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Background: Children may be at higher risk than adults from pesticide exposure, due to their rapidly developing physiology, unique behavioral patterns, and interactions with the physical environment. This preliminary study conducted in Ecuador examines the association between household and environmental risk factors for pesticide exposure and neurobehavioral development.

Methods: We collected data over 6 months in the rural highland region of Cayambe, Ecuador (2003–2004). Children age 24–61 months residing in 3 communities were assessed with the Ages and Stages Questionnaire and the Visual Motor Integration Test. We gathered information on maternal health and work characteristics, the home and community environment, and child characteristics. Growth measurements and a hemoglobin finger-prick blood test were obtained. Multiple linear regression analyses were conducted.

Results: Current maternal employment in the flower industry was associated with better developmental scores. Longer hours playing outdoors were associated with lower gross and fine motor and problem solving skills. Children who played with irrigation water scored lower on fine motor skills (8% decrease; 95% confidence interval = −9.31 to −0.53), problem-solving skills (7% decrease; −8.40 to −0.39), and Visual Motor Integration test scores (3% decrease; −12.00 to 1.08).

Conclusions: These results suggest that certain environmental risk factors for exposure to pesticides may affect child development, with contact with irrigation water of particular concern. However, the relationships between these risk factors and social characteristics are complex, as corporate agriculture may increase risk through pesticide exposure and environmental contamination, while indirectly promoting healthy development by providing health care, relatively higher salaries, and daycare options.

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Animal studies show that several pesticides including organochlorines, pyrethyroids, organophosphates, and carbamates are developmental neurotoxins.[1–4] However, few epidemiologic studies have focused on the effects of pesticides on neurobehavioral development and function.[5–8] Infants and young children are more susceptible to environmental toxins due to their developing and still immature physiology.[9–13] The developing central and peripheral nervous systems are especially susceptible to adverse affects of neurotoxins such as organophosphates and carbamates.

In addition to their increased susceptibility, young children are also more likely to be exposed to environmental toxins through their behaviors. For infants under the age of 6 months, inhalation and breast milk are the main potential routes of exposure.[14] As infants begin to crawl and spend much of their day on the floor or soil, dermal exposure and oral ingestion become the principal avenues of exposure.[15,16] Sucking, chewing, and biting, are part of normal development and contribute to increased exposure. As children begin to walk, they spend less time on the floor, but their feeding patterns and gender-related behaviors may contribute to exposure risk. Children may eat their meals sitting on the floor or outside on the ground. Boys may be more likely to play outside, whereas girls may be more likely to assist their mothers in washing contaminated clothing or cleaning contaminated food sources.[15]

For young children in agricultural communities, several additional factors may influence exposure risk. The proximity of the child's home to large agricultural industries or farmland may increase exposure, depending on the direction of the wind, water run-off, and ground contamination. Lu and colleagues[17] found that children whose parents worked with pesticides or who lived in close proximity to farmland treated with pesticide had higher exposures to organophosphate pesticides compared with other children in the same community. Similarly, Simcox and colleagues[18] found that children of agricultural families had a higher risk of exposure to organophosphate pesticides than children of nonagricultural families in the same region. Inhalation may be a key exposure route among children who live in close proximity to the flower farms. Dermal and oral exposure may occur through exposure to pesticide residues on parents' work clothes and boots.[19]

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Use of pesticides on private crops, as well as the participation of the child in the cultivation and harvest of these crops, also contribute to the child's exposure profile. Fenske and colleagues[20] found that use of pesticides in a home garden was associated with an increase of an organophosphate metabolite in children's urine samples.

In Ecuador, large-scale agricultural products such as bananas and cut-flowers are important contributors to the national economy. The cut-flower industry is now the country's third most important export, following petroleum and bananas. Pesticide use in this industry is common, with the organophosphate, carbamate, and dithiocarbamate classes of pesticides being the most frequently used.

In 2001, the Centro de Estudios y Asesoría en Salud (CEAS) in Quito, Ecuador, in collaboration with the Canadian International Development Research Center (IDRC), initiated the EcoSalud Project in the Cayambe-Tabacundo region of Ecuador. This project addresses the social impact of the cut-flower industry through a wide range of approaches, including sociology, anthropology, ecology, and epidemiology. This preliminary study considered household and community risk factors for pesticide exposure and their potential effects on neurobehavioral development of young children aged 24 to 61 months living in the region.

METHODS

Study Population

The sample population for the EcoSalud project was drawn from 2 northern and 2 southern sections of the Cayambe-Tabacundo region. The present study focused on 3 northern communities. Communities A and B were at lower altitudes and were likely to have higher exposures, with more community members employed in the flower industry and living in closer proximity to the flower plantations. Community C was at a higher altitude and was likely to have lower exposure. It was located farther from the flower plantations and had few flower industry employees. These communities were selected based on exposure status and because of close ties between researchers and community leaders, allowing for greater accessibility to the communities' members.

A census was taken to establish the sampling frame. All mothers who had been living in the community for at least a year and who had any children aged 3 to 61 months were eligible to participate. Mothers were interviewed about all their eligible children, up to 3 children total. Informed consent was obtained from the mothers for their participation as well as that of their children. Consent forms were read to the mother and consent was documented by the mother's signature or fingerprint. In total, 219 mothers (91% of those eligible) and 283 children (91% of those eligible) participated. This analysis of household and community risk factors for exposure to pesticides includes children 24 to 61 months of age ($n = 142$). Approval for this project was obtained from the Institutional Review Board at the University of Michigan, as well as from CEAS in Quito, Ecuador.

Procedures

The Ages and Stages Questionnaire (ASQ) (2nd Edition; Paul H. Brookes Publishing Co., Baltimore, MD) is a developmental screening test that was directly administered to the child. We administered an additional test to children aged 48 to 61 months, to assess their visual-motor integration (VMI) skills. Two trained testers assessed the participating children in each of the 3 communities. Mothers were administered a questionnaire to obtain information on sociodemographic characteristics, maternal occupational history, maternal and child health characteristics, and the child's socialization and exposure profiles. Blood by finger-prick was obtained to assess the child's hemoglobin levels (HemoCue, Lake Forest, CA). Height (centimeters), weight (kilograms), and head circumference (centimeters) of the child were measured. All survey instruments were pretested and piloted to ensure clarity and comprehensibility.

Risk Factors for Pesticide Exposure

Potential risk factors for pesticide exposure were classified within 3 domains: 1) community and household variables; 2) maternal and paternal work variables; and 3) child activities. As community and household risk factors, we used distance of the home to a flower farm ($>$200 m, $\leq$200 m), pesticide use on domestic crops (yes/no), pesticide use within the home (yes/no), and use of the potentially contaminated plastics/wood from the flower farms at home (yes/no). Maternal work exposure included whether she had worked in the flower industry in the past 6 years (yes/no, and mean number of years), whether she had worked in the flower industry while pregnant with the child (yes/no), and her current work status (currently not working, currently working but not in flowers, currently working in flowers). Mothers also reported father's current work status. Child's play activities included the distance from a flower farm ($>$200 m, $\leq$200 m), number of hours per day the child plays outdoors (mean number of hours/day; $<5/\geq5$ hours/day), and whether the child plays with irrigation water (no or rarely, sometimes or frequently).

Neurobehavioral Development

Ages and Stages Questionnaire

The use of a screening test that combines both parent-report and direct evaluation approaches has been shown to be an effective and valid way to assess a child's developmental progress.[21,22] The questionnaire is standardized for use in children ages 3 to 61 months and is composed of 19 age-specific questionnaires that cover 5 broad developmental dimensions: communication, fine and gross motor skills, problem solving, and personal-social skills. Each domain is scored from 0 to 60 points, with 60 being a perfect score. A continuous score is calculated for each age-specific questionnaire, with scores summarized for each developmental domain.

Before administering the ASQ (Spanish version), the tool was adapted into the local vernacular and contextually inappropriate questions were removed to prevent cultural and language bias. For example, all references to the baby/child were changed to the term "guagua," a Quichua term commonly used in the region. Testing was conducted using the home-visit procedure outlined in the manual, in which the tester attempts to elicit all behaviors directly from the child during the assessment.[22] This procedure varies slightly from one that relies solely on parent-report and is more appropriate in a setting where the parent may not be able to complete the questionnaire on her own. Testers brought all materials required for direct assessment as listed in the manual. Mothers were encouraged to participate in the activities with their child throughout the session. When the particular activity could not be carried out directly, the mother was asked about the child's behavior at home.

Visual Motor Integration

The use of targeted developmental testing may be more able to detect subtle delays in development in this population due to chronic low level exposure to pesticides. To supplement the use of a general screening tool, we included one targeted developmental test that assessed the integration of visual perception and motor skills—skills that have been shown to be affected by pesticide exposure in adults and children.[5,7,23,24] Children aged 48 to 61 months were tested using the Beery-Buktenica VMI developmental test (4th Edition, Revised ed., Modern Curriculum Press).[25] The VMI is a developmental sequence of geometric forms to be copied with paper and pencil by the child. It is designed to assess the extent to which the child can integrate his visual and motor abilities. The VMI manual was used as a reference for scoring, with each drawing scored as 0 for incorrect and 1 for correct. Points were totaled and the raw score was recorded. A total score of 160 was possible. Based on the age of the child, the raw score was converted into a standard score equivalent. The VMI manual presents a standardized mean score of 100 and standard deviation (SD) of 15.[25]

Covariates

Standardized z-scores for anthropometric measures of chronic malnutrition were calculated using the 1978 Centers for Disease Control (CDC)/World Health Organization (WHO) growth reference curves, which are a normalized version of the 1977 National Center for Health Statistics (NCHS) growth reference curves.[26] Chronic malnutrition (stunting) was defined by a height-for-age z-score 2 SDs below the reference median. Presence of anemia (yes/no) was determined after taking into account the child's age and the altitude of the community of residence.[27]

The child's exposure to developmentally fostering experiences was assessed by attendance at the daycare center (yes/no) and the type and frequency of stimulating activities at home. For the latter, a set of 6 questions was adapted from a UNICEF multicountry survey.[28] The 6 activities between mother and child included reading, counting and drawing, looking at pictures (from any type of media), singing songs, going out of the house together, and playing.

Maternal and sociodemographic characteristics included maternal age and education level, father's education level, mother's ethnicity and predominant language preference (Quichua /Spanish, Spanish only), marital status, monthly household income in US dollars ($0–150, $151–250, or >$250), and housing construction. Maternal age was examined as a continuous variable and as a dichotomous variable based on the median (≤25 years old, >25 years old). Maternal education, categorized as none/partial primary, completed primary school, or partial/completed high school, was used to assess education level and as a proxy for literacy. Mother's education and her ability to read were correlated ($r = 0.52$) as were mother's education and her ability to write ($r = 0.54$). Father's education level was categorized similarly. A housing scale (ranging from 0 to 7), was constructed from the following housing characteristics: roof composition; floor composition; wall composition; type of water used in home; bathroom type; and access to electricity. This housing scale was categorized as poorer (≤3), midlevel (4–5), and better (6–7).

Statistical Analysis

We examined distributions of the pesticide-related community and household factors, child play activity, child's health and nutrition, maternal characteristics, and the sociodemographic characteristics of the child's family. Cross-tabulation of all risk factors were examined to assess collinearity, and χ^2 tests were conducted to assess differences across risk factor categories.

Developmental delay was analyzed separately for each developmental domain screened by the ASQ, and for the VMI test score. We assessed differences in mean developmental scores among the various pesticide-related risk factors using ANOVA and *t* tests. We constructed regression models for each developmental domain to assess the effects of household environmental exposures to pesticides on development after controlling for key confounders. Prior analyses have examined the sociodemographic and health characteristics associated with neurobehavioral development in this population.[29,30] In this paper, we consider those variables associated with development (as measured by the ASQ) in our population and associated with our exposure variables as potential confounders. Due to the limited sample size, only those variables found to be associated with each ASQ domain and the exposure variable of interest were included in the regression model for a given domain.

We report potential associations, with confidence intervals and the percent change in developmental scores between exposure groups. Effect size was calculated to compare the magnitude of effect of the main exposure variables on the developmental scores across exposure groups.[31] The measure of effect size, Cohen's *d*, is calculated by taking the difference in the mean score of each exposure group divided by the SD; this parameter is independent of sample size. Effect size is cautiously interpreted as small for $d = 0.2$, medium for $d = 0.5$, and large for $d = 0.8$. Data were entered into SPSS 11.5 (SPSS, Chicago, IL) and were analyzed in SPSS and SAS Version 8 (SAS Institute, Cary, NC). Nutritional data were analyzed in EpidInfo's NutStat program software (CDC, Division of Public Health Surveillance and Informatics, 2003).

RESULTS

A total of 142 children aged 24 to 61 months were included in this analysis, with 57 children aged 48 to 61 months included in the VMI testing group. Table 1 displays the maternal and health and sociodemographic characteristics of the study population. Three-quarters of the mothers in this population identified as indigenous, and 89% reported their predominant language as Spanish. Approximately 80% of the mothers reported being married or living with their partner in a free union. About half (56%) of the mothers had completed primary schooling and 21% of the fathers achieved a partial or complete high school education. Almost half (45%) of the mothers reported a monthly household income of $150 or less and about half reported a midlevel housing quality.

There was a high prevalence of both anemia (51%) and stunting (60%) among the children. About a quarter (22%) of the children had suffered from at least one infection in the past 3 months. Nearly two-thirds (63%) participated in 3 or more developmentally stimulating activities with their mother at home. A lower percentage (24%) of children were engaged in such activities outside the home, specifically at a daycare center.

Table 2 displays the risk factors for pesticide exposure included in the analyses. Approximately half of the mothers reported working in the flower industry in the previous 6 years (53%), working an average of 45 months. Thirty-five percent of the mothers reported currently being employed in the flower industry, and 39% reported having worked in the flower industry during their pregnancy. More than half of the children resided in a household where pesticides were used on domestic crops (78%) and inside the home (57%). Over half of the children spent 5 or more hours playing outdoors (mean = 5.6 hours), and about half of the children occasionally or frequently played with irrigation water (49%), with no substantial difference between boys and girls.

Table 3 presents the unadjusted mean developmental outcome score (with SD) and the percent change for each neurobehavioral domain by risk factors for pesticide exposure. Given the correlations among several of the exposure variables, we focused on those variables that describe distinct exposure domains (eg, maternal employment, pesticide use within and around the home, and child play activities).

Tables 4 and 5 display the results of adjusted regression models for the ASQ developmental outcomes and the VMI scores, respectively. Results of the unadjusted and adjusted analyses were similar. Mother's current employment in the flower industry was associated with better scores among their children for all 5 ASQ domains. Specifically, current employment in flowers was more strongly associated with better communication and problem-solving skills. Employment in the flower industry during pregnancy, however, showed a positive association only with the problem-solving skills domain. Pesticide use on domestic crops was also associated with better gross motor and personal-social scores, while conversely, pesticide use within the home was associated with lower communication scores (effect size: $d = 0.2$).

More hours spent outdoors (>5 hours/d) was associated with lower gross and fine motor and problem solving skills.

TABLE 1. Maternal and Child Characteristics of the Study Population, Children 24–61 Months of Age (n = 142), Cayambe-Tabacundo Region, Ecuador (2003)

	No. (%)
Mother's characteristics	
Mother's age; yrs	
≤25	64 (45)
25+	78 (55)
Ethnicity of mother*	
Indigenous	108 (77)
Mestizo/white	32 (23)
Language most used	
Spanish/Quichua mix	15 (11)
Spanish	127 (89)
Marital status	
Married	88 (62)
Free union	29 (20)
Single/separated/widowed	25 (18)
Mother's education level	
None or partial elementary	40 (28)
Completed elementary school	80 (56)
Partial or completed high school	22 (16)
Socioeconomic characteristics	
Monthly household income (U.S. dollars)†	
$0–150	63 (45)
$151–250	41 (29)
>$250	37 (26)
Father's education level	
None or partial elementary	23 (16)
Completed elementary school	64 (45)
Partial or completed high school	30 (21)
No father	25 (18)
Housing construction	
Poorer	25 (18)
Midlevel	73 (51)
Better	44 (31)
Child health characteristics	
Sex of child	
Male	71 (50)
Female	71 (50)
Anemia‡	
No	69 (49)
Yes	73 (51)
Stunting	
No	57 (40)
Yes	85 (60)
Infection health score†§	
0	110 (78)
≥1	31 (22)
Maternal-child stimulation at home	
3+	89 (63)
≤3 activities	52 (37)
Daycare attendance	
No	108 (76)
Yes	34 (24)

*Information missing for 2 mothers.
†Information missing for 1 mother/child.
‡Cut points for anemia were specific for age and community altitude, and ranged from 12.3 g/dL to 13.2 g/dL.
§In last 3 months, cold, sore throat, lung infection, cough, ear infection, or other infection.

TABLE 2. Potential Risk Factors for Pesticide Exposure in the Study Population, Children 24–61 Months of Age (n = 142)

Exposure variables	
Mother worked in the flower industry in past 6 yrs; no. (%)	
No	67 (47)
Yes	75 (53)
Total no. mos worked in flowers in past 6 yr (n = 75); mean	44.7
Mother worked in flower industry during pregnancy; no. (%)	
No	87 (61)
Yes	55 (39)
Mother's current work status; no. (%)	
Currently is not working or has not worked in past 6 yrs	83 (59)
Currently works, but not in flower industry	10 (7)
Currently works in flower industry	49 (35)
Father's current work status; no. (%)	
Currently is not working or has not worked in past 6 yrs	26 (19)
Currently works, but not in flower industry	44 (31)
Currently works in flower industry	46 (33)
No father	25 (18)
Use of pesticides on domestic crops; no. (%)	
No	32 (23)
Yes	110 (78)
Use of pesticides within the home; no. (%)	
No, rarely	61 (43)
Sometimes/frequently	81 (57)
Use of plastics/wood at home; no. (%)	
Neither	45 (32)
Either plastics or wood	97 (68)
Child play activities	
Distance between place where child passes day to flower farm (meters); no. (%)	
>200	97 (68)
≤200	45 (32)
Number of hours child plays outdoors daily; no. (%)	
<5	50 (35)
5+	92 (65)
Mean	5.61
Child plays with irrigation water; no. (%)	
No/rarely	73 (51)
Occasionally/frequently	69 (49)

Occasional or frequent playing in irrigation water was associated with poorer fine motor and problem solving scores. On average, a child who played with irrigation water scored approximately 5 points lower on fine motor skills (8.2% decrease; 95% CI= −9.3 to −0.5); effect size, $d = 0.4$), 4.4 points lower on problem solving skills (7.3% decrease; −8.4 to −0.4;; effect size, $d = 0.3$), and 5.5 points less on the VMI test (3.4% decrease; −12.0 to 1.1; effect size, $d = 0.2$) compared with children who rarely or never played with irrigation water. Current employment in the flower industry, longer hours spent outdoors playing, and contact with irrigation water were all associated with lower VMI scores; however, estimates are imprecise, as the VMI was administered only to a subset of the study, thus reducing the sample size.

DISCUSSION

These results suggest that exposures related to the cut-flower industry may be associated with child development and health, although these associations are complex. Some factors associated with the presence of the industry may be harmful, such as exposure to pesticides, whereas other factors such as increased resources and opportunities for creating a stimulating environment may be beneficial.

The environmental impacts of agricultural industry are well-established, particularly on the contamination of irrigation and drainage canals from pesticide residues.[32–34] In communities where cut-flower farms are located, irrigation canals are located close to the flower plantations and pass throughout the community, where the population then uses this water for domestic purposes. It is not uncommon for these canals to receive waste waters from the farms. Environmental sampling done in this region showed increasing contamination in the water systems and river basins sediment according to proximity to the contaminating sources. Systems closest to the flower farms showed the highest level of contamination with pesticide, metal, and biologic residues.[35]

Few studies have examined child exposure during play to potentially-contaminated surface water such as irrigation water. Most studies focus on the effects of exposure via ingesting contaminated drinking water. The results of the present study provide evidence that children's exposure to this water was associated with lower developmental screening test scores. Future research should incorporate continued environmental sampling of irrigation waters and sediment in the region to determine the type and the level of contamination in these surfaces.

Pesticide use on domestic crops and their proximity to children's outdoor play area were either not associated with developmental delay or were associated with better developmental outcomes. We considered several explanations for these seemingly anomalous findings. The risk of children living and playing in close proximity to the flower farms might be offset by the benefits of having a mother employed in the flower industry, which corresponds to a higher monthly household income, higher parental educations, and better access to health care and daycare. As we were not able to take direct measures of pesticide exposure in the physical environment of the child, we had to rely on indirect or proxy measures, and they may not correlate well with actual exposure. Alternatively, there may be unmeasured confounders. For example, domestic pesticide use may correlate with economic security; pesticides are an added expense for the family and only those who can afford the chemicals may be using them at home.

Current maternal employment in the flower industry and maternal employment in the flower industry during pregnancy were associated with better communication and problem-solving skills. Previous studies have found that take-home exposures via contaminated parental work clothing and equipment contribute to household pesticide residue contamination.[17,18,36] In this population, however, it is uncommon for flower farm workers to take home their work clothes and

TABLE 3. Developmental Outcomes Stratified by Household and Community Risk Factors for Pesticide Exposure for Children age 24–61 Months (n = 142)*

		Communication		Gross Motor		Fine Motor		Problem Solving		Personal-Social		VMI[†]	
	Total No.	Mean ± SD	% Change	Mean ± SD	% Change	Mean ± SD	% Change	Mean ± SD	% Change	Mean ± SD	% Change	Total No.	Mean ± SD
Mother's current work status													
Currently is not working, or has not worked in past 6 yrs, or currently works but not in flower industry	93	41.2 ± 13.5	5%	44.7 ± 11.2	3%	40.8 ± 13.9	6%	30.6 ± 14.1	9%	40.3 ± 12.9	4%	37	92.0 ± 14.2
Currently works in flower industry	49	44.4 ± 12.7	—	46.7 ± 13.1	—	44.5 ± 13.2	—	36.2 ± 12.8	—	42.9 ± 12.0	—	20	97.4 ± 15.8
Mother worked in flower industry during pregnancy													—
No	87	43.3 ± 11.6	4%	46.1 ± 10.7	3%	41.4 ± 13.3	3%	30.7 ± 14.3	8%	40.9 ± 12.6	1%	37	91.4 ± 13.8
Yes	55	40.7 ± 15.5	—	44.3 ± 13.6	—	43.1 ± 14.5	—	35.5 ± 12.6	—	41.7 ± 12.7	—	20	98.6 ± 15.9
Use of pesticides on domestic crops													
No	32	42.2 ± 17.6	0%	40.3 ± 15.6	11%	42.2 ± 14.2	0%	32.8 ± 15.0	1%	37.1 ± 14.0	9%	15	96.0 ± 17.6
Yes	110	42.3 ± 11.8	—	46.9 ± 10.2	—	42.0 ± 13.7	—	32.5 ± 13.6	—	42.4 ± 12.0	—	42	93.2 ± 13.9
Use of pesticides within the home													
No/rarely	61	43.9 ± 1.7	5%	43.9 ± 12.8	5%	41.4 ± 15.1	2%	32.2 ± 14.3	1%	40.6 ± 13.4	2%	30	94.3 ± 15.4
Sometimes/frequently	81	41.1 ± 14.3	—	46.6 ± 11.1	—	42.6 ± 12.7	—	32.8 ± 13.6	—	41.6 ± 12.0	—	27	93.4 ± 14.5
Number of hours child plays outdoors daily													
<5	50	42.1 ± 13.6	2%	47.9 ± 10.6	2%	44.3 ± 12.0	8%	34.9 ± 12.7	8%	42.1 ± 12.6	3%	24	98.8 ± 13.2
5+	92	42.4 ± 13.2	—	44.1 ± 12.4	—	40.9 ± 14.5	—	30.1 ± 14.7	—	40.2 ± 12.7	—	33	90.3 ± 15.1
Child plays with irrigation water													
No/rarely plays with irrigation water	73	42.8 (14.0)	5%	45.0 (13.3)	3%	44.5 ± 12.9	6%	30.6 ± 14.1	9%	40.3 ± 12.9	4%	27	98.6 ± 15.9
Occasionally/frequently plays with irrigation water	69	41.7 (12.5)	—	45.9 (10.2)	—	39.5 ± 14.2	—	36.2 ± 12.8	—	42.9 ± 12.0	—	30	89.7 ± 12.6

*Differences in mean scores were assessed using ANOVA and *t* test; higher score corresponds to better developmental skills.
[†]Mean scores presented are the standard score equivalent of the child's raw score.[25] VMI tests only administered to children age 48–61 months (n = 57). VMI published standardized scores have a mean of 100 and standard deviation.

TABLE 4. Adjusted Regression Models for 5 ASQ Developmental Domains for Distinct Risk Factors for Pesticide Exposure in the Household and Community Environment for Children Age 24–61 Months (n = 142)

Household/Community Exposure Variable	ASQ Domain*	β (SE)†	% Change (95% CI)
Mother currently works in flower industry	Communication	4.13 (2.23)	6.9 (−0.3 to 8.5)
	Gross motor	2.38 (2.04)	4.0 (−1.7 to 6.4)
	Fine motor	2.12 (2.45)	3.5 (−2.7 to 7.0)
	Problem solving	5.03 (2.20)	8.4 (0.7 to 9.4)
	Personal–social	1.87 (2.31)	3.1 (−2.7 to 6.5)
Mother worked in flower industry during pregnancy	Communication	−1.71 (2.19)	−2.9 (−6.1 to 2.6)
	Gross motor	−0.86 (1.99)	−1.4 (−4.8 to 3.1)
	Fine motor	0.59 (2.35)	1.0 (−4.1 to 5.2)
	Problem solving	3.57 (2.11)	6.0 (−0.6 to 7.8)
	Personal-social	0.15 (2.21)	0.3 (−4.2 to 4.5)
Pesticides on domestic crops	Communication	−1.03 (2.56)	−1.7 (−6.1 to 4.0)
	Gross motor	4.86 (2.18)	8.1 (0.6 to 9.2)
	Fine motor	0.57 (2.71)	1.0 (−4.8 to 5.9)
	Problem solving	0.04 (2.47)	0.1 (−4.8 to 4.9)
	Personal–social	4.12 (2.46)	7.0 (−0.7 to 9.0)
Pesticides in home	Communication	−4.51 (2.15)	−7.5 (−8.8 to −0.3)
	Gross motor	2.10 (1.87)	3.5 (−1.6 to 5.8)
	Fine motor	1.50 (2.28)	2.5 (−3.0 to 6.1)
	Problem solving	−0.52 (2.09)	−1.0 (−4.7 to 3.6)
	Personal–social	0.02 (2.12)	0.0 (−4.2 to 4.2)
Child plays outdoors ≥5 h/d	Communication	−0.06 (2.24)	−0.1 (−4.5 to 4.4)
	Gross motor	−2.52 (1.94)	−4.2 (−6.3 to 1.3)
	Fine motor	−2.12 (2.41)	−3.5 (−6.9 to 2.6)
	Problem solving	−3.28 (2.16)	−5.5 (−7.5 to 1.0)
	Personal–social	1.89 (2.18)	3.2 (−2.4 to 6.2)
Child plays with irrigation water	Communication	−2.23 (2.15)	−3.7 (−6.5 to 2.0)
	Gross motor	0.72 (1.85)	1.2 (−2.9 to 4.4)
	Fine motor	−4.92 (2.22)	−8.2 (−9.3 to −0.5)
	Problem solving	−4.39 (2.03)	−7.3 (−8.4 to −0.4)
	Personal–social	−2.09 (2.08)	−3.5 (−6.2 to 2.0)

*Communication domain is adjusted for presence of anemia, presence of infection in past 3 months. Gross motor domain is adjusted for age of child, presence of infection in past 3 months, stimulation at home, and housing construction. Fine motor domain is adjusted for daycare attendance. Problem solving domain is adjusted for age of child, mother's education. Personal-social domain is adjusted for presence of infection in past 3 months, stimulation at home.

†Mean difference in score.

TABLE 5. Adjusted* Regression Models for VMI Developmental Outcome for Distinct Risk Factors for Pesticide Exposure in the Household and Community Environment for Children Age 24–61 Months (n = 57)

Household/Community Exposure Variable	Developmental Outcome	β (SE)†	% Change (95% CI)
Mother currently works in flower industry	VMI	−3.53 (3.94)	−2.2 (−11.4 to 4.4)
Mother worked in flower industry during pregnancy	VMI	0.19 (3.74)	−0.1 (−7.3 to 7.7)
Pesticides on domestic crops	VMI	−0.39 (3.82)	−0.2 (−8.0 to 7.3)
Pesticides in home	VMI	−2.41 (3.29)	−1.5 (−9.0 to 4.2)
Child plays outdoors ≥5 h/d	VMI	−3.90 (3.43)	−2.4 (−10.8 to 3.0)
Child plays with irrigation water	VMI	−5.46 (3.26)	−3.4 (−12.0 to 1.1)

*Adjusted for daycare attendance and household monthly income.

†Mean difference in score.

equipment. Therefore, assessing take-home pathways for exposure via parental work clothing may be less relevant for this population, whereas other aspects of maternal employment in the flower industry (ie, increased monthly income, more education, better access to daycare and health services) may benefit the child's health and development.

This preliminary study has several limitations. We relied on indirect exposure measurement through the use of a questionnaire, which may lead to exposure misclassification. Additionally, domestic pesticide use is common in this region. We were not able to obtain information on the type or quantity of pesticides used, either in the work environment or at home, therefore leading to another potential source of measurement error. Future investigations should incorporate the use of biomarkers and environmental sampling to supplement information gathered by questionnaire.

There are several limitations of a general screening tool such as the ASQ[22] in this population. The optimal test for assessing developmental delay is one that directly evaluates the child and assesses specific developmental functions and domains. These domain-specific tests are particularly useful when there are sufficient data to generate hypotheses about which domains may be affected. However, global tests of development can be expensive, difficult to administer in a field setting, and time-consuming. In a developing country such as Ecuador, where assessment of neurobehavioral development must be conducted in a field setting with minimal cost, a screening test was the most appropriate option in a preliminary investigation. Another limitation of using the ASQ is that, although standardized and validated in a large and multicultural population in the United States, the instrument may not be culturally appropriate for rural Andean populations. Validation of these developmental tools in this and similar cultures is needed.

Our results highlight the importance of social relationships and opportunities that may interact with adverse exposures to affect neurobehavioral development. Neurobehavioral development is a complex and dynamic process that is affected by numerous factors, including parent-child interaction, household relationships, maternal health, physical environment, the child's physical and mental health, and social organization. Associated social opportunities that increase access to daycare or health care for the child may counteract the effects of pesticide exposure. In researching effects of pesticide exposure on child neurobehavioral development, recognition of other environmental factors within a larger ecosystem is critical.

Our analysis highlights the complexity of considering exposure risks that come with new economic endeavors. One would expect to see gains in health of a population that has experienced the economic growth that the cut-flower industry has brought to as has this region of Ecuador. The economic benefits of the flower industry may improve child health in some respects, whereas pesticide exposure and environmental contamination may cause harm in others. Both aspects must be considered when assessing the impact of specific industrial exposures on child development and health.

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REFERENCES

1. Eriksson P, Ahlbom J, Fredriksson A. Exposure to DDT during a defined period in neonatal life induces permanent changes in brain muscarinic receptors and behaviour in adult mice. *Brain Res*. 1992;582:277–281.
2. Eriksson P, Fredriksson A. Neurotoxic effects of two different pyrethroids, bioallethrin and deltamethrin, on immature and adult mice: changes in behavioral and muscarinic receptor variables. *Toxicol Appl Pharmacol*. 1991;108:78–85.
3. Chanda SM, Pope CN. Neurochemical and neurobehavioral effects of repeated gestational exposure to chlorpyrifos in maternal and developing rats. *Pharmacol Biochem Behav*. 1996;53:771–776.
4. Miller DB. Neurotoxicity of the pesticidal carbamates. *Neurobehav Toxicol Teratol*. 1982;4:779–787.
5. Guillette EA, Meza MM, Aquilar MG, et al. An anthropological approach to the evaluation of preschool children exposed to pesticides in Mexico. *Environ Health Perspect*. 1998;106:347–353.
6. Young JG, Eskenazi B, Gladstone EA, et al. Association between in utero organophosphate pesticide exposure and abnormal reflexes in neonates. *Neurotoxicology*. 2005;26:199–209.
7. Ruckart PZ, Kakolewski K, Bove FJ, et al. Long-term neurobehavioral health effects of methyl parathion exposure in children in Mississippi and Ohio[see comment]. *Environ Health Perspect*. 2004;112:46–51.
8. Rohlman DS, Arcury TA, Quandt SA, et al. Neurobehavioral performance in preschool children from agricultural and non-agricultural communities in Oregon and North Carolina. *Neurotoxicology*. 2005;26:589.
9. Faustman EM, Silbernagel SM, Fenske RA, et al. Mechanisms underlying children's susceptibility to environmental toxicants. *Environ Health Perspect*. 2000;108(Suppl 1):13–21.
10. Whyatt R. Intolerable risk: the physiological susceptibility of children to pesticides. *J Pest Reform*. 1989;9:5–9.
11. Scheuplein R, Charnley G, Dourson M. Differential sensitivity of children and adults to chemical toxicity. I. Biological basis. *Regul Toxicol Pharmacol*. 2002;35:429–447.
12. Wargo J. *Our Children's Toxic Legacy*. New Haven, CT: Yale Press; 1996.
13. National Research Council. *Pesticides in the Diets of Infants and Children*. Washington, DC: National Academy Press; 1993.
14. Eskenazi B, Bradman A, Castorina R. Exposures of children to organophosphate pesticides and their potential adverse health effects. *Environ Health Perspect*. 1999;107(Suppl 3):409–419.
15. Hubal EAC, Sheldon LS, Burke JM, et al. Children's exposure assessment: a review of factors influencing children's exposure, and the data available to characterize and assess that exposure. *Environ Health Perspect*. 2000;108:475–486.
16. Aprea C, Strambi M, Novelli MT, et al. Biologic monitoring of exposure to organophosphorus pesticides in 195 Italian children. *Environ Health Perspect*. 2000;108:521–525.
17. Lu C, Fenske RA, Simcox NJ, et al. Pesticide exposure of children in an agricultural community: evidence of household proximity to farmland and take home exposure pathways. *Environ Res*. 2000;84:290–302.
18. Simcox NJ, Fenske RA, Wolz SA, et al. Pesticides in household dust and soil: exposure pathways for children of agricultural families. *Environ Health Perspect*. 1995;103:1126–1134.
19. Loewenherz C, Fenske RA, Simcox NJ, et al. Biological monitoring of organophosphorus pesticide exposure among children of agricultural workers in central Washington state. *Environ Health Perspect*. 1997; 105:1344–1353.
20. Fenske RA, Lu C, Barr D, et al. Children's exposure to chlorpyrifos and parathion in an agricultural community in central Washington state[see comment]. *Environ Health Perspect*. 2002;110:549–553.
21. Glascoe FP. Evidence-based approach to developmental and behavioural surveillance using parents' concerns. *Child Care Health Dev*. 2000;26: 137–149.

22. Squire J, Potter L, Bricker D. *The ASQ User's Guide*. 2nd ed. Baltimore, MD: Brooks Publishing Co.; 1999.
23. Cole DC, Carpio F, Julian J, et al. Neurobehavioral outcomes among farm and nonfarm rural Ecuadorians. *Neurotoxicol Teratol*. 1997;19: 277–286.
24. Farahat TM, Abdelrasoul GM, Amr MM, et al. Neurobehavioural effects among workers occupationally exposed to organophosphorous pesticides. *Occup Environ Med*. 2003;60:279–286.
25. Beery KE. *The Beery-Buktenica Developmental Test of Visual-Motor Integration: VMI.* 4th ed. Revised ed. Cleveland: Modern Curriculum Press.
26. Dibley MJ, Goldsby JB, Staehling NW, et al. Development of normalized curves for the international growth reference: historical and technical considerations. *Am J Clin Nutr*. 1987;46:736–748.
27. CDC. CDC criteria for anemia in children and childbearing-aged women. MMWR *Morb Mortal Wkly Rep*. 1989;38:400–404.
28. UNICEF. UNICEF Indicators Projects: Family Psychosocial Care Practices Measures. 2003; Version 16.
29. Handal AJ, Lozoff B, Breilh J, et al. Socio-demographic and nutrition correlates of neurobehavioral development in Ecuadorian children. *Pan Am J Public Health.* In press.
30. Handal AJ, Lozoff B, Breilh J, et al. Effect of community of residence on neurobehavioral development in infants and young children in a flower growing region of Ecuador. *Environ Health Perspect*. 2007;115: 128–133.
31. Cohen, J. *Statistical Power Analysis For The Behavioral Sciences.* 2nd ed. Hillsdale, NJ: Lawrence Earlbaum Associates; 1988.
32. Lopez-Rios O, Lechuga-Anaya M. [Pollutants in water bodies in the south of Sonora.] *Salud Publica Mex*. 2001;43:298–305.
33. Ritter L, Solomon K, Sibley P, et al. Sources, pathways, and relative risks of contaminants in surface water and groundwater: a perspective prepared for the Walkerton inquiry. *J Toxicol Environ Health A*. 2002; 65:1–142.
34. Hunt JW, Anderson BS, Phillips BM, et al. Ambient toxicity due to chlorpyrifos and diazinon in a central California coastal watershed. *Environ Monit Assess*. 2003;82:83–112.
35. Breilh J, Campana A, Hidalgo F, et al. Floriculture and the health divide: a struggle for fair and ecological flowers. In: CEAS, ed. *Latin American Health Watch: Alternative Latin American Health Report*. Quito: Global Health Watch; 2005;66–79.
36. Thompson B, Coronado GD, Grossman JE, et al. Pesticide take-home pathway among children of agricultural workers: study design, methods, and baseline findings. *J Occup Environ Med*. 2003;45:42–53.