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# Determinants of manganese levels in house dust samples from the CHAMACOS cohort

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## Abstract

**Introduction—**Manganese (Mn) is an essential nutrient, but at high exposure levels Mn is a neurotoxicant. The fungicides maneb and mancozeb are approximately 21% Mn by weight and more than 150,000 kg are applied each year to crops in the Salinas Valley, California. It is not clear, however, whether agricultural use of these fungicides increases Mn levels in homes.

**Materials and methods—**We collected house dust samples from 378 residences enrolled in the Center for the Health Assessment of Mothers and Children of Salinas (CHAMACOS) study with a second sample collected approximately nine months later from 90 of the residences. House dust samples were analyzed for Mn using inductively coupled plasma optical emission spectroscopy. Information from interviews, home inspections, and pesticide use reports was used to identify potential predictors of Mn dust concentrations and loadings.

**Results—**Mn was detectable in all dust samples. The median Mn concentration was 171 μg/g and median Mn loading was 1,910 $\mu g/m^2$ at first visit. In multivariable models, Mn dust concentrations and loadings increased with the number of farmworkers in the home and the amount of agricultural Mn fungicides applied within three kilometers of the residence during the month prior to dust sample collection. Dust concentrations of Mn and other metals (lead, cadmium and chromium) were higher in residences located in the southern Salinas Valley compared those located in other areas of the Salinas Valley. Dust loadings of Mn and other metals were also higher in residences located on Antioch Loam soil than other soil types, and in homes with poor or average housekeeping practices.



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**Competing financial interests:** One of the authors (AB) has served as a consultant on cases unrelated to the issues covered in this paper and has participated as a member of the Science Advisory Board for The Organic Center, a non-profit organization that provides information for scientific research about organic food and farming. The other authors declare they have no actual or potential competing financial interests.

**Conclusions—**Agricultural use of Mn containing fungicides was associated with Mn dust concentrations and loadings in nearby residences and farmworker homes. Housekeeping practices and soil type at residence were also important factors related to dust metal concentrations and loadings.

**Keywords**

house dust; exposure science; GIS; manganese; metals; pesticides

## 1. Introduction

Manganese (Mn) is an essential nutrient but is neurotoxic at high exposure levels (ATSDR 2008). In children, higher Mn levels in blood and hair have been associated with poorer mental development (Claus Henn et al. 2010), lower full-scale and verbal intelligence quotient (Menezes-Filho et al. 2011), lower verbal and performance intelligence quotient (Riojas-Rodriguez et al. 2010), poorer attention and non-verbal memory (Takser et al. 2003), increased externalizing behavior problems (Ericson et al. 2007), and more oppositional and hyperactive behaviors (Bouchard et al. 2011). An interaction between Mn and lead resulting in poorer overall, cognitive and language development has also been observed (Lin et al. 2013). Water contaminated with Mn is one potential exposure pathway (Bouchard et al. 2011; Wasserman et al. 2006). However, exposure may also occur through airborne Mn released from Mn mining operations (Riojas-Rodriguez et al. 2010), ferro-manganese production facilities (Haynes et al. 2010; Lucchini et al. 2012; Menezes-Filho et al. 2009) or agricultural use of Mn containing fungicides (Gunier et al. 2013; Takser et al, 2004).

House dust serves as a reservoir for persistent compounds including metals (Whitehead et al. 2013). House dust is an important exposure pathway for young children because they spend more time close to the ground and have greater hand to mouth activity (Roberts et al. 2009). Lead dust loading, the amount of lead per area of floor space, has been shown to be a significant predictor of blood lead levels in children (Lanphear et al. 1995). Previous studies have demonstrated that levels of pesticides are higher in house dust of homes with farmworkers (Curl et al. 2002; Harnly et al. 2009) and residences located near agricultural pesticide applications (Fenske et al. 2002; Gunier et al. 2011; Harnly et al. 2009; Lu et al. 2000; Ward et al. 2006).

Maneb and mancozeb are fungicides that contain approximately 21% Mn by weight (FAO 1979). A study conducted in Quebec, Canada, found that self-reported proximity to agricultural pesticide applications was related to higher blood Mn (Takser et al. 2004). Agricultural use of these Mn fungicides exceeds 150,000 kg per year in the Salinas Valley of California with more than 90% used on lettuce (CDPR 2011). In a previous analysis, we found higher levels of Mn in shed teeth of children from the CHAMACOS study who lived within 3 km of agricultural applications of Mn fungicides, whose mothers were farmworkers, and among those with higher Mn dust loading in their homes (Gunier et al. 2013).

In the present study, we evaluated potential predictors of Mn concentrations and loadings in house dust from the homes of CHAMACOS participants, including agricultural work, proximity to agricultural use of Mn fungicides, soil type at the residence, and the amount of bare soil near the homes.

## 2. Material and Methods

### 2.1. Study population

Between September 1999 and November 2000, the Center for Health Assessment of Mothers and Children of Salinas or the CHAMACOS study enrolled 601 pregnant women from health clinics in the agricultural Salinas Valley primarily serving low-income families. Participants were eligible if they spoke English or Spanish and qualified for state funding of well-pregnancy care (within 200% of the Federal poverty level). A total of 513 participated in a prenatal home visit, of which 385 participated in a visit when the child reached six months of age. We included participants in this analysis (n=378) if there was adequate house dust sample available to measure Mn concentrations from either the prenatal (n=371) or six month home visit (n=7). Mothers included in this analysis were younger ($p<0.001$) and more likely to be nulliparous ($p=0.02$) than those with a prenatal visit that were not included. Residences included in this analysis were more likely to have a doormat ($p<0.0001$) and poor housekeeping practices ($p=0.001$) than residences not included, but there was no difference in the number of farmworkers in the home, agricultural use of Mn fungicides near the home prior to sample collection or residential location. For 90 homes, samples were analyzed from both prenatal and 6 month visits (postnatal) to assess Mn levels within homes over time. Written informed consent was obtained from all participants and all research was approved by the University of California, Berkeley Committee for the Protection of Human Subjects prior to commencement of the study.

### 2.2. Dust sample collection

We collected house dust samples using a High Volume Small Surface Sampler (HVS3, Envirometrics, Inc., Seattle, WA), which allows for the determination of dust loading in grams per square meter of floor area in order to better characterize Mn available for contact by children (Roberts et al. 2009). Details of our dust sample collection methods are provided elsewhere (Harnly et al. 2009); briefly dust samples were collected primarily from a one square meter area of floors. One dust sample per residence was collected for pesticide analyses. The pesticide dust sample was taken from (in order of priority): carpet in the central living area; if none then carpet in the bedroom; if none then bare floor in the central living area; if sample volume was judged inadequate then stuffed furniture in the central living area was sampled. We collected dust samples at mean of 18 (SD=6) weeks gestation (prenatal) and again when the child was 7 (SD=1) months old (postnatal). If an adequate amount of dust (> 1 g) could not be collected from the floor, furniture in the room was sampled using an attachment (n=7). Field interviewers collected information on recent cleaning and made an assessment of the overall quality of housekeeping (poor, average or excellent), recorded the type of surface sampled (floor, carpet or furniture) and measured the area of floor sampled in square meters. Samples were collected from the living room (n=403), child's bedroom (n=58) or other room (n=6). We defined season of dust sample collection as winter (December–February); spring (March–May); summer (June–August); and fall (September–November). Field staff also recorded the latitude and longitude coordinates of the residence using a hand-held global positioning system unit (eTrex, Garmin, Kansas, USA).

### 2.3. Laboratory analysis of dust samples

We stored dust samples at −80 °C for approximately 10 years before shipping them on dry ice for analysis. Samples were sieved to 150 μm and then weighed. We digested ~500 mg of each dust sample in 7.5 N nitric acid overnight and quantified Mn concentrations using inductively coupled plasma optical emission spectroscopy. We used Mn standards to develop a calibration curve and the procedural limit of detection (LOD) was 0.1 μg Mn/g dust and was derived via repeated analyses of analytical blanks within each analytical run using the formula three times the average standard deviation of blanks, then converting this analytical LOD to a procedural LOD using the typical dust sample weight that was processed, accounting for dilutions during processing. We analyzed 26 dust samples in triplicate to determine the reproducibility of Mn measurements and the coefficient of variation for Mn among these samples was 2.7%. We calculated Mn dust loading ($\mu g/m^2$) by multiplying the Mn concentration (μg/g) by the dust loading ($g/m^2$). Using the same methods, we also quantified cadmium (Cd), chromium (Cr) and lead (Pb) concentrations in dust samples for evaluation of our Mn prediction models.

### 2.4. Questionnaire data

Participants were interviewed shortly before and at the time of dust sample collection. Information obtained included demographic characteristics such as maternal age and education, maternal country of birth, parity, household income and number of people supported by this income, and housing density (number of persons/room). We also obtained visit specific information on the number of agricultural workers in the home, number of doormats at the home, housing type (house, apartment or condominium), number of pets in the home, whether an air conditioner was present in the home, smoking in the home and self-reported distance to the nearest agricultural field.

### 2.5. Geographic data

Based on the latitude and longitude coordinates, we used geographic information system (GIS) software (ArcInfo 10, ESRI, Redlands, CA) to evaluate possible geographic predictors of Mn levels in house dust, including region within the Salinas Valley (North County, Salinas, South County), population density, and median year built for homes in the census tract of the residence (U.S. Bureau of Census 2003). To account for variations in soil Mn concentrations, we linked each residence to detailed soil maps to identify the soil type (USDA 2009). We also used the National Elevation Data at 30 m resolution to assign an elevation to each home (USGS 2006).

The California Department of Pesticide Regulation maintains the California Pesticide Use Report (PUR) system (CDPR 2011). Pesticide applicators are required to report the active ingredient, quantity applied, acres treated, crop treated, date and location to one square mile in area (Public Land Survey Section (PLSS)) for all agricultural pesticide applications. We computed nearby maneb and mancozeb use for combinations of distance from the residence (buffer radii of 500, 1000 and 3000 meters) and time prior to dust sample collection (4, 8 and 16 weeks). We weighted fungicide use near homes based on the proportion of each square-mile PLSS that was within each buffer around a residence (Gunier et al. 2011). To account for the potential downwind transport of fungicides from the application site, we obtained data on wind direction from the five closest meteorological stations in the study area (CDWR 2013). We determined the direction of each PLSS centroid relative to residences and weighted fungicide use in a section by the percentage of time that the wind blew from that direction for each time period. We calculated the average wind speed and total precipitation during the 4, 8 and 16 weeks prior to dust sample collection using data from the nearest weather station.

We estimated the proportion of bare soil within 3000 meters of each residence from Landsat remote-sensing data images collected at three time points during dust sample collection: October 4, 1999, April 29, 2000 and October 22, 2000 (USGS 2013). We calculated the soil line index using the Landsat image produced closest in time prior to the dust sample collection by taking the ratio of Landsat band 4 (near infrared) to band 3 (red); bare soil was identified as areas with a ratio between 0.95 and 1.05 (Dematte et al. 2009). We used data from the only available particulate air pollution monitor in the Salinas Valley to determine the average particulate matter < 10 μm ($PM_{10}$) and particulate matter < 2.5 μm ($PM_{2.5}$) concentrations for the four and eight weeks prior to dust sample collection (CARB 2008). We also estimated Mn emissions from vehicle traffic at each residence by calculating the traffic density using previously published methods (Gunier et al. 2003) that involve summing vehicle kilometers traveled for all major roads (CDOT 2003) by the length of the road segments within 1,000 m of the residence.

### 2.6. Statistical analysis

Both Mn dust concentrations and loadings were not normally distributed based on results from the Shapiro-Wilk test for normality. Therefore, we used non-parametric methods including the Kruskal-Wallis test to evaluate bivariate relationships between categorical predictors and Mn dust concentrations and loadings, Spearman rank correlation coefficients to evaluate bivariate relationships with continuous predictor variables and Wilcoxon matched-pair signed-ranks test to compare prenatal and postnatal Mn dust concentrations and loadings in the 90 homes with repeat samples. The distribution of Mn dust concentrations was more normally distributed than the distribution of log-transformed concentrations; therefore we present results for the untransformed Mn concentrations. The distribution of Mn dust loadings was skewed to the right with a long tail; therefore we present results for the geometric mean (GM) Mn dust loadings. Variance components models with random intercepts for each residence were used to determine the intraclass correlation coefficient (ICC) of Mn dust concentrations and natural-logarithm transformed Mn dust loadings to assess the variability of Mn dust levels over time within a home. We used linear mixed-effects models with random intercepts to identify significant predictors ($p<0.1$) of Mn dust concentrations and natural-log transformed Mn dust loadings and estimate the amount of variability in measured levels explained by the model while accounting for the correlation among repeat samples collected from the same residence. We first identified potential explanatory variables that were associated with Mn dust concentrations or loadings with $p<0.2$ from bivariate analyses. We then used manual forward selection to derive final multivariate models including all explanatory variables that predicted Mn concentrations or loadings with $p<0.1$.

We constructed generalized additive models (GAM) with a 3-degrees-of-freedom cubic spline to evaluate the shape of the curves between predictor variables and Mn dust concentrations and loadings in our study population. None of the tests for digression from linearity were significant ($p<0.1$), suggesting that relations did not depart from linearity. We therefore included linear terms in multiple linear regression models. We evaluated outliers using standardized residuals from the multivariate mixed-effects models. There were no outliers from the Mn dust concentration and loading models with standardized residuals less than −3 or greater than 3, therefore we report results including all dust measurements. We conducted sensitivity analyses using log-transformed concentrations and loadings of Cd, Cr and Pb as the dependent variables in our final Mn dust models to evaluate whether predictor variables were associated specifically with Mn levels or related to dust concentrations and loadings of metals in general.

We performed a 10-fold cross-validation to evaluate whether our models were over fitting the data by setting aside 10% of the data and rerunning the models (Shao 1993). To further evaluate non-linear responses and interactions among predictor variables, we used a machine learning procedure called SuperLearner to estimate the prediction model (van der Laan et al. 2007). SuperLearner is a statistical package available in the programming language R (version 2.15.2, The R Foundation for Statistical Computing, 2012) that combines a set of candidate prediction algorithms specified by the user to create the best performing algorithm and performs a cross-validation procedure to assess the model fit. We used the following learners from R packages for the prediction model: generalized linear models (glm), generalized additive models (gam), Bayesian generalized linear models (bayesglm), random forest models (randomForest) and Lasso and elastic-net regularized generalized linear models (glmnet).

As an exploratory analysis and to visualize the spatial distribution of Mn dust loading in our study area, we created a map of predicted values using co-kriging methods. Kriging is a method of spatial interpolation used for predicting values in unmeasured locations using the spatial correlation as a function of distance between measurements and co-kriging combines information on other important predictor variables at that location (Stein and Corsten 1991). Our co-kriging model included soil type and agricultural use of Mn fungicides within 3 km of the residence as predictor variables. We assessed spatial autocorrelation of Mn dust concentrations and loadings as well as the residuals from our multivariable models using the global Moran's I coefficient, which is similar to other correlation coefficients with values between negative one and one and zero indicating no spatial autocorrelation.

# 3. Results

## 3.1. Distribution of metal concentrations and loadingss in dust

All house dust samples had detectable levels of Mn. The GM and geometric standard deviation (GSD) of Mn dust concentrations during the prenatal and postnatal visits were 150 (2) and 129 (2) μg/g of dust respectively (Table 1). The GM and GSD for Mn dust loadings were 435 (7) and 259 (7) μg/m$^2$ of floor area sampled at the prenatal and postnatal visits. The maximum Mn dust concentration was 414 μg/g and the maximum Mn dust loading was more than 25,000 μg/m$^2$. Among the 90 residences with dust samples collected at the prenatal and postnatal visits, both Mn dust concentrations ($p<0.001$) and loadings ($p<0.05$) were significantly higher at the prenatal visit. The intraclass correlation coefficients for residences with repeat samples collected were 0.58 for Mn dust concentration and 0.55 for Mn dust loading. The GM and GSD for Cd, Cr and Pb dust concentrations were 2 (2), 26 (2) and 49 (3) μg/g, respectively during the prenatal visit. The GM and GSD of Cd, Cr and Pb dust loadings were 6 (7), 76 (7) and 142 (8) μg/m$^2$, respectively during the prenatal visit.

## 3.2. Bivariate analyses of manganese predictors

Table 2 provides the levels of Mn dust concentrations and loadings from the prenatal visit (n=371) for categorical predictor variables. Concentrations and loadings of Mn in house dust were significantly higher in homes with farmworkers, residences located in the southern portion of the Salinas Valley, residences located on Antioch Loam soil and residences with poor housekeeping. There were also significantly higher Mn dust concentrations and loadings in homes where the mother was born in Mexico or had a 6$^{th}$ grade education or less. There was a difference in Mn dust concentrations by maternal age, but the relationship was inverse U-shaped with the highest mean concentration in the middle age group. The Mn dust loadings were higher in residences without a doormat, and in samples collected in the bedroom versus the living room or kitchen. There was no difference in Mn dust concentration or loading based on parity, family income or season of dust sample collection.

Table 3 presents the distributions of continuous predictor variables and Spearman correlation coefficients with Mn dust concentration and loading are presented. The Spearman correlation was strongest with the number of farmworkers in the home ($r_s=0.24$, $p<0.01$). There was a negative correlation between precipitation during the 8 weeks prior to dust sample collection and Mn dust concentration ($r_s=-0.13$, $p<0.01$) and Mn dust loading ($r_s=-0.12$, $p<0.05$), and a positive relationship with elevation for both Mn dust concentration and loading ($p<0.01$). Average wind speed during the 16 weeks prior to dust sample collection was correlated with Mn dust concentration ($r_s= 0.15$, $p<0.01$) and loading ($r_s= 0.13$, $p<0.01$). The median agricultural Mn fungicide use within 3 km of the residence during the 8 weeks prior to dust sample collection was 372 kg and the correlations with Mn dust concentration ($r_s=0.11$, $p=0.04$) and loading ($r_s=0.10$, $p=0.05$) were significant when agricultural Mn fungicide use was weighted by the proportion of time the wind blew from the direction of the section where the fungicides were applied. Traffic density within 1,000m of the residence, and $PM_{2.5}$ and $PM_{10}$ concentrations during the four weeks prior to sample collection were not correlated with either Mn dust concentrations or loadings. The correlation with Mn dust loading ($r_s=0.94$) was stronger with total dust loading (g/m$^2$) than with Mn dust concentration ($r_s$=0.62). Total dust loading (g/$m^2$) was also correlated with Mn dust concentration ($r_s$=0.40).

### 3.3 Multivariate analyses

Significant determinants of Mn dust concentration from multivariable linear mixed-effects models are presented in Table 4. Dust Mn concentrations were higher in residences in southern Monterey County (β=35 μg/g, 95% CI: 21, 50) and homes located on Antioch Loam soil (β=22 μg/g, 95% CI: 8, 36). There were no homes in southern Monterey County located on Antioch Loam soil so these predictors are independent. Mn dust concentration was also associated with greater house dust loading (g/$m^2$) with an increase of 10 μg/g dust (95% CI: 6, 13) per interquartile range increase (8.7 g/$m^2$) in dust loading. Dust Mn concentrations were higher in homes with poor housekeeping (β=18 μg/g, 95% CI: 9, 27), homes with at least one farmworker living in the residence (β=15 μg/g, 95% CI: 2, 29), and with greater wind weighted agricultural Mn fungicide use within 3 km during the 4 weeks prior to dust sample collection (β=6 μg/g, 95% CI: 1, 12). House dust loading explained the greatest amount of variability in Mn dust concentration (6.4%), followed by residence in the southern Salinas Valley (4.0%), poor or average vs. excellent housekeeping practices (3.6%), residence located on Antioch Loam soil (2.7%) and having a farmworker in the home (1.9%). In sensitivity analyses using concentrations of Cd, Cr and Pb, we found that residential location in the southern Salinas Valley was associated with higher concentrations of all four metals measured in dust.

The percentage change in Mn dust loading for select predictor variables are shown in Table 5 and Figure 1. Residence located in southern Monterey county was associated with an increase of 278% (95% CI: 143, 489) in Mn dust loading and residences located on Antioch Loam soil had 141% (95% CI: 61, 262) higher Mn dust loading. The Mn dust loading levels increased 13% (95% CI: 3, 23) per farmworker living in the home and 26% (95% CI: 6, 50) per interquartile range (37 kg) increase in wind weighted agricultural Mn fungicide use within 3 km of the residence during the four weeks prior to dust sample collection. Residences that did not have a doormat and residences with poor housekeeping practices also had higher Mn dust loading. In sensitivity analyses using dust loadings of Cd, Cr and Pb, we found that residential location in the southern Salinas Valley, housekeeping practices and residential location on Antioch Loam soil were related to dust loadings of all four metals measured. The cross-validation results showed that the models were not over fit, with the same independent variables significant ($p<0.1$) in each subset of the data, similar regression coefficients (±10%) and overall adjusted $R^2$ values for Mn dust concentration and loading.

Figure 2 shows the location of residences where prenatal dust samples were collected and estimated Mn dust loading levels from a universal co-kriging model with soil type and agricultural Mn fungicide use within 3 km of the residence included as predictor variables. Based on this exploratory analysis, the highest estimated Mn dust loading levels were in the southern Salinas Valley and the lowest levels were in the northwestern county. There was significant spatial autocorrelation of Mn dust loading (Moran's I=0.11, p=0.04), but no spatial autocorrelation for Mn dust concentrations or the residuals from the multivariable mixed effects models for Mn dust concentration or loading. The mean squared errors were similar using SuperLearner and linear regression models to predict Mn dust concentrations (3,887 vs. 3,903) and loadings (3.1 vs. 3.2), suggesting there were not strong interactions or other non-linear relationships that were missed using linear regression models.

## 4. Discussion

We found that farmworkers living in the home and agricultural use of Mn fungicides within 3 km of the residence during the month prior to dust sample collection were associated with higher Mn concentrations and loadings in house dust. These factors were specific to Mn levels in dust and were not associated with dust concentrations or loadings of Cd, Cr or Pb. Although Mn dust concentrations and loadings were higher in homes of women born in Mexico, there was a strong relationship between being born in Mexico and having a farmworker living in the home and therefore country of birth was not significant in multivariable the models. Our findings suggest that agricultural use of fungicides-containing Mn is a source of Mn in homes. In a previous analysis, we observed an association between levels of Mn in prenatal dentin of shed teeth from children and Mn dust loading from prenatal dust samples, suggesting that higher levels of Mn in the home increases exposure to pregnant women (Gunier et al. 2013). Our results add to the existing evidence that household proximity to agricultural pesticide applications and parental occupational take-home increases levels of pesticide constituents in the home (Curl et al. 2002; Fenske et al. 2002; Gunier et al. 2011; Harnly et al. 2009; Ward et al. 2006). Maneb, the primary Mn containing fungicide used in our study area during the study period from 1999 – 2001 (>95%), was not reregistered for sale in the U.S. after 2011 (USEPA 2011). Mancozeb, another Mn-containing fungicide, has largely replaced maneb use on lettuce in the Salinas Valley based on 2011 PUR data (CDPR 2014) and continues to be applied in the U.S. on other crops.

We identified several factors that were the largest predictors of increased Mn dust loading that also contribute broadly to increased dust loadings of metals. Residences located in the southern Salinas Valley had higher dust concentrations and loadings of Mn as well as Cd, Cr and Pb. The predominant wind direction in our study area is from the Pacific Ocean to the west and then down the Salinas Valley to the south, likely resulting in PM accumulating in air downwind of agricultural sources and higher background levels of metals in southern Salinas Valley air than in the northern Salinas Valley air, which is upwind of agricultural fields. We observed higher dust concentrations and loadings of Mn and other metals in residences located on Antioch Loam soil than residences located on other soil types. All soil contains naturally occurring Mn and other metals, and levels vary by soil type, but little information is available on differences in metal concentrations by soil type (ATSDR 2008). Homes with greater dust loading had higher average dust concentrations of metals suggesting that the additional dust entering homes is enriched in Mn and other metals from soil, agricultural use of Mn fungicides or other sources. We also found a strong, inverse linear relationship between dust loading levels of Mn and other metals in residences and more frequent or effective housekeeping practices and the number of doormats used at the home. Maternal education was associated with Mn levels in dust; however, lower maternal education was also related to poorer housekeeping practices and not having a doormat at the residence.

The dust sample collection methods also affected Mn dust loading including whether samples were collected from carpet or bare floors, and from the child's bedroom or the living room suggesting that the type of surface sample and location of dust sample collection can influence the concentration and loading of Mn in house dust. Single Mn dust measurements appear to be a reasonable long-term indicator of indoor levels with intraclass correlation coefficients among repeat dust samples collected 9 months apart of 0.6 for both concentrations and loadings, indicating moderate correlation of Mn levels within a home over time. Although moderately correlated, Mn concentrations decreased in the present study from the prenatal to postnatal visit.

In summary, we found that both Mn dust concentrations and loadings were related to the number of farmworkers in the home, agricultural use of Mn fungicides near the home, soil type at the residence, housekeeping practices and location of the residence in the southern Salinas Valley. Dust loading and whether the sample was collected during the prenatal or postnatal visit were also significant determinants of Mn dust concentrations, while having a doormat at the home was significantly associated with Mn dust loadings.

Table 6 summarizes median Mn dust concentrations measure in this study and previously published studies. In the present study, the median Mn dust concentration from samples collected during the prenatal home visit (178 μg/g) was higher than previous studies with no known source of Mn (Callan et al. 2013; Chattopadhyay et al. 2003; Kurt-Kurakos et al. 2012; Zota et al. 2011) except for a small study conducted in the United Kingdom (Turner and Simmonds 2006). Zota et al. observed a higher median Mn dust concentration (156 μg/g) among thirteen residences located 500 m or less from agricultural fields than residences located farther from fields, although specific information on pesticide use was not available and the median Mn concentration was lower than our study (Zota et al. 2011). Concentrations of Mn in house dust were higher than our study in other locations with potential Mn sources including residences located in a district with heavy industry and vehicle traffic in Hong Kong, China (Tong and Lam 2000) and in Ottawa, Canada where methylcyclopentadienyl manganese tricarbonyl (MMT) was added to gasoline (Rasmussen eta al 2001). Comparison of Mn dust concentrations across studies should be done cautiously because different dust extraction methods may have been used and dust with different particle size may have been included. The results from these studies provide evidence that nearby Mn emission sources increase Mn dust concentrations inside homes which is likely to lead to higher exposure from non-dietary ingestion, especially among young children, and may indicate higher exposure to Mn from inhalation as well.

The median Mn dust loading in our study was 488 μg/m$^2$, higher than levels reported (50 μg/m$^2$) in the only other study that measured Mn dust loading (Zota et al. 2011). The relatively higher Mn loadings in our study reflect the dusty conditions in this agricultural community with at least one farmworker living in most of the homes (79%). We observed a stronger correlation between Mn loading and total dust loading ($r_s = 0.94$) than between Mn loading and Mn dust concentration ($r_s = 0.62$). In a Canadian study of other metals in dust, stronger correlations were also observed between metal loading and dust loading ($r_s = 0.79$ – 0.92) than between metal loading and metal concentration ($r_s = 0.17$ – 0.58), indicating that dust loading is the primary factor driving metal loadings in homes (Rasmussen et al. 2013).

There were several limitations of this study. Our samples were collected from a relatively small geographic area in the Salinas Valley and from a homogeneous population of mostly Mexican-American farmworkers. We only had one dust sample for most homes with a second sample collected from 25% of the homes within about nine months; therefore longer-term variability could not be evaluated in our study. Because there was only one air monitor available that measured PM concentrations in our study area, we were only able to account for temporal variation in PM and not spatial variation. Another important limitation is that we did not collect air samples or soil samples to distinguish between deposition from windblown particles and dust tracked into the homes on shoes. We did not compare latitude and longitude coordinates collected using handheld global positioning system units to address geocoded locations and were not able to evaluate the accuracy of residential locations. As a result, there is likely to be some exposure misclassification in our estimates of proximity to agricultural Mn fungicide use due to measurement error. The identification of bare soil around residences using remote sensing data was challenging due to similarities between urban developed land and bare soil, differences in reflectance between dry and wet bare soil and complex cropping patterns present in the Salinas Valley. There was no spatial autocorrelation for Mn dust concentration among residences and only weak spatial autocorrelation for Mn dust loadings which suggests that differences in Mn levels in the homes were not strongly related to geographic factors. Finally, our models were only able to explain 20–30 % of the variability in Mn concentrations and loadings in dust.

Strengths of our study include the availability of comprehensive agricultural pesticide use data, a relatively large sample size and a dust sample collection method that allowed for the calculation of Mn dust loading in addition to Mn dust concentration. We had extensive interview data on potential Mn sources and detailed information on sample collection procedures. An additional strength was the use of GIS methods to estimate proximity to recent agricultural applications of Mn containing fungicides, traffic density, bare soil around the residence and soil type at the residence. We conducted extensive sensitivity analyses using advanced model selection and GIS techniques that further supported the assumption of linearity in the prediction models. Our results provide additional evidence that agricultural pesticide applications result in higher levels of these compounds in nearby homes, especially for pesticides that contain more persistent active ingredients such as Mn (Fenske et al. 2002; Gunier et al. 2011; Harnly et al. 2009; Lu et al. 2000; Ward et al. 2006).

Future studies should include house dust samples from non-agricultural areas for comparison. It would also be informative to collect and analyze air samples for $PM_{10}$ and Mn concentrations during the month prior to sample collection and to measure Mn concentrations in soil samples from the yard to evaluate the contribution of windblown dust from nearby agricultural fields in comparison to take-home dust from agricultural workers in the home. Air samples collected at participating homes would be helpful for assessing the spatial variability of windblown dust and contribution from longer range transport of particles into homes downwind of agricultural fields. Air samples could also potentially provide information for evaluating the differences in toxicity associated with exposure from inhalation compared to non-dietary ingestion.

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## 5. Conclusions

Concentrations and loadings of Mn in house dust were related to agricultural applications of Mn fungicides within 3 km of the residence during the month prior to dust sample collection and to the number of farmworkers living in the home. This study provides further evidence that pesticide levels could be reduced in homes by storing shoes and clothes of agricultural workers outside the home. Dust loadings of metals in general could be reduced by cleaning the home more frequently or effectively and using a doormat. Dust loadings of Mn and other metals were also related to soil type at the residence and location of the home within the Salinas Valley.

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## Abbreviations

| | |
|---|---|
| **Cd** | cadmium |
| **CHAMACOS** | Center for the Health Assessment of Mothers and Children of Salinas |
| **Cr** | chromium |
| **GIS** | geographic information system |
| **GM** | geometric mean |
| **GSD** | geometric standard deviation |
| **LOD** | limit of detection |
| **MMT** | methylcyclopentadienyl manganese tricarbonyl |
| **Mn** | manganese |
| **Pb** | lead |
| **PM** | particulate matter |
| **PUR** | pesticide use report |

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## Highlights

- Manganese dust concentrations and loadings were higher in farm worker residences.
- Manganese dust levels were higher in homes near applications of Mn fungicides.
- Dust metal loadings were related to soil type and housekeeping practices.
- Dust metal loadings were higher in homes located downwind of agricultural fields.

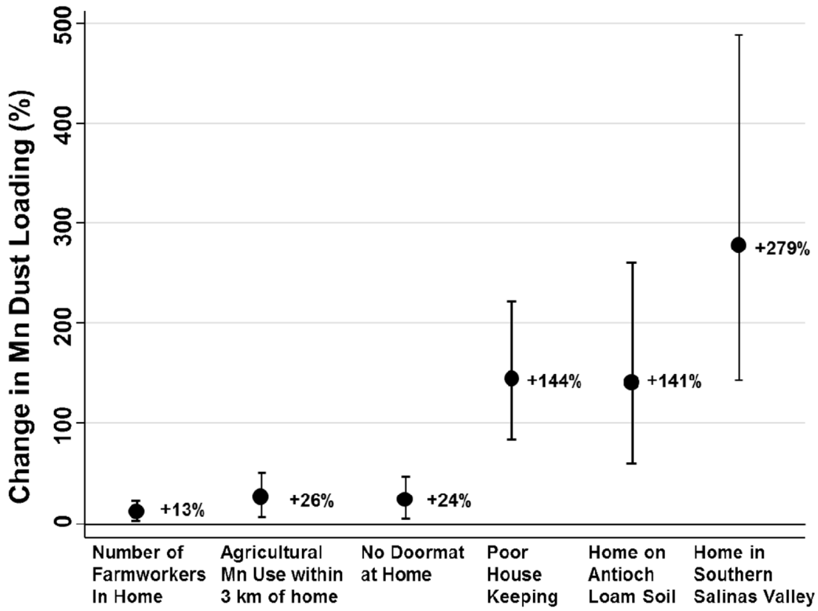


**Figure 1.**

Percentage change in manganese dust loading (μg/$m^2$) for select predictor variables estimated from multivariable mixed effects models (n=464).

*Adjusted for maternal education, room and surface sampled.

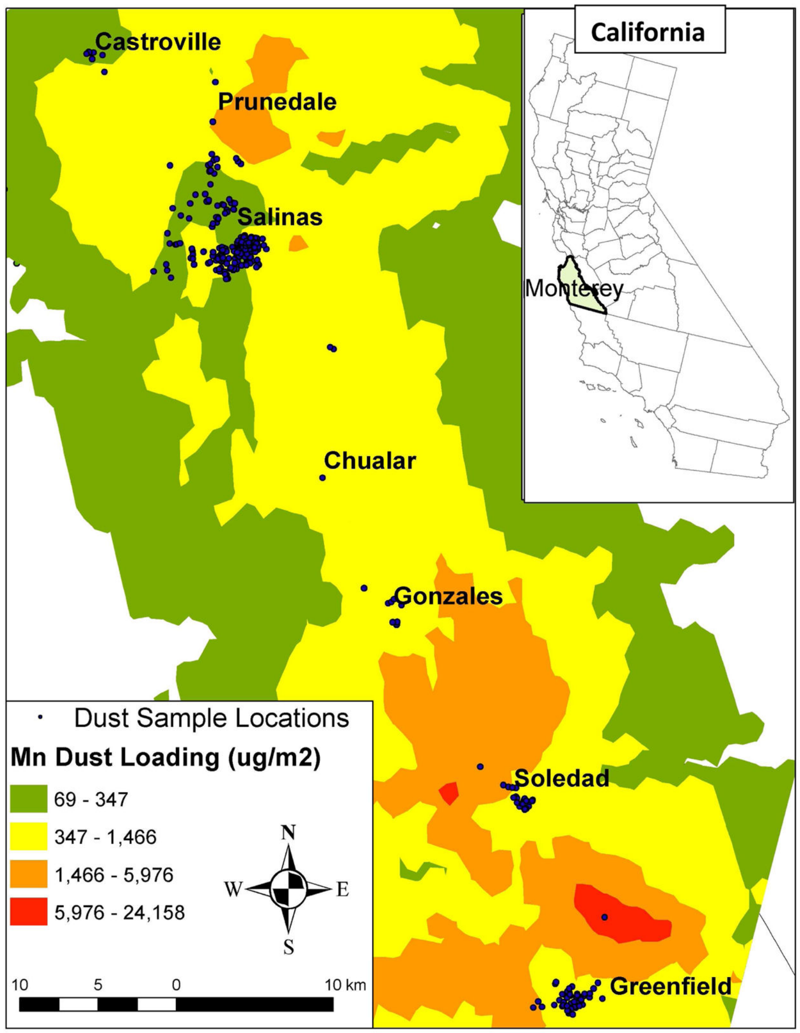


**Figure 2.**

Predicted manganese dust loading (μg/m$^2$) from a co-kriging model for prenatal visit (n=371) in the Salinas Valley, California.

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## Table 1

Distributions of manganese dust concentrations (μg/g) and loadings (μg/$m^2$) by home visit for CHAMACOS study cohort.

| Variable | N | Mean (SD) | GM (GSD) | Min | 25$^{th}$ | 50$^{th}$ | 75$^{th}$ | Max. |
|---|---|---|---|---|---|---|---|---|
| Prenatal Visit Mn Concentration | 371 | 171 (68) | 150 (2) | 2 | 134 | 178 | 209 | 414 |
| Postnatal Visit Mn Concentration | 97 | 145 (53) | 129 (2) | 10 | 119 | 148 | 181 | 239 |
| Prenatal Visit Mn Loading | 371 | 1,910 (3,600) | 435 (7) | 1 | 117 | 488 | 1,740 | 25,200 |
| Postnatal Visit Mn Loading | 97 | 1,040 (2,070) | 259 (7) | 1 | 122 | 271 | 710 | 10,600 |

## Table 2

Distributions of manganese dust concentrations and loadings from prenatal home visit (n=371) by demographic and residential characteristics.

| Characteristic | N (%) | Mn Concentration (μg/g) Mean (SD) | Mn Loading (μg/m$^2$) GM (GSD) |
|---|---|---|---|
| Mother's Age | | | |
| 18 – 24 | 184 (50%) | 172 (69) | 434 (8) |
| 25 – 29 | 125 (34%) | 178 (62) | 518 (7) |
| 30 – 45 | 62 (16%) | 153 (77)* | 270 (8) |
| Mother's birth country | | | |
| Mexico | 312 (84%) | 174 (67) | 496 (7) |
| United States/other | 59 (16%) | 152 (73)* | 262 (7)** |
| Mother's Education | | | |
| ≤ 6$^{th}$ grade | 156 (42%) | 182 (69) | 662 (8) |
| > 6$^{th}$ grade | 215 (58%) | 162 (66)** | 324 (7)** |
| Parity | | | |
| 0 | 142 (38%) | 172 (69) | 422 (7) |
| ≥ 1 | 229 (62%) | 170 (68) | 438 (8) |
| Family Income | | | |
| ≤ Poverty line | 220 (60%) | 173 (68) | 518 (8) |
| > Poverty line | 149 (40%) | 167 (70) | 351 (7) |
| Housekeeping Quality | | | |
| Poor or average | 243 (65%) | 182 (67) | 654 (5) |
| Excellent | 128 (35%) | 149 (66)** | 184 (9)** |
| Number of doormats | | | |
| 0 | 84 (23%) | 184 (66) | 665 (7) |
| 1 | 134 (36%) | 163 (64) | 446 (6) |
| 2+ | 153 (41%) | 171 (72) | 334 (7)* |
| Surface sampled | | | |
| Carpet | 323 (87%) | 171 (70) | 493 (7) |
| Bare floor/furniture | 48 (13%) | 169 (53) | 245 (9) |
| Room sampled | | | |
| Bedroom | 49 (13%) | 178 (56) | 812 (9) |
| Living room/kitchen | 322 (87%) | 170 (70) | 403 (7)* |
| Antioch Loam Soil | | | |
| Yes | 59 (16%) | 193 (61) | 898 (5) |
| No | 312 (84%) | 167 (69)** | 370 (7)** |
| Residential Location | | | |
| North County | 15 (4%) | 129 (65) | 83 (15) |
| Salinas | 248 (67%) | 164 (67) | 369 (7) |
| South County | 106 (29%) | 194 (65)** | 797 (7)** |

| Characteristic | N (%) | Mn Concentration (μg/g) Mean (SD) | Mn Loading ($\mu g/m^2$) GM (GSD) |
|---|---|---|---|
| Farmworker in Home | | | |
| Yes | 291 (79%) | 178 (66) | 536 (7) |
| No | 79 (21%) | 146 (70)** | 198 (8)** |

Kruskal-Wallis rank test

* p<0.05;

** p<0.01

## Table 3

Distributions of continuous predictor variables and Spearman correlations with manganese dust concentration (μg/g) and loading (μg/m$^2$) from prenatal visit (n=371).

| Variable | Median ($25^{th}$ – $75^{th}$) | Spearman Correlation Coefficient | |
|---|---|---|---|
| | | Mn Concentration (μg/g) | Mn Loading (μg/m$^2$) |
| Housing density (persons/room) | 2 (1 – 2) | 0.06 | 0.11* |
| Number of farmworkers | 1 (0 – 3) | 0.24** | 0.24** |
| Census tract population density (persons/mi$^2$) | 2,940 (103 – 5,560) | −0.12* | −0.06 |
| Precipitation 4 weeks (mm) | 5 (0 – 25) | −0.11* | −0.05 |
| Precipitation 8 weeks (mm) | 16 (4 – 51) | −0.13** | −0.12* |
| Elevation (m) | 28 (25 – 56) | 0.25** | 0.21** |
| Agricultural Mn Pesticide Use 500 m, 4 weeks (kg) | 0 (0 – 2) | 0.10* | 0.11* |
| Agricultural Mn Pesticide Use 500 m, 8 weeks (kg) | 1 (0 – 5) | 0.06 | 0.05 |
| Agricultural Mn Pesticide Use 1000 m, 4 weeks (kg) | 3 (0 – 12) | 0.14** | 0.11** |
| Agricultural Mn Pesticide Use 1000 m, 8 weeks (kg) | 10 (1 – 26) | 0.08 | 0.05 |
| Agricultural Mn Pesticide Use 3000 m, 4 weeks (kg) | 177 (19 – 348) | 0.11* | 0.08 |
| Agricultural Mn Pesticide Use 3000 m, 8 weeks (kg) | 372 (97 – 665) | 0.12* | 0.09 |
| Agricultural Mn Pesticide Use Wind weighted 3000 m, 4 weeks (kg) | 14 (0 – 37) | 0.12* | 0.09† |
| Agricultural Mn Pesticide Use Wind weighted 3000 m, 8 weeks (kg) | 39 (10 – 75) | 0.11* | 0.10* |
| Traffic density (VKT) 1000 m | 1,370 (430 – 48,600) | −0.06 | 0.01 |
| Particulate Matter < 2.5 μm 4 weeks (μg/m$^3$) | 7 (6 – 9) | 0.02 | 0.03 |
| Particulate Matter < 10 μm 4 weeks (μg/m$^3$) | 18 (15 – 22) | 0.04 | 0.06 |
| Percentage of Bare Soil 3000 m (%) | 17 (14 – 22) | −0.01 | 0.00 |
| Dust loading (g/m$^2$) | 4 (1 – 11) | 0.40** | 0.94** |

*p<0.05;

**p<0.01

**Table 4**

Determinants of manganese dust concentrations (μg/g) from mixed-effects model using samples collected at prenatal and postnatal visits (n=465).

| Predictor Variable | β (95% CI) | p-value | Partial $R^2$ |
|---|---|---|---|
| Dust loading (per IQR=8.7 g/$m^2$) | 10 (6, 13) | <0.0001 | 6.4% |
| Residence in South County | 35 (8, 36) | <0.0001 | 4.0% |
| Housekeeping (Poor/average vs. excellent) | 18 (9, 27) | <0.0001 | 3.6% |
| Antioch Loam soil type (yes vs. no) | 22 (8, 36) | 0.002 | 2.7% |
| Farm worker in the home | 15 (2, 29) | 0.02 | 1.9% |
| Wind weighted Agricultural Mn Use 3000 m, 4 weeks (per IQR =37 kg) | 6 (1, 12) | 0.03 | 0.9% |
| Sample collection time (Prenatal vs. postnatal) | 20 (10, 31) | <0.0001 | 0.9% |
| Overall model | | <0.00001 | 22.2% |

## Table 5

Determinants of manganese dust loadings (μg/$m^2$) from mixed-effects model using samples collected at prenatal and postnatal visits (n=465).

| Predictor Variable | % Change (95% CI) | p-value | Partial $R^2$ |
|---|---|---|---|
| Housekeeping (Poor/average vs. excellent) | 144 (85, 222) | <0.0001 | 8.1% |
| Residence in South County | 278 (143, 489) | <0.0001 | 7.5% |
| Antioch Loam soil type (yes vs. no) | 141 (61, 262) | <0.0001 | 5.4% |
| Farm worker in the home | 13 (3, 23) | 0.007 | 1.4% |
| Wind weighted Agricultural Mn Use 3000 m, 4 weeks (per IQR =37 kg) | 26 (6, 50) | 0.009 | 1.4% |
| Doormat used (yes vs. no) | 24 (4, 48) | 0.02 | 1.3% |
| Overall model | | <0.0001 | 27.0% |

* Adjusted for maternal education, room and surface sampled.

## Table 6

Summary of studies that have measured manganese concentrations in house dust.

| Study and Year Published | Location of Residences | Samples | Median Mn (μg/g) | Presumed Sources of Mn |
|---|---|---|---|---|
| Gunier et al. 2013 | Salinas, California | 371 | 178 | Agricultural fungicides |
| Gunier et al. 2013 | Salinas, California | 291 | 182 | Farmworker in home |
| Gunier et al. 2013 | Salinas, California | 106 | 198 | Located in southern County |
| Callan et al. 2013 | Western Australia | 156 | 53[a] | None |
| Kurt-Kurakos et al. 2012 | Istanbul, Turkey | 39 | 136 | None |
| Zota et al. 2011 | Ottawa, Oklahoma | 55 | 121 | None |
| Zota et al. 2011 | Ottawa, Oklahoma | 13 | 156 | Agricultural fields < 0.5 km |
| Turner et al. 2006 | United Kingdom | 32 | 501 | None |
| Chattopadhyay et al. 2003 | Sydney, Australia | 82 | 48 | None |
| Rasmussen et al. 2001 | Ottawa, Canada | 48 | 267 | MMT in gasoline |
| Tong et al. 2000 | Hong Kong, China | 151 | 216 | None |
| Tong et al. 2000 | Hong Kong, China | 34 | 283 | Heavy industry and traffic |

[a]Geometric mean concentration, median not reported.