta-analyses of large datasets and from studies involving small time frames of drug exposure are reviewed. Further, 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; neurodevelopment; paracetamol

Key message: Despite worldwide acceptance in pediatric medicine, careful examination reveals no valid objections to the conclusion, based on extensive evidence, that early life exposure to acetaminophen (paracetamol) causes neurodevelopmental injury in susceptible babies and children. Nevertheless, some debate has centered around the prenatal period, where evidence is relatively limited compared to evidence pointing toward the postnatal period as the greatest time of absolute and relative risk.
Graphical abstract
1. Introduction. Mounting evidence for the induction of neurodevelopmental injury by early life exposure to acetaminophen.

Acetaminophen (paracetamol) as well as the antidote for acetaminophen overdose (N-acetylcysteine) are listed by the World Health Organization as essential medicines for children (1). Despite worldwide acceptance of the drug in pediatric medicine, 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 (2). A summary of this evidence is shown in Figure 1 and in Table 1. Evidence demonstrates that, while most babies and children are relatively unharmed by exposure to the drug, some babies and children are at risk due to the presence of oxidative stress (2, 3). Evidence points in particular toward induction of autism spectrum disorder (ASD), with possible connections to both developmental delays and attention deficits (2). Further, evidence points toward exposure between birth and approximately 5 years of age as being the period of highest risk, with the risk during prenatal exposure being significant in numerous studies (4), but less consequential (2, 3). Much of this evidence has been reviewed in detail recently (2, 3), and will not be reviewed in detail again here.

A recent, exhaustive review of the literature, complete with citation tracking, demonstrated that, within the medical profession, acetaminophen is widely assumed to be safe when used as directed in the pediatric population (5). 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 (5). Indeed, in adults, the liver was found to be the target of acetaminophen toxicity during the 1960s (6-8). At that time, however, the view that babies metabolize drugs in a manner identical to adults was already known to be a dangerous assumption (9); this knowledge has yet to be applied to the toxicity of acetaminophen in human children (5). More than a decade later, a study using laboratory animals demonstrated that this assumption should not be applied to the metabolism of acetaminophen (10). 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 (10), a finding that has been recently verified (11). 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 (12). 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 (13).
A recent summary statement by Bauer and colleagues examined the potential role of acetaminophen exposure *during pregnancy* in the induction of neurodevelopmental problems (14). That summary statement called for increased awareness of a potential role of acetaminophen in the induction of neurodevelopmental problems, but was criticized heavily by the American College of Obstetricians and Gynecologists (ACOG). In their response (15), the ACOG concluded that available studies “show no clear evidence that proves a direct relationship between the prudent use of acetaminophen during any trimester and fetal developmental issues”. The ACOG further concluded that “physicians should not change clinical practice until definitive prospective research is done”. In considering the response of the ACOG, it is important to note that evidence related to the induction of neurodevelopmental problems by acetaminophen during the prenatal period is concerning but yet somewhat limited (2, 16, 17). As shown in Table 1, only one line of evidence out of a total of 20 relates specifically to the prenatal period. All of the other 19 lines of evidence are consistent with involvement of the postnatal period and, in some cases, indicate that the postnatal period is the time of greatest sensitivity to acetaminophen-induced neurodevelopmental injury. The relative safety of the prenatal period compared to the postnatal period is perhaps not surprising given the the particularly efficient metabolism of acetaminophen by the mother during pregnancy (18) and the limited capacity of neonates to metabolize pharmaceuticals (19). Unfortunately, however, considerable public debate concerning acetaminophen-induced neurodevelopmental problems has focused on the Bauer consensus statement involving the prenatal period (14) and is fueled by ongoing lawsuits involving prenatal use of the drug (20). Thus, although substantial controversy currently surrounds the view that acetaminophen causes neurodevelopmental injury in susceptible individuals, this debate and the controversy surrounding it are primarily focused on relatively limited evidence pointing specifically toward the prenatal period. Tragically, the public debate has not yet moved toward postnatal use of the drug, where relative and absolute risks are greater and evidence of neurodevelopmental injury is strong to the point of being conclusive (2).

As shown in Table 1, evidence pointing toward the conclusion that early life exposure to acetaminophen causes neurodevelopment injury in susceptible babies and children during the postnatal period is much more robust than evidence limited to prenatal exposure only (Table 1). The extensive evidence pointing at neurodevelopmental injury in the postnatal period has not been directly challenged, although the conclusion that the drug is hazardous for neurodevelopment might still be considered controversial by some. Indeed, objections to some of the evidence described recently (2) and summarized in Figure 1 and in Table 1 has been voiced. Herein we review several issues that may be considered controversial in the field, considering each line of evidence independently but also as a component of the whole. 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 2A. 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 (21) 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) (22) 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 (23), 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 (24). Further, data from the “Avon Longitudinal Study of Parents and Children” (ALSPAC) study in England (25) were not reported consistently, with use of acetaminophen in babies from 0-6 months during 1991-1992 described as 6% (26) and 84% (27), 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 found in only 5 countries. Figure 2B 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.
3. Associations between the incidence of ASD and early life exposure to acetaminophen.

At least three of the numerous circumstantial lines of evidence (2) 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 (28), 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 (29-31). 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.

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 (32), 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 (29), Arrowsmith et al. (30), and results of the systematic review shown in Figure 3). Further, as pointed out by Saugstad (33), 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 (34), long after the measured incidence of ASD began to rise (Figure 3).

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 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 (35)). Based on this argument, it has been concluded by some that no chemical can conceivably account for the increased rates of ASD (36, 37). 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 (35). 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 (38, 39) demonstrate that some environmental factor or factors do, at least under some circumstances, play a pivotal role in the induction of ASD (2). 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 (2, 3).

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 (40). The fact that association does not prove causation is both undeniable and widely appreciated. However, at the same time, causation cannot exist without association, and multiple, independent associations coupled with other lines of independent evidence support causation. 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 (41). 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 dysfunction seems reasonable if not expected. Such recalibration could account for short term shifts in data concerning the incidence of ASD. Nevertheless, 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 (42), has consistently been distinguished by a deficit in social awareness (43), and was viewed as rare by the very knowledgeable individuals in the US and independently in Europe who discovered the condition 80 years ago (44, 45).

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 (26) 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 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 (2). Since the lack of reliability of the multivariate analysis in this context has been dealt with in some detail previously (2), 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 (46), a physician who saw his son regress into ASD following a vaccination (47). 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 (46). 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 (2).

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)” (48). As pointed out correctly by Schultz in response (49), 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” (40, 48). 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 (32, 50). 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 (51) and Rimland (28) had suggested that vaccines might cause ASD. Further, bias that vaccines cause ASD has persisted in parents of children with ASD (52, 53), 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, t(46)(46)(46)As correctly described by Schultz (49), 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 (46) contributes to the body of abundant circumstantial evidence pointing toward use of acetaminophen as one cause of neurodevelopmental injury in susceptible babies and children (2). 5. Clues from the metabolism of acetaminophen.

It is probably not surprising that both sulfation and glutathione-dependent pathways are aberrant in the same population, since these pathways are metabolically connected (41, 72, 73). 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).

As described above, the mechanisms by which acetaminophen induces cell injury and death are well established, and the fact that acetaminophen-induced cytotoxicity affects the developing brain is well established. However,
6. Discussion

In this narrative review and 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 lines 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 a quarter of a million children less than 2 years old, raised no substantial flags concerning safety of early life exposure to acetaminophen (74). Unfortunately, based on the approach used in the study by Tan and colleagues, the results obtained are to be expected 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 (2). In addition, consistent with our recent results (5), 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 neurodevelopmental outcomes would require a substantial effort that is unlikely to occur in the near future, as previously discussed (5).

Studies in animal models are, at present, sufficient to conclude that early life exposure to acetaminophen causes neurodevelopmental problems (5). The observation is robust, encompassing both laboratory rats and mice and a variety of study designs (see review by Patel and colleagues (2) and recent studies from the University of New Orleans (75, 76)). 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 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 also fed an exceedingly healthy diet (2) and are often largely free of 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 (77).

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 3) 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 (5). However, other factors undoubtedly contributed. For example, (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 present, 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 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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<table>
<thead>
<tr>
<th>Summary of evidence leading to the conclusion that early life exposure to acetaminophen in susceptible children causes neurodevelopmental injury</th>
<th>Relevance to prenatal versus postnatal exposure: nature of evidence</th>
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<tbody>
<tr>
<td>1. Laboratory mice and rats develop long-term brain damage and exhibit behavioral changes following early life APAP exposure at doses that are similar to or even less than doses received by human babies and children (12, 78-81).</td>
<td>Points toward the postnatal period more so than the prenatal period: Laboratory animal studies.</td>
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<td>2. In laboratory rats, APAP affects the developing male brain more than the female brain (80). ASD also affects males more than females (82).</td>
<td>Postnatal: Laboratory animal studies.</td>
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<tr>
<td>3. APAP causes death of cortical neurons in adult laboratory rats at concentrations lower than it causes liver failure (13). Affected cortical neurons are implicated in ASD (83, 84).</td>
<td>Postnatal: Laboratory animal studies.</td>
</tr>
<tr>
<td>4. Despite the fact that APAP targets the brain, APAP use in babies and children was only proven safe for acute side effects, not for neurodevelopment (5).</td>
<td>Postnatal: Systematic review of the literature.</td>
</tr>
<tr>
<td>5. Male circumcision, often performed using APAP as an analgesic, is associated with a dramatic increase in the risk for early-onset (infantile) ASD (39).</td>
<td>Postnatal: Association with human behavior.</td>
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<tr>
<td>6. An unexpectedly high prevalence of ASD was identified in South Korea (85, 86), where APAP-containing products for children were repeatedly found to contain amounts of drug exceeding the package label (87).</td>
<td>Postnatal: Association with human behavior.</td>
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<tr>
<td>7. Ultra-Orthodox Jews (88) and Arabs (88, 89) in Israel have a reported prevalence of ASD less than half of that of other Israelis. Israelis have high rates of circumcision concomitant with ritual use of alcohol. Alcohol use depletes glutathione, particularly in the brain (90), thereby increasing susceptibility to APAP-induced injury. Thus, use of traditional circumcision practices without APAP by some communities in Israel could account in part for their lower rates of ASD compared to other Israelis.</td>
<td>Postnatal: Association with human behavior.</td>
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<td>8. Analysis of 61,430 babies in the Danish National Birth Cohort found an odds ratio (OR) of 1.3 (CI 1.02-1.66) for ASD associated with postnatal APAP exposure (26). This result is especially concerning since heavy use of the drug among non-susceptible children will cause dramatic underestimation of the actual risk (2).</td>
<td>Postnatal use: Epidemiologic study with some control for indication.</td>
</tr>
<tr>
<td>9. The ratio of regressive to infantile ASD rose at the same time as pediatric APAP use rose (91) after aspirin was associated with Reye’s syndrome (3).</td>
<td>Postnatal: Temporal association.</td>
</tr>
<tr>
<td>10. The incidence of ASD began to increase in the early 1980s, coinciding with the increase in APAP use after aspirin was associated with Reye’s syndrome (3).</td>
<td>Postnatal: Temporal association.</td>
</tr>
<tr>
<td>11. The incidence of ASD has steadily increased (3) as direct-to-consumer advertising (92) and perhaps other factors have driven up use of pharmaceutical products.</td>
<td>Prenatal and postnatal: Temporal association.</td>
</tr>
<tr>
<td>12. Use of APAP in pregnant women is associated with long-term effects that include lower IQ, increased ASD, and increased ADHD (26, 93-105).</td>
<td>Prenatal use: Epidemiologic studies, some with controls for indication.</td>
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<tr>
<td>14.</td>
<td>APAP given alongside vaccine administration but not vaccination alone is associated with ASD (46).</td>
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<td>15.</td>
<td>Many parents believe that their children’s ASD was induced by a vaccine based on their own observations or the observations of trusted social networks. (52, 53) APAP is frequently used with vaccinations, although vaccinations alone do not cause ASD.</td>
</tr>
<tr>
<td>16.</td>
<td>APAP use during early childhood is associated with a dramatic increase in regressive ASD (46).</td>
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<tr>
<td>17.</td>
<td>APAP use in adults temporarily blunts social trust (106) and awareness (107), emotional responses to external stimuli (108), and the ability to identify errors (109), indicating that the drug targets regions of the brain affected in patients with ASD.</td>
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<td>18.</td>
<td>Cystic fibrosis is associated with unusually efficient (effective) metabolism of APAP (110, 111), and some evidence suggests that the prevalence of ASD may be very low in patients with cystic fibrosis (3).</td>
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<td>19.</td>
<td>Genetic and immune factors associated with an increased risk of ASD have a detrimental effect on the body’s ability to metabolize APAP (3, 54, 70).</td>
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<td>20.</td>
<td>APAP is known to be highly toxic in the presence of oxidative stress. The mechanism by which this toxicity occurs has been established for decades (112), and involves the formation of the potent toxin, NAPQI (113-115). More recent studies indicate that concomitant mitochondrial damage (116) is important in the process.</td>
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Table 1. Current evidence leading to the conclusion that early life exposure of individuals to acetaminophen (APAP) causes long-term neurodevelopmental problems. Whether the evidence applies to the prenatal period, the postnatal period, or potentially both is indicated.
**Figure 1.** Summary of evidence pointing toward the induction of neurodevelopmental disorders by early life exposure to acetaminophen (APAP). Numbering of individual lines of evidence are as in Table 1. References and more detailed descriptions of each line of evidence are listed in Table 1. The twenty lines of evidence are separated into 5 miscellaneous lines of evidence, with the remaining 15 lines of evidence divided evenly into 5 categories: Studies in animal models, associations with human activities, associations in time, postnatal observations, and molecular mechanism of action. Lines of evidence numbers 10 and 11 derive from the same data, and lines of evidence numbers 14 and 16 derive from the same study. These lines of evidence are therefore not independent, which is indicated by the connecting lines in the diagram. DNBC = Danish National Birth Cohort.
Figure 2. Studies tracking acetaminophen use in the general population 5 years of age or less. In diagram A, the systematic search strategy is shown. 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. In diagram B, variation in studies probing the use of acetaminophen (APAP) in babies and children under 6 years old is illustrated. 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 (117, 118) and one evaluating data from 2006-2007 (119).
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 (35) 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) (35). 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 (120). 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 (121). In the inset, previously published survey data (28) from the Autism Research Institute and the Autism Society of America are shown (28). 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 (2, 3, 41), such factors are expected to influence the incidence of ASD (41).
Figure 4. Metabolism of acetaminophen (APAP) 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 N-acetyl-p-benzoquinone imine (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.
Figure 5. Schematic diagram illustrating relative sensitivities to laboratory animal pups and of human infants and children to acetaminophen (APAP)-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.


87. Hall C, Smith M. Increased cGMP enforcement has gone international: South Korean action against Johnson & Johnson serves as warning. White Collar Watch. 2013;June.


Early life exposure of susceptible individuals to APAP causes neurodevelopmental injury
A

Initial search: 2785 manuscripts

Title review: excluded studies describing pharmacokinetics, pharmacology (formulation, effects, dosage, toxicity, etc.), case reports, prenatal use only, i.e., acetaminophen use only, use in hospital or clinical practice, use by individuals selected based on medical condition(s) or drug use, overdose/poisoning/suicide and use in older children (selected age or > 5 years). Also excluded reports, commentaries and letters

146 manuscripts

Initial full text review: excluded most studies without data describing use of acetaminophen in the general population under 6 years of age

70 manuscripts

Second full text review: excluded remaining studies without data describing use of acetaminophen in the general population under 6 years of age

46 manuscripts

B

Percent APAP usage

0 20 40 60 80 100

United States Italy New Zealand Norway Spain

n = 2838

6-12 months 2014

n = 1340

6-12 months 1999-2002

n = 68

12-24 months 2003-2006

n = 2849

6-15 months 2003-2004

n = 514

6-24 months 2002

n = 411

12-24 months 2009

n = 505

0-15 months 1997-2002

n = 6061

0-12 months 2002

n = 13414

0-12 months 2002

n = 326

6-18 months 2002-2006

n = 8455

0-12 months 2002-2003

n = 53569

0-12 months 1999-2008

n = 1016

0-6 months 2001-2004

n = 1533

6-18 months 2004-2008

n = 1406

3 years 2007

n = 13462

0-12 months 2000-2003
1982-1986: Government warnings on association between aspirin and Reye Syndrome

1990-2007: Spending per year on pharmaceutical advertising to consumers increases from $47 million to $5 billion
Glucuronidation (not the major pathway in babies)

APAP (acetaminophen)

Sulfation (dominant pathway in babies, impaired in many children with ASD)

Sulfate (non toxic, eliminated from body)

Oxidative pathway (cytochrome P450-dependent)

Glutathione-dependent detoxification (impaired in many children with ASD)

Beta-D-glucosiduronic acid (non toxic, eliminated from body)

Glutathione (non toxic, eliminated from body)

NAPQI (highly toxic)
Laboratory animals (standard laboratory environment)

Laboratory animals (environment modified to increase oxidative stress)

Humans

Risk threshold for profound, permanent neurological impairment at clinical dosage (15 mg/kg APAP)

Number of individuals

Increasing risk of injury from APAP (oxidative stress)