Limitations of the Columbia University Study of Children’s Environmental Health

Introduction

Researchers at Columbia University have published health-related claims based on information collected from a group of New York City mothers and their children born between 1998 and 2003. These investigators have sought to identify statistical correlations of various developmental characteristics in children with prenatal childhood exposure to environmental factors, including chemical exposures, as well as family and neighborhood status. The study estimated exposure to chlorpyrifos based on measurements in umbilical cord blood at birth and maternal blood near delivery. No samples were taken at gestational periods that would have the greatest developmental interest. The researchers reported that exposure to chlorpyrifos prior to birth can be linked to decreased body length and weight at birth, to later childhood developmental and behavioral issues, and to intelligence and brain anomalies.

Chlorpyrifos opponents have used the findings of this study to fuel arguments against the use and re-registration of chlorpyrifos.

At face value, the Columbia University research has a strong methodological design and raises serious questions, and further research into this area will most certainly be conducted. There are compelling reasons, however, why the reported outcomes of these papers are not likely to have been caused by chlorpyrifos:

- Inconsistency with similar studies in humans and with studies in animals.
- Exposures reported are low and are well below the no effect level in numerous other studies.
- Exposure was only estimated at one point in time.
- Internal evidence within the research raises concerns about the validity of reported findings.
- Alternative hypotheses may explain the observed results.

The weight of all the evidence (human and animal) does not support a cause and effect connection between current levels of chlorpyrifos exposure and adverse human effects.

Inconsistency with Similar Studies

Researchers at Mount Sinai Hospital and the University of California at Berkeley conducted similar research on human effects and prenatal exposures to chlorpyrifos. But the results of their research are inconsistent with the findings of the Columbia University researchers (see following tables). Taken together, the findings from these three groups of researchers do not show a clear link between chlorpyrifos exposure and child health. It should also be noted that the most recent Columbia University study, which employed brain scans, dealt with findings that have not yet been evaluated by any other study.
The reported outcomes of these evaluations are summarized as follows:

**RESULTS FOR FETAL GROWTH**

<table>
<thead>
<tr>
<th></th>
<th>Head Size</th>
<th>Body Weight</th>
<th>Body Length</th>
</tr>
</thead>
<tbody>
<tr>
<td>Columbia University</td>
<td>No Effect</td>
<td>Decreased</td>
<td>Decreased</td>
</tr>
<tr>
<td>University of California at Berkeley</td>
<td>No Effect</td>
<td>No Effect</td>
<td>No Effect</td>
</tr>
<tr>
<td>Mount Sinai Hospital</td>
<td>No Effect</td>
<td>No Effect</td>
<td>No Effect</td>
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</table>

*Note: Berkeley and Mount Sinai researchers noted no statistically significant decrease in fetal growth outcomes associated with exposures based on the chlorpyrifos breakdown product TCPy.*

**RESULTS FOR DEVELOPMENT**

<table>
<thead>
<tr>
<th></th>
<th>Psychomotor Development</th>
<th>Mental Development</th>
</tr>
</thead>
<tbody>
<tr>
<td>Columbia University</td>
<td>Decreased test scores (36 months)</td>
<td>Decreased test scores (36 months)</td>
</tr>
<tr>
<td>University of California at Berkeley</td>
<td>No Effect (6, 12, 24 months)</td>
<td>No Effect (6, 12, 24 months)</td>
</tr>
<tr>
<td>Mount Sinai Hospital</td>
<td>No effect (12, 24 months)</td>
<td>No effect (12, 24 months)</td>
</tr>
</tbody>
</table>

*Note: Berkeley and Mount Sinai researchers found no statistically significant decrease in development with exposures based on the chlorpyrifos breakdown product TCPy or DEP.*
### RESULTS FOR BEHAVIOR

<table>
<thead>
<tr>
<th></th>
<th>ADHD</th>
<th>Attention Problems</th>
<th>Pervasive Developmental Disorder</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Columbia University</strong></td>
<td>Increased (3 years)</td>
<td>Increased (36 months)</td>
<td>Increased (36 months)</td>
</tr>
<tr>
<td><strong>University of California at Berkeley</strong></td>
<td>No Effect (2, 3.5 years)</td>
<td>No Effect (2, 3.5, 5 years)</td>
<td>No Effect (2 years)</td>
</tr>
<tr>
<td></td>
<td>Conflicting (Increased in 2 of 5 analyses at 5 years)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Mount Sinai Hospital</strong></td>
<td>Not tested</td>
<td>Not tested</td>
<td>Not tested</td>
</tr>
</tbody>
</table>

*Note: Berkeley researchers found no consistent increase in adverse behavior with exposures based on the chlorpyrifos breakdown product TCPy or DEP.*

### RESULTS FOR INTELLIGENCE

<table>
<thead>
<tr>
<th></th>
<th>Working memory</th>
<th>Full Scale IQ</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Columbia University</strong></td>
<td>Decreased test scores (7 years)</td>
<td>Decreased test scores (7 years)</td>
</tr>
<tr>
<td><strong>University of California at Berkeley</strong></td>
<td>No Effect (7 years)</td>
<td>Conflicting (1 of 2 analyses decreased)</td>
</tr>
<tr>
<td><strong>Mount Sinai Hospital</strong></td>
<td>No effect (6–9 years)</td>
<td>No effect (6–9 years)</td>
</tr>
</tbody>
</table>

*Note: Berkeley and Mount Sinai researchers found no consistent decrease in intelligence scores with exposures based on the organophosphate breakdown product DEP. The breakdown product TCPy was not analyzed.*
Inconsistency with Animal Data

Laboratory animal studies conducted over decades as part of the required registration package for chlorpyrifos have failed to identify neurodevelopmental or neurobehavioral effects from exposure (and at exposures far greater than those reported in the Columbia cohort).

A recent review of the animal data concluded that few behavioral parameters were affected following gestational exposures to 1 mg/kg-d [1,000,000 ng/kg/body weight] chlorpyrifos with some inconsistencies across laboratories [12]. Some of these variable effects are attributed to dosing administered subcutaneously or intravenously using a neurotoxic carrier solvent, resulting in exposures that are not relevant to human exposure due to product use. While the authors of the Columbia research try to draw parallels between their findings and animal research, the doses used in those laboratory studies were well in excess of those needed to inhibit blood cholinesterase.

In effect, what this broader analysis reveals is that there is no viable, validated, animal model or series of studies to corroborate the findings that have been reported in the Columbia studies. Across both academic research publications and regulatory guideline studies, there remains no plausible mode of action or identification of consistent effects related to neurodevelopment or neurobehavior to support the findings as reported in the Columbia studies.

Exposures Well Below the No Effect Level

For children in the study group born during 1998 to 2003, Columbia researchers reported links between levels of chlorpyrifos in maternal/umbilical cord blood and fetal growth and reduced scores on various standard psychological, behavior and intelligence tests as well as brain anomalies.

If the effects reported by Columbia University researchers were actually caused by chlorpyrifos, they would have to be caused at a level thousands of times below both current endpoints used for risk assessment and exposures shown to cause no effect in rats (see table below). For a product that has been extensively studied throughout the 40-year history of its use, newly reported effects at doses this low would seem to be highly unlikely.

<table>
<thead>
<tr>
<th>Outcome</th>
<th>Dose (ng/kg/BW)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NOAEL for behavioral effects in animals\textsuperscript{12}</td>
<td>1,000,000</td>
</tr>
<tr>
<td>EPA *BMDL\textsubscript{10} Point of Departure for Human Risk Assessment\textsuperscript{13}</td>
<td>30,000</td>
</tr>
<tr>
<td>EPA’s Chronic Reference Dose\textsuperscript{13}</td>
<td>300</td>
</tr>
<tr>
<td>Estimated daily adult exposure from the Columbia study\textsuperscript{14} (see page 105 in Reference 14)</td>
<td>27</td>
</tr>
</tbody>
</table>

\textsuperscript{*Note: BMDL\textsubscript{10} \approx “No effect” level (lower confidence limit for 10\% RBC ChE inhibition)
As demonstrated by the previous table, the repeat dose endpoint which is protective of adverse effects from exposure to chlorpyrifos based on animal data is 30,000 ng/kg/BW (nanograms per kilogram of body weight) per day. By contrast, the Columbia University researchers reported that the mean level of chlorpyrifos in blood was 4 picograms (pg) per gram, which, assuming for the moment that the exposure was uniform and constant over time, would be the equivalent of 0.027 µg/kg/day, or 27 ng/kg/BW.¹⁴

Is it possible that effects such as those reported in the Columbia research actually occurred in prior animal work but were not detected? For the physical measurements (body weight and body length), this is highly unlikely. In numerous guideline laboratory studies, offspring of several animal species exposed during fetal development had measurements taken of their at-birth body weight, body length and other parameters. No effects on these parameters were noted at doses less than 30,000 ng/kg/BW per day.

In terms of neurological development, obviously no direct comparison can be made between laboratory tests of animal response following exposure to chlorpyrifos versus the performance of children on standardized psychological tests. But it should be noted that, in prior animal research based on EPA accepted protocols, no exposure-related effect was found with chlorpyrifos for measured behavioral parameters (including tests of motor activity, auditory startle, delayed spatial alteration). Additionally, no exposure-related neuropathology was noted, even at doses high enough to cause significant brain cholinesterase inhibition (1,000,000 ng/kg/BW per day).

The enormous discrepancy between the Columbia University findings and those of extensive prior animal research raises serious questions of interpretation. Internal evidence within the Columbia University studies also raises concerns about the validity of their reported findings:

<table>
<thead>
<tr>
<th></th>
<th>Animal Research</th>
<th>Columbia University Research</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Body Weight &amp; Length</strong></td>
<td>No effect found at oral dose of 30,000 ng/kg BW/day¹³</td>
<td>Effect reported at estimated mean dose 27 ng/kg in blood</td>
</tr>
<tr>
<td><strong>Neurological Development</strong></td>
<td>No effect found at oral dose of 1,000,000 ng/kgBW/day¹⁵</td>
<td>Effect reported at estimated mean dose 27 ng/kg in blood</td>
</tr>
</tbody>
</table>

*Note: Estimated fetal doses from Columbia data, based on air monitoring and cord blood, ranged from 0.5 – 63 pg/g.*¹⁴

**Exposure Only Estimated at One Point in Time**

The authors estimated the *in utero* chlorpyrifos exposures based on levels detected in maternal or umbilical cord blood at the time of delivery. They then assumed that these levels were similar for each child prenatally. Since the samples were taken from the children at birth, however, the levels of chlorpyrifos detected do not necessarily reveal the extent of their exposures throughout their prenatal period. Further, the later studies, as the children aged, provided no information about the multitude of other factors that could have affected brain development in the intervening years. *A conclusion of an adverse outcome for a complex, developing organism*
based on the correlation to a single snapshot exposure which represents a broad potential window of fluctuating exposure is overwhelmingly tenuous.

Internal Evidence of Problems with Earlier Columbia University Findings

Given that the findings of the Columbia University researchers are inconsistent with the findings of other, similar human research and that they are in direct conflict with what is known about dose-response from extensive animal research, it is worth evaluating whether the design and execution of these studies may have unintentionally skewed their results. There are many reasons to believe that this may have been the case.

*Exposures Too Low for Accurate Measurement:* Based on the analytical method used, the researchers would have had difficulty accurately categorizing children by their exposures because most of the levels of chlorpyrifos detected in blood were quite low, with 90-97% of the results below 15 pg/g serum. The analytical method used for this analysis was validated in the serum matrix only down to 15 pg chlorpyrifos/g. Levels below this validation limit could have been misclassified into exposure tertiles or groups.

*Missing Data:* Maternal IQ has been shown to have significant bearing on the IQs of offspring, and the authors note that the average IQ score for these women was 86. When IQ scores were missing from several dozen women in this research, however, the researchers arbitrarily substituted the sample mean for the missing data. And when cord blood data were missing for 12 percent of the subjects, the researchers substituted data from maternal blood. It is impossible to determine to what extent these decisions influenced the research conclusions.

*Inconsistent Trends on Test Scores:* According to Columbia University researchers, children born in 1999 when chlorpyrifos was still authorized in the U.S. for in-home use performed more poorly at age three on developmental tests than three-year-old children born in 2000, when chlorpyrifos was being phased out of residential use in the United States. On its face, this might seem like a fairly convincing demonstration that the reductions in test performance were caused by exposure to chlorpyrifos.

But a closer look at these findings raises significant doubt:

In 2001, the levels of chlorpyrifos detected in umbilical cord blood were even lower than in 2000. If chlorpyrifos exposure was linked with lower test scores, then the children born in 2001 ought to have performed better on these tests than those born the previous year. But in fact, the children born in 2001 (i.e., with presumed lesser exposure) performed worse on the tests than those born in 2000 (i.e., when exposures were higher).³

* Differences in Test Scores Fell within Test/Retest Variation:* Reported differences in performance between the “high” vs. “low” exposure groups fell well within the expected test/retest variability for the behavior and development tests.¹² Consequently, the clinical meaning of these findings is unclear.

Despite efforts of the researchers to reduce error and uncertainty in the data, serious questions remain about the validity of the findings of the Columbia study. The U.S. EPA recommends placing “higher confidence in results which are replicated or reproduced from multiple studies”, in their Framework for Incorporating Human Epidemiologic and Incident Data in Health Risk Assessment [16]. To date, none of the results from the Columbia study have been consistently replicated.
Alternate Explanations

Observation of a statistical correlation in an epidemiology study provides no basis for a cause-and-effect assumption in the absence of confirmatory, experimental laboratory data. In fact, there is plausible reason to suppose, given the complex number of factors affecting childhood development, that unmeasured and unreported variables may have confounded the results observed by the Columbia researchers.

No Adjustment for Blood Lipids: Chlorpyrifos binds to blood lipids, and blood lipid levels were not evaluated by the Columbia University research. Increased levels of certain lipids (e.g., elevated LDL cholesterol) have been associated with adverse birth outcomes in prior research, including reduced head circumference, reduced birth weight and reduced ponderal index (similar to body mass index). Women with higher levels of lipids in their blood would be expected to have higher measurable levels of chlorpyrifos as well. Consequently, these findings may be masking some unhealthy lipid levels operating as contributing factor.

Other Exposures: Other reviewers have noted that there appeared to be a higher prevalence of exposure to alcohol and tobacco in the group of women evaluated by the Columbia University research compared to similar research conducted by researchers at Mount Sinai Hospital and the University of California at Berkeley. While researchers did try to control for potential confounding from ethanol intake and second-hand tobacco smoke, other researchers have noted that there were clear limitations in what was possible. Also not controlled as potential confounders in the Columbia University research was the potential effect of maternal depression, duration of breast feeding, household income, and father’s presence in the home, all of which can have a profound impact on the development of a child.

Unique Developmental Challenges Among Children: Test scores from the research showed that about one-third of all the children in this inner city cohort had cognitive and developmental delays, regardless of whether or not the researchers considered them exposed to chlorpyrifos. This suggests that chlorpyrifos may be masking a yet-to-be-discovered underlying effect (e.g., social or economic factors that may potentially influence children’s performance on standard tests).

A Marker for Poor Living Conditions: Due to the phase-out of residential use of chlorpyrifos after 2000, chlorpyrifos levels declined sharply thereafter in Columbia study newborns. Concurrently, pyrethroid insecticide use increased in this cohort over time, as a replacement to the organophosphates. A recent publication from the Columbia researchers identified an association of decreased child development scores at 3 years and piperonyl butoxide levels [a marker of pyrethroid exposure] in study children. The similarity of these findings to those of chlorpyrifos suggest that, in the absence of supporting animal data or confirming human studies, the levels of piperonyl butoxide and chlorpyrifos measured at birth may be markers of pest infestation and poor living conditions rather than indications of a toxic exposure.

The Weight of Accumulated Evidence

Finally, the weight of accumulated evidence over decades of study does not support claims of chlorpyrifos-related risk to child development.

After evaluating thousands of available studies on chlorpyrifos, including those conducted by Columbia University researchers, an international panel of 13 eminent physicians, medical scientists, toxicologists and epidemiologists concluded in an extensive report published in 2008 that “there is no scientific support for a cause-and-effect connection between current levels of chlorpyrifos exposure and adverse human development.”
Similarly, a 2011 weight of evidence evaluation integrating the results of available epidemiology studies (including Columbia University research) and laboratory animal studies concluded that "The weight of the available evidence more strongly indicates that a causal association between chlorpyrifos exposure and neurodevelopmental effects in the absence of AChE inhibition in the brain is not plausible for humans, and the few positive associations observed in epidemiology studies would be attributed to alternative explanations." ¹⁹

REFERENCES