



House Memorial 42: Parkinson's Disease and Pesticide Exposure

A Review of the Association between Pesticide Exposure and
Parkinson's Disease

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Executive Summary

The NMDOH developed conclusions and recommendations about the potential association of pesticide exposure and Parkinson's disease (PD) based on the literature assessed. Two main categories were analyzed: studies without genetic interactions and studies with genetic interactions. Within these two categories, the following categories of pesticides were identified: 1) general, 2) organochlorine, 3) organophosphorus, 3) botanical, 4) quaternary ammonium, and 5) carbamate or dithiocarbamate. There were many ways in which the reviewed studies described exposure to pesticides, including spraying pesticides, handling or direct contact, using or applying pesticides, performing jobs likely involving pesticide use, or unspecified exposure such as proximity to agricultural spraying.

To assess the existing epidemiological evidence for the association between exposure to pesticides and PD that could pertain to specific potentially exposed populations, the reviewed studies were grouped into two broad exposure/use categories: 1) exposure with pesticide use specified, including general use (occupational or residential use could not be distinguished), agricultural use (including farming), other occupational use, and residential use (including gardening), and 2) exposure with pesticide use not specified, including in an agricultural setting, in another occupational setting, in proximity to a pesticide use area, and in a residential setting.

Because there are no available data about the specific pesticides historically utilized, the amount, and where these pesticides were used, assessing the risk of pertinent populations in New Mexico is not currently possible. However, the following conclusions and recommendations apply to anyone in New Mexico who uses pesticides.

1. Pesticide Exposure without Genetic Interactions

Of the selected studies which addressed exposure to pesticides and the risk of PD, the majority of the evidence suggested an association between PD and pesticide exposure, including: herbicides and insecticides as well as chemical groups of pesticides such as organochlorine (lindane), organophosphorus (chlorpyrifos), quaternary ammonium (paraquat), botanicals (rotenone), and a mixture of dithiocarbamate (maneb) with paraquat. Furthermore, there appears to be good epidemiologic evidence for the association between the general pesticide use category and PD development. The association between PD and more specific pesticide use settings appears to be inconclusive for 1) agricultural use of pesticides, which included farming, 2) other occupational use, and 3) residential use of pesticides, which included gardening. The majority of the studies falling into the exposure with pesticide use not specified category provided inconsistent and inconclusive evidence for

the association with PD development. This included pesticide exposure with use not specified in agricultural, other occupational and residential settings as well as proximity to a pesticide use area. However, there were other factors which were stronger predictors for PD including family history of PD, head trauma, and lack of smoking. **Recommendations:** 1) Individuals who decide to use pesticides should first protect themselves and others from exposure by following the directions for application that are appropriate for the given pesticide. Gardeners who want to reduce the amount of pesticides they use may wish to learn more about Integrated Pest Management (IPM) principles:

http://aces.nmsu.edu/pubs/_circulars/cr-655/welcome.html. 2) Under the federal Worker Protection Standard, agricultural workers and pesticide handlers must be trained and informed about pesticides used on the establishment. Violations should be reported to NMDA.

2. Pesticide Exposure with Genetic Interactions

Among selected studies which addressed the effect of exposure to pesticides on genetically susceptible individuals/populations for the development of PD, the majority of the evidence on the genes studied suggested that genes modulated the risk between pesticide exposure and the risk of PD. These studies suggested that genotypes may interact with pesticide exposure to increase the risk for PD. Specific pesticides involved in gene-environment interactions with the development of PD included organophosphorus insecticides (specifically, chlorpyrifos and diazinon), the quaternary ammonium herbicide paraquat, and a mixture of maneb (dithiocarbamate) with paraquat. Specific genotypes implicated in these interactions included *CYP2D6*, *MDR1*, *MnSOD*, *NQO1*, *NOS1* SNPs, *GSTT*, *PON1*, *DAT*, and *GSTT1*. Other factors that play a role in PD include family history of PD, head trauma, and smoking status. Some individuals may be more susceptible to PD with pesticide exposure than others based on specific genes. **Recommendation:** 1) Individuals who decide to use pesticides should first protect themselves and others from exposure by following the directions for application that are appropriate for the given pesticide. Gardeners who want to reduce the amount of pesticides they use may wish to learn more about Integrated Pest Management (IPM) principles: http://aces.nmsu.edu/pubs/_circulars/cr-655/welcome.html. 2) Under the federal Worker Protection Standard, agricultural workers and pesticide handlers must be trained and informed about pesticides used on the established. Violations should be reported to NMDA.

Background

2013 Legislative Session

During the 2013 legislative session, House Memorial 42 (HM 42) passed, which identified public concerns about pesticide exposure and the development of Parkinson's disease (PD). This memorial requested a review of the historical agricultural use of pesticides by the New Mexico Department of Agriculture (NMDA) and a review of the pertinent scientific literature on the relationship of pesticide use and the development of PD by the New Mexico Department of Health (NMDOH). Using this information, the NMDOH was directed to prepare a report assessing the risk of pertinent populations of New Mexicans with recommendations, as appropriate. This document incorporates information provided by NMDA and provides the methods utilized to conduct a systematic review of the literature as well as the resulting findings and recommendations.

Federal Regulation of Pesticides

The United States federal government passed the first laws regulating pesticides in 1910. The Insecticide Act, as it was known, was intended to protect consumers from impure or fraudulently labeled insecticides and fungicides. The Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) replaced it in 1947, requiring for the first time that all pesticides be registered and labeled according to minimum standards. It wasn't until 1970 that Congress created the US Environmental Protection Agency (EPA) and transferred administration of FIFRA to it from USDA.

Since then there have been many changes to the laws governing the registration, labeling, and use of pesticides. Before 1974, there were no standards for worker reentry into pesticide treated fields, and thus the agricultural worker protection standard was passed in 1992. Many pesticides have disappeared and new, safer chemicals – for pesticide workers, the public, and the environment – have been developed. In general, modern pesticides are effective at lower rates, are safer to apply, and have fewer side effects or unintended consequences in the environment.

Under FIFRA, EPA registers pesticides and prescribes label directions to prevent unreasonable adverse effects on human health or the environment. Under the Federal Food, Drug and Cosmetic Act (FFDCA), EPA establishes tolerances (maximum legally permissible levels) for pesticide residues in food. Tolerances are enforced by the Food and Drug Administration's Health and Human Services Department and the US Department of Agriculture's Food Safety and Inspection Service. In 1996, Congress passed the Food Quality Protection Act (FQPA). This law sets a "reasonable certainty of no harm" standard when setting tolerances for pesticides in food. It requires EPA to periodically re-evaluate existing registrations and tolerances to make sure they meet the requirements of current standards. By 2006

EPA reviewed nearly 10,000 tolerances, recommending the revocation or modification of 4,600 tolerances and confirming the safety of 5,200 tolerances.

Agricultural workers and pesticide handlers are protected under the federal Worker Protection Standard, which requires employers to train and inform their people about the pesticides used on the establishment. Appendix I, provided by the NMDA, lists the pesticides of most interest to the public group Pesticides and Parkinson's Committee (PPC), the class of the pesticide, and historical use. Therefore, the list is not meant to be comprehensive of all pesticides ever used. Additionally, the NMDA does not have information which would allow an historical assessment of how much of each pesticide was used and where. Appendix III, also provided by the NMDA is a report conducted by the United States Geological Survey (USGS). The document describes a method which could be used to calculate annual county-level pesticide use for selected herbicides, insecticides, and fungicides applied to agricultural crops grown in the conterminous United States from 1992 through 2009.

Pesticide Regulation in New Mexico

In New Mexico, pesticides and pesticide applicators are regulated by the Pesticide Control Act, administered by the NMDA. Rules promulgated under this law govern pesticide registration, the licensing of pesticide applicators, recordkeeping requirements, standards for use, storage and disposal of pesticides, penalties for noncompliance, inspection of application equipment, and applicator safety. Pesticides comprise a wide range of substances designated to deter or kill insects (insecticides), rodents (rodenticides), plants (herbicides), fungi (fungicides), etc. Pesticides also have subclasses based on their chemical components (e.g., organophosphate) or their application method (e.g., fumigant).

Registration

All pesticides must be registered with NMDA before they can be distributed or used in the state. Any product that claims to control, mitigate, or repel a pest is a pesticide, including insecticides, herbicides, fungicides, rodenticides, disinfectants, sanitizers, insect repellents, microbial pesticides, desiccants, and more. This includes consumer products like insect repellent, weed killers, sanitizers, wasp sprays, etc., purchased at big box stores, grocery stores, and drug stores, in addition to pesticides purchased and used by agricultural producers and professional pest control applicators. There are approximately 11,000-12,000 pesticides registered annually with NMDA. These products are formulated from an estimated 1,100 active ingredients.

Registered pesticides may be restricted – available for purchase and use only by certified applicators – by either federal or state law. Products restricted by federal law are automatically

restricted in the state. Certain 2,4-D herbicides that are not federally restricted are restricted in New Mexico by state law. About eight percent of federally registered pesticides are restricted.

Licensing

Any person who wants to purchase or use a restricted pesticide must be certified and licensed through NMDA. In addition, anyone who applies any pesticide for hire must hold a commercial applicator or operator/technician license. Currently there are about 7,600 individuals licensed in New Mexico. Nearly 3,000 of those individuals are licensed with commercial businesses that perform pest control for hire.

Federal and state laws require applicators to 1) use only registered pesticides and to follow all label directions 2) keep records of pesticide applications 3) only apply pesticides in categories they are certified in; and more.

Inspection/Enforcement

Federal and state laws require that dealers sell restricted pesticides to licensed applicators only. Applicators must follow all label directions; keep records of their pesticide applications; only apply pesticides in categories they are certified in; not be careless or negligent in their use of pesticides; and more.

NMDA inspects those businesses that sell pesticides. In addition to pesticide dealers who sell restricted pesticides, markets like feed stores, hardware stores, box stores, discount stores, garden centers, and others are inspected to make sure the pesticides for sale are registered and are displayed, stored and sold safely and in accordance with federal and state laws.

NMDA inspects pesticide applicators. Inspectors verify they are applying pesticides in accordance with the product labels and with state and federal law. Agricultural establishments are also inspected to assure compliance with the Worker Protection Standard. Although some inspections are conducted during a pesticide application, most compliance monitoring is based on the records kept by the applicators.

NMDA investigates an average of about 35 complaints a year alleging pesticide misuse, drift, or other violations. When violations can be documented NMDA takes enforcement action against the applicator. Complaints may involve damage to non-target plants, personal exposure, ineffective pest control, or licensing issues.

Methods

Literature Search

The NMDOH conducted a systematic search for relevant information in the scientific literature via the search engine PubMed (<http://www.ncbi.nlm.nih.gov/sites/entrez?db=PubMed>). In order to reduce the chance that information supporting a particular conclusion would be preferentially identified while other information was missed, the literature search was designed to be as comprehensive as possible. The search was conducted in July 2013 to identify all peer-reviewed articles published before that date. Key and free-text words included “Parkinson’s disease,” “pesticide,” “insecticide,” “fungicide,” and “rodenticide.” In addition to the articles identified through PubMed, the public group PPC also submitted articles for potential inclusion. In order to allow enough time to review articles, the deadline for the public to submit articles was August 2, 2013.

Inclusion and Exclusion Criteria

Because the focus of HM 42 is on the association between pesticide exposure and PD development, the NMDOH included studies (whether through PubMed or submitted by the public) if they addressed health outcomes among people. To be included, the studies had to address not only PD but also pesticide exposure. For this review, articles were included if they focused on PD. The signature signs and symptoms of PD include bradykinesia,¹ resting tremor,² cogwheel rigidity,³ and postural reflex⁴ impairment. Only peer-reviewed journal articles, government agency reports, and reports from nationally or internationally recognized organizations and authorities in public health such as the World Health Organization (WHO) were included for review. The scientific peer-review process ensures the quality and relevance of research activities, and helps maintain scientific objectivity and credibility. Government agencies have internal procedures and processes which guide reviews.

Articles were excluded if they met any of the following criteria: 1) animal model (anything but human), 2) cell lines or cell cultures, 3) treatment/protective factors, 4) no measures of association (odds ratio, relative risk), 5) miscellaneous (no pesticide exposure or PD not addressed; mechanism of action study; biomarkers of exposure; case report (report of a single case of PD); predictive toxicity (extrapolation of animal toxicity data to predict human effects or extrapolation of toxicity based on chemical structure); letter to the editor; summary of conference), or 6) article written in language other than English. Additionally, we excluded review articles that summarized original articles that we have

¹ Slowness of movement

² Tremor that occurs when muscle is relaxed

³ Increase in muscle tone causing resistance to externally imposed joint movements

⁴ Automatic movements that control equilibration when the body is upright and moving

already included, except for those reviews that attempted to evaluate the level of evidence for the association between exposure to pesticides and PD development.

Data Extraction Form

Eight epidemiologists from NMDOH extracted data from the studies selected for review and recorded the information on a standardized form (see end of report). Extracted data included information about study participants such as race/ethnicity, age, and sex, measure of association (odds ratio), statistically significant findings, and any recommendations the study made.

Data Synthesis

Two of the epidemiologists working on the data extractions also conducted the data synthesis and analysis. Data from the extraction form were summarized into two main categories of PD articles: 1) pesticide exposure without genetic interactions and 2) pesticide exposure with genetic interactions.

Results

Selection of Studies

Through the PubMed search, a total of 1547 potentially relevant publications were identified (Appendix II). In addition, 94 article titles were submitted by the PPC for review. Of these, 13 articles had been duplicated within the PPC document, 29 were excluded based on our criteria, and 52 were possible inclusions. Of the 52, 51 were already included in the PubMed search results. Thus one article was added to bring the total of articles to review to 1548. Of these, 1444 were excluded based on the previously identified inclusion/exclusion criteria. The final list of articles that were included for this review can be found in the References section of the report.

1. Pesticide Exposure without Genetic Interactions

Of the selected studies which addressed the association between pesticide exposure and Parkinson's disease without investigating genetic interactions, there were four categories: 1) pesticides (general): specific pesticide not identified (includes general categories such as insecticide, fungicide, herbicide), 2) organochlorine, 3) organophosphorus, 4) botanical, 5) quaternary ammonium, and 6) carbamate or dithiocarbamate.

There were many ways in which the reviewed studies described exposure to pesticides, including spraying pesticides, handling or direct contact, using or applying pesticides, performing jobs likely involving pesticide use, or unspecified exposure such as a proximity to agricultural spraying.

To assess the existing epidemiological evidence for the association between exposure to pesticides and PD that could pertain to specific potentially exposed populations, we grouped studies into two broad exposure/use categories: 1) exposure with pesticide use specified, including general use (occupational or residential use could not be distinguished), agricultural use (including farming), other occupational use, and residential use (including gardening), and 2) exposure with pesticide use not specified, including in an agricultural setting, in another occupational setting, in proximity to a pesticide use area, and in a residential setting.

The studies are summarized here by the type of pesticides evaluated for potential association with PD. The studies presenting positive associations between pesticide exposure and risk of PD are summarized first.

Pesticides (General)

Of the 61 studies analyzing an association between general exposure to pesticides and PD development, 35 provided statistically significant evidence to support this association and 26 studies did not have statistically significant evidence. Herbicides and insecticides as functional categories of pesticides each had the same number of studies associated with increased risk of PD (n=7). Out of the seven studies reporting increased risk of PD due to herbicide use, statistically significant ORs ranged from 1.33 to 4.1, with the highest OR involving occupational, long-term exposure greater than 20 years compared with no exposure. Among seven studies reporting increased risk of PD due to insecticide use, statistically significant ORs ranged from 1.53 to 5.81, with the highest OR involving exposure lasting 10 years or longer, as compared to those with less than 10 years' exposure. Fungicides had the least number of studies (n=2), with ORs of 2.2 and 5.6. The remainder of the studies with a positive association fell under the general pesticides category. Out of these studies, there were statistically significant ORs ranging from 1.21 to 17.12. However, the study finding the OR of 17.12 did not adjust for family history of PD, which had an OR of 21.4. The next highest OR was 5.63.

There appears to be good epidemiologic evidence for the association between the general pesticide use category and PD development; of the 26 studies falling into this exposure/use category, 17 studies reported a positive association with the risk for PD and 9 did not. The association between PD and pesticide use appears to be inconclusive for 1) agricultural use of pesticides, which included farming (of the 14 studies, 6 reported a evidence for the association and 8 did not), 2) other occupational use (of the 16 studies, 9 reported positive evidence and 7 did not), and 3) residential use of pesticides, which included gardening (of the 6 studies falling into this pesticide us category, 3 studies provided a positive association with PD and 3 did not).

The majority of the studies falling into the pesticide use not specified category provided no evidence for an association with PD. This included pesticide use not specified in agricultural, other occupational and residential settings as well as proximity to a pesticide use area.

For all of these studies, factors that tended to increase the risk of PD due to pesticide exposure included increased duration and frequency of exposure, family history of PD, and head trauma. Smoking appeared to act as a protective factor, decreasing the risk of PD. Studies with occupational exposure tended to have a longer duration/frequency of pesticide use compared to residential exposure such as gardening. Collectively, the studies support a multifactorial etiology of PD, influenced by family history of PD, head trauma, and pesticide exposure.

The limitation of the studies included exposure misclassification, recall bias, small sample size, and competing risk factors that may not have been taken into consideration through statistical analyses. For example, with respect to recall bias, many studies mentioned that PD patients were more likely to recall pesticide exposure compared to controls. For competing risk factors, at least one paper demonstrated that farming as an occupation was an independent risk factor for PD, regardless of pesticide exposure status.

Positive association

The studies which, for the most part, found a positive and statistically significant association between general pesticide exposure and risk of PD are summarized here. In some cases, however, the studies were mixed with both positive and no associations. A positive association means the authors found an increased risk [odds ratio (OR) or relative risk (RR) over one (1)] of PD among those exposed to pesticides compared to unexposed individuals. Confidence intervals (CI) that include one (1) indicate that the association is not statistically significant.

Golbe and co-authors (1990), conducted a case-control study, which mainly focused on nutritional/dietary risk factors for PD, however, they also investigated the association between PD and rural living or pesticide use. This study involved 106 PD patients with spouses as controls (71 males and 35 females). Pesticide exposure (assessed by a telephone-administered questionnaire) was positive if respondents answered yes to the question “sprayed pesticides or insect spray at least once a year for five years?” PD development was associated with pesticide exposure (OR=7.0, $p<0.05$).

Butterfield *et al.* (1993) conducted a case-control study of PD but instead focused on young-onset Parkinson’s disease (YOPD). The authors chose this population in order to minimize recall bias (shorter life history to remember) and on the premise that early PD may be associated with higher exposures. YOPD patients diagnosed before the age of 50 (n=63) were frequency-matched on sex, year

or birth, and year of diagnosis with rheumatoid arthritis patients (RA) in the Oregon-Washington region of the US. There was a higher proportion of YOPD cases with a family history of PD and more non-whites among RA controls than the YOPD group. A questionnaire was used to assess exposure and employment history. YOPD subjects with a history of farm employment or residency, had a mean of 10.6 years of exposure compared with a mean of 6.4 years for controls (t -test $p=0.101$). Logistic regression models were used to estimate the YOPD risk associated with exposure to pesticides at a frequency of more than 10 times a year in any one year (insecticides, herbicides, and fumigants). The ORs were adjusted for age, age at diagnosis, race, sex, educational level and family history of PD. The risk for YOPD was statistically significantly associated with exposure to three pesticide classes: herbicides (OR=3.46, $p=0.011$; adjusted odds ratio (aOR)=3.22, $p=0.033$), past residency in a fumigated house (OR=3.29, $p=0.068$; aOR=5.25, $p=0.045$) and exposure to insecticides (OR=4.04, $p=0.002$; aOR=5.75, $p=0.001$). There was no statistically significant association between YOPD development and exposure to fungicides (crude OR=1.50, $p=0.742$), rodenticides (OR=2.42, $p=0.618$), and “ever lived within ¼ mile of agricultural spraying?” (OR=1.99, $p=0.106$). Logistic regression analysis comparing the relative strength of association between insecticides and herbicides exposure found adjusted ORs of 4.84 for insecticides ($p=0.008$) and 1.53 for herbicides ($p=0.510$), when both variables were modeled simultaneously.

Semchuk *et al.* (1993) conducted a population-based, case-control study to test the hypothesis of multi-factorial etiology of PD development, using occupational and chemical exposure data, medical, and smoking history data, and family history of PD and essential tremors in Calgary, Canada. There were 130 cases (75 males and 55 females) with neurologist-confirmed idiopathic PD and two community controls of 260 subjects (150 males and 110 females) matched by sex and age (± 2.5 years). Exposure and family history information was obtained by personal interview. Occupational and chemical exposures included exposure to pesticides (herbicides, insecticides, and fungicides), work-related exposure to aluminum, carbon monoxide, cyanide, manganese, mercury, mineral oils, and ionizing radiation. Information on family history of PD and essential tremor, smoking, and history of various viral and other medical conditions was collected together with a detailed lifetime occupational history, including exposure dates and descriptive information on all work-related contact with pesticides and other chemicals. Occupational and chemical exposures were coded for the period between the subject’s 16th and 55th birthdays and for each 10-year age interval between the subject’s 16th and 55th birthdays. Both univariate and multivariate conditional logistic regression analyses methods were used to estimate the risk for PD (crude and adjusted ORs, respectively) associated with the following

variables: family history of PD, previous head trauma, exposure to herbicides, family history of essential tremor, and smoking status. The crude OR for herbicide exposure was 3.06 (CI=1.34-7.00, $p<0.01$). When adjusted for different combinations of the variables previously cited, the adjusted ORs ranged from aOR=2.83 (CI=1.13-7.06, $p<0.05$) to aOR=3.09 (CI=1.27-7.56, $p<0.05$). When all five variables were included in the model, family history of PD was the strongest predictor of PD risk followed by history of head trauma. The results of this analysis demonstrate that the ORs for PD remained at approximately three, even after controlling for the effects of the other variables, such as family history of PD and previous head trauma. The authors concluded that their results support the hypothesis of a multifactorial etiology of PD, which likely involves genetic, environmental, head trauma and possible other factors.

Hubble and co-authors (1993) conducted a small case-control study in Kansas involving two sites: one rural (Hays) and one urban (Kansas City) to investigate the effect of rural residency on the development of PD. In the urban population, there were 32 cases and 31 controls. In the rural population, there were 31 cases and 44 controls. All subjects were examined by a neurologist and the PD diagnosis was based on the presence of two or more of the cardinal signs of PD (i.e., tremor, rigidity, and bradykinesia) and responsiveness to the PD drug, levodopa. Patients with historical signs or symptoms of neurological disorders were excluded. Information on exposure factors was collected by self-administered questionnaire and included ever living or working on a farm, pesticide use for more than 20 days for any one year, pesticide use for more than 20 days for 5 years, living or working on a farm with livestock, smoking more than 100 cigarettes during a lifetime, head trauma, having received professional help for depression, and others. Lifetime occupational histories were obtained. Rural living was defined as residency in a town of less than 2500 population. Logistic regression modeling was conducted on the pooled data from urban plus rural sites. However, rural living was so common among subjects in the control (urban) site (51% had lived in a rural setting for more than 20 years) that it did not serve to distinguish controls from patients. On the other hand, pesticide exposure emerged as a variable distinct from rural living and served as a strong predictor of PD among these subjects. Three significant predictors of PD emerged in order of the strength of their association with PD: pesticide factor (used pesticides for more than 20 days in any given year and having done so for more than 5 years), reported family history of neurologic disorders, and history of depression. When all three predictors were positive, the probability for PD in the subject was 92.3% (OR=12.0). Pesticide use had an OR of 3.42 (CI=1.27-7.32, $p=0.0041$) in the stepwise logistic regression. Several variables were not predictive of PD: head trauma, ethnicity, history of central nervous system (CNS) infection, and fresh

produce consumption, but the sample size was also small. Recommendations included investigation into the identity of specific chemical agents that may be responsible for the development of PD and exploration of the nature of familial influence.

A case-control study conducted in a geographically defined horticultural region in Okanagan Valley, British Columbia aimed to identify possible risk factors for idiopathic PD (Hertzman *et al.*, 1994). There were 127 PD cases (71 males and 56 females) and 245 controls (140 males and 105 females), categorized into two groups: 121 subjects (80 males and 41 females) with chronic cardiac disease (CD) and 124 randomly selected voters (60 males and 64 females). There was approximately the same number of males and females within the different age groups. There were similar years of residence in the Okanagan Valley for all groups. The occupational exposure questionnaire included questions on 79 different agricultural chemicals used in the tree fruit industry, including: 1) any pesticide; 2) specific pesticides: paraquat, azinphos-methyl, ferbam, phosalone; and 3) pesticides grouped into: chlorophenoxy compounds (chlorophenoxy), organochlorines, organophosphates, carbamates, borates, copper salts, dithiocarbamates, insecticides, herbicides, fungicides, and acaricides. Exposure was defined as handling the chemical or working in an area that had been recently sprayed with the chemical. Full occupational history information was collected. Separate analyses were conducted for PD vs. CD and PD vs. voters groups using logistic regression and adjusting for sex and age. The study found a statistically significant association between PD in men and having an occupation in which there was probable exposure to pesticides through handling or direct contact. However, no specific pesticide exposure was associated with PD development for either gender. For men's exposure to pesticides, the aOR was 2.03 (CI=1.00-4.12) as compared with CD controls; aOR=2.32 (CI=1.10-4.88), compared with voters. For women's exposure to pesticides, aOR=1.11 (CI=0.32-3.80) for comparison with CD controls and OR=1.36 (CI=0.48-3.85) as compared with voters group. There were no statistically significant findings when exposure was grouped by specific pesticides or occupation. The authors concluded that although occupational exposure to agricultural chemicals may predispose individuals to the development of PD, the pathogenesis is multi-factorial rather than related to any specific chemical. The authors were concerned with recall bias and therefore they used two control groups, assuming that patients with chronic diseases may be more introspective and therefore, more likely to identify past exposures. They also noted that educational attainment may correlate positively with the ability to recall the name of the pesticide and negatively with the level of exposure to the pesticide. Thus, they recommended that future studies should control for these factors.

Semchuk and Love (1995) examined the effects of exposure misclassification in proxy-derived data on agricultural work, pesticide use, rural living, well water drinking, head trauma, smoking status, and family history of PD or essential tremor on Parkinson's disease risk estimates. The data were collected in 1989 as part of a population-based case-control study of PD in Calgary, Canada. For each neurologist-confirmed PD case, two matched (by sex and age) community controls were randomly selected. Forty cases and 77 controls were randomly selected as index respondents. The cases, controls, and one proxy respondent (spouse or offspring) for each index respondent were interviewed using a structured questionnaire. The data were analyzed using conditional logistic regression. Odds ratios estimated the risk of PD associated with environmental variables (rural living, farm living and well water drinking), agricultural variables (agricultural work, crop farming, grain farming, herbicide use, insecticide use, and fungicide use), and other variables (family history of PD, head trauma, family history of essential tremor, and smoking status). Incorporation of proxy-derived data for 30% of the cases and controls, or both, resulted in misclassification of exposure for some variables, in some cases leading to considerable attenuation of the ORs. The adjusted PD risk estimates were calculated for the three probable risk factors (herbicide use, family history, and head trauma) when only self-respondent data were used and when the analysis was replicated using proxy-derived data for about 30% of the cases and controls. Specifically, the study results show that the adjusted odds ratio for PD development and herbicide use was 3.09 (CI=1.27-7.56) when only self-respondent data were used for the analysis and the adjusted ORs remained reduced for the mixed self- and proxy-respondent data use situation. In summary, the study results indicate that pooling dichotomously classified data derived in part from self- and proxy respondents may result in biased estimates of PD risk associated with agricultural exposure (including pesticide use), family history of PD, and head trauma.

A case-control study conducted in Germany investigated a number of potential etiologic factors for PD development, including exposure to pesticides (use of herbicides, insecticides, organochlorines, alkylated phosphates and carbamates), farming activity, well-water drinking, exposure to wood preservatives, head trauma, the number of previous episodes of general anesthesia, and other factors such as the number of amalgam fillings (Seidler *et al.*, 1996). PD cases (n=380) from 9 neurological clinics across Germany were recruited for the study and 379 neighborhood controls and 376 regional controls were enrolled. The cases were determined by using the UKPDS Brain Bank clinical diagnostic criteria. A detailed residential history was taken from each participant. To assess pesticide exposure, participants were asked about years of pesticide application and frequency of use. Then, a toxicological expert categorized all pesticides named by subjects with regular pesticide use, which fell into five groups:

organochlorines, alkylated phosphates and carbamates, inhibitors of cellular metabolism (cellular respiration, enzyme activity, membrane excitation), and “other” category. Occupational exposure was assessed by "ever" versus "never" exposure to a list of neurotoxic substances. In addition, a job exposure matrix was constructed for job titles and industries named by participants. Smoking and educational status were included as covariates in the multivariate analyses to adjust for potential confounding; both variables were strongly inversely associated with PD. Therefore, ORs were adjusted for smoking and educational status, for patients vs. neighborhood control subjects, and for patients vs. regional control subjects. The study showed a significantly increased OR for pesticide usage and the risk of PD, but none for rural factors. Duration of herbicide and insecticide use prior to PD diagnosis showed a positive trend across the number of exposure years; however, the ORs and p-values for trend were statistically significant only in the comparison with regional controls. The study found that PD patients were more likely than control subject to have used herbicides (aOR significant only for comparison with regional control subjects for exposure duration of 41-80 years (aOR=3.0, CI=1.5-6.0), insecticide (aOR significant only for comparison with regional control subjects for exposure duration of 1-40 years (aOR=1.8, CI=1.1-2.7) and 41-80 years (aOR=2.5, CI=1.4-4.5). The authors also analyzed the risk of PD for organochlorine and organophosphate exposures. These results are provided in subsequent sections. A dose-response gradient was seen with increasing number of exposure years to wood preservatives (ever vs. never), but that trend was not statistically significant. The gradient was more pronounced when exposure was considered only for 15 years prior to PD diagnosis. The authors concluded that environmental factors may play a role in the etiology of PD, possibly acting through a genetic predisposition. They recommended conducting more precise exposure assessments, possibly involving biological monitoring for newly diagnosed PD cases as well as setting up a population-based PD registry to capture the disease incident rates, which would likely approximate a prospective assessment of PD risk factors.

A case-control study by Gorrel *et al.* (1998) investigated exposure to pesticides (herbicides, insecticides, and fungicides), farming, well water use, and rural living as the risk factors for PD. Cases and controls were drawn from residents of the tri-county metropolitan Detroit area who were receiving primary medical care from the Henry Ford Health System (HFHS). Cases (n=144) diagnosed by staff neurologists were selected from the HFHS databases. Controls (n=464) were frequency matched on sex, race, and age. Controls who reported any of eight symptoms of potentially undiagnosed PD were excluded. Participants were administered a risk factor questionnaire by trained interviewers face to face. The questionnaire focused on occupational history but also included questions on pesticide use

and farming. Logistic regression was used for the analysis. Exposure to herbicides and insecticides at work was significantly greater for PD patients than for controls (i.e., for occupational herbicide exposure, aOR=4.10, CI= 1.57-12.24, p=0.012; for insecticide exposure aOR=3.55, CI= 1.75-7.18, p<0.01); but there was no association between PD and exposure to fungicides (aOR=1.60, CI=0.47-5.45, p=0.453). The association with PD was greater in those with ≥ 10 years of occupational exposure to insecticides (aOR=5.81, CI= 1.99-16.97) compared to those with < 10 years of exposure (aOR=2.39, CI= 0.89-6.40). Farming was also significantly associated with PD (OR=2.79; CI= 1.03-7.55). Farming as a variable was found to be independent from the pesticides exposure variable: the OR for farming was relatively unchanged after adjusting for any of the three pesticide classes. Furthermore, the association between occupational exposure to herbicides and the risk for PD increased dramatically for those with 10 or more years of well water exposure (increase from OR= 4.10 to OR=13.98, CI=1.46-133.53). There was, however, no statistically significant association between the risk for PD and exposure to residential spraying of insecticides (OR=1.02, CI=0.62-1.65, p=0.951; aOR=1.03, CI=0.63-1.70, p=0.92) nor while gardening as a hobby (for exposure to herbicides, insecticides, or fungicide aOR=1.39, CI=0.84-2.28, p=0.200; aOR=0.90, CI=0.58-1.38, p=0.619; and aOR=0.96, CI=0.55-1.66, p=0.879, respectively), or as a resident or worker on a farm (for exposure to herbicides, insecticides, and fungicides aOR=1.64, CI=0.70-3.82, p=0.253; aOR=1.28, CI=0.69-2.40, p=0.435; aOR=0.96, CI=0.29-3.12, p=0.944; respectively). In summary, the results of this study show a significant association of occupational exposure to herbicides and insecticides with PD and no association between the risk for PD and residential spraying of insecticides or gardening or being a resident or worker on a farm.

Priyadarshi *et al.* (2000) searched a medical abstract database for articles about exposure to or use of pesticide or both (i.e., pesticide use and exposure) and PD as of August 1999 to conduct a meta-analysis. Articles were excluded for the following reasons: 1) language was other than English, 2) pesticide not included as a risk factor, 3) duplication of the studies with same cohort, 4) insufficient data for determining an estimate of relative risk or a confidence interval, 5) disease studied was not specifically designated as PD. A total of 19 case-control studies from various countries were examined. The OR for the studies reporting positive association between PD and exposure to pesticides ranged from 1.1 to 7 and included a total of 2110 PD cases; eight of these studies reported estimated ORs that were statistically significant. Significant heterogeneity was detected among the studies (p<0.01). The random-effect model including all studies yielded a combined OR of 1.94 (CI=1.49 – 2.53). For those studies conducted only in the US, the OR was 2.15 (CI =1.14-4.05).

Ritz *et al.* (2000) conducted an ecological study using death certificate data in California. There were 7516 deaths with an underlying cause of PD for the study time period (1984–1994) and 15,222 deaths with PD mentioned as a contributing cause of death for the same time period (except for 1994, when contributing cause data were not available). Exposure to pesticides was categorized by county based on pesticide use reports, and then agricultural census data were employed to create measures of land per county treated with pesticides. Prevalence odds ratios (PORs) were used to estimate the risk of pesticide exposure for PD mortality compared to death from (underlying) ischemic heart disease (n=498,461). Logistic regression models were used to control for the effect of age, sex, race, birthplace, year of death, and education. The POR for residents in low, moderate or high restricted pesticide-use counties compared to long term-residents of no agricultural pesticide-use counties were 1.52 (CI=1.35 – 1.72), 1.49 (CI=1.31–1.69) and 1.49 (CI=1.30–1.71), respectively. While an elevated risk was observed, a dose-response was not.

Priyadarshi *et al.* (2001) conducted a meta-analysis of case-control studies examining the associations of rural living, well water drinking, farming, and exposure to pesticides with PD. For the pesticide studies the number of cases ranged from 38 to 224 and the number of controls ranged from 38 to 464. The number of females and males were similar in both cases and controls in most studies. Of the 14 studies that examined the association of exposure to pesticides with PD, one study show a negative association, two studies showed no association and the remaining 11 studies reported a positive association. The random-effect model including all studies yielded a combined OR of 1.85 (CI=1.31 – 2.60). For studies conducted in the United States, the combined OR was 2.16 (CI=1.95 –2.39). The overall conclusion from this study was the risk of PD increased with longer exposure duration to pesticides.

Zoron *et al.* (2002) carried out a case-control study to investigate the association of familial and environmental risk factors with PD. Neurologist-confirmed cases of PD (n=136) were age- and sex-matched, with 272 controls affected by neurological diseases unrelated with PD. The risk of developing idiopathic PD was associated with the following factors: positive family history of PD, positive family history of essential tremor (ET), age of mother at subject's birth, rural birth, rural living, well water use, farming as an occupation, exposure to pesticides, head trauma, exposure to general anesthesia and to ionizing radiations, food restriction, concentration camp imprisonment and smoking status. Of 136 cases, 74 were women. In the conditional multiple logistic regression analysis, the following factors increased the risk for PD: positive family history of PD (OR= 41.7, CI=12.2 -142.5, p<0.0001), positive family history of ET (OR=10.8, CI=2.6-43.7), age of mother at subject's birth (OR=2.6, CI=1.4-3.7,

$p < 0.0013$), farming as an occupation (OR= 7.7, CI=1.4-44.1, $p=0.0212$) and well water use (OR=2.0, CI=1.1-3.6, $p=0.0308$). Smoking showed an inverse relationship with PD (OR= 0.7, CI=0.4-1.1, $p < 0.06$). After adjusting for smoking, exposure to pesticides (aOR=1.6, CI=1.0-2.4, $p=0.035$) and rural living (aOR =1.5, CI=1.0-2.4, $p=0.044$) were no longer significant. The mean length of exposures to pesticides was significantly different in cases and control subjects (4.1 years, SD 10.9 and 2 years, SD6.4, respectively; $p < 0.05$). The mean length of exposure to well water use was significantly longer in cases (5.7 years, SD 13.2) when compared to control subjects (2 years, SD6; $p < 0.01$).

Baldi *et al.* (2003b) conducted a prospective cohort study of 1122 men and 1670 women. Occupational and environmental exposure to pesticides and PD or Alzheimer's diagnosis was assessed by questionnaire. Nineteen job titles for 320 subjects were estimated to involve non-null pesticide exposure. In men, the risk of PD was associated with pesticide exposure (aRR=5.63, CI=1.47-21.58); no association was found for women.

Baldi *et al.* (2003a) conducted a case-control study of residents in two regions in Southwestern France. Cases were 70 years old, over and lived in the study regions at the time of interview, and were recruited through area hospitals. Cases were matched 1:3 with controls for age and sex. Controls were enrolled in the PAQUID study – a study on pathological cerebral aging and loss of independence in the elderly. PD status was excluded for controls based on a symptom questionnaire. The agricultural study region largely included vineyards, where fungicides were used heavily, as compared to insecticides or herbicides. Occupational exposure was assessed through the gathering of occupational histories and having engineers to construct a job exposure matrix, rather than relying on subjects' recall. For occupational exposure, there was an increased risk for PD, when taking into account age, sex, educational level and smoking (aOR=2.2, CI=1.1-4.3). Demonstration of a dose-response relationship failed when exposure was calculated by quartiles. With exposure assessed from lowest to highest, the first two exposure quartiles were not significant while the 3rd was statistically significant (aOR=6.6, CI=1.7-25.0); however, the highest exposure quartile had aOR=0.7 (CI= 0.1 – 3.1) and was not significant.

Baldereschi *et al.* (2003) investigated the association of major lifestyle-related risk factors with the prevalent cases of PD identified by the Italian Longitudinal Study on Aging. 5632 individuals were randomly selected from the population of eight centers and screened for parkinsonism using both a questionnaire and a neurological examination; 113 prevalent PD cases were identified. PD was defined among those affected by parkinsonism by exclusion of all other possible causes. Age, male sex, and having a pesticide-use license were significantly related to PD. Heavy smoking was inversely related with PD. Age (OR=1.1, CI= 1.06-1.15) and having a pesticide-use license (aOR=3.68, CI= 1.57-8.64)

retained their significant correlation with PD in the multivariate analysis after adjustment for all the variables under investigation (i.e., age, sex, years of schooling, smoking status (never, 0.01–19.99 pack years, ≥ 20 pack years), and having a pesticide-use license). In multivariate analyses for men and women separately, having a pesticide use license was positively associated with PD only in men (aOR=4.41, CI=1.84-10.56 for men). In this study, farming for at least 10 years was not associated with the risk for PD (aOR=1.2, CI=0.72-1.97).

Gorell *et al.* (2003) sought to determine the relative contribution of potential PD risk factors to the development of PD. Subjects from a health system in tri-county Detroit metropolitan area were recruited. Lifetime cumulative exposure was calculated for each pesticide group. Variables included occupational exposure to selected metals, family history of PD, occupational exposure to pesticides, occupational exposure to farming, and smoking. Cases (n=144) and 464 controls, selected between 1988 to 1992, were matched on sex, race, and age. An interviewer-administered risk factor questionnaire provided a history of all jobs held for 6 months or longer. An industrial hygienist reviewed each job history and assessed the possibility of exposure to different metals. Long-term (>20 years) exposure to manganese had the highest adjusted odds ratio (aOR=10.6, CI=1.1-106.0, $p=0.044$), while occupational exposure to herbicides aOR=4.1 (CI=1.37-12.23) and occupational exposure to insecticides aOR=3.55 (CI= 1.75-7.18, $p=0.001$) were also associated with PD. For farming in general as an occupation aOR=2.79, (CI=1.03-7.54, $p=0.043$). The final multivariate model after stepwise logistic regression had a population attributable risk (PAR) of 54.1% and included the following variables: smoking less than or equal to 30 pack years, PD in first- and second-degree relatives, occupational insecticide exposures, occupational lead and copper exposure (both >20 years). These results suggest that PD is a multi-factorial condition, with various environmental and genetic risk factors playing a role in its development.

Yesavage *et al.* (2004) assessed whether pharmacy database information from the US Department of Veterans Affairs (VA) medical centers could be used to screen for areas of higher PD prevalence among patients exposed to pesticides. Pharmacy datasets (1997-2001) were used to compare the use of anti-parkinsonian medications and two VA medical centers in California (in Palo Alto near the ocean and in Fresno downwind from extensively farmed parts of the Central Valley). These areas were seen as a proxy for pesticide exposure (low=Palo Alto, higher=Fresno). Patients at Fresno exhibited higher ORs for the use of PD medications (carbidopa/levodopa prescriptions) than patients at Palo Alto; OR (Fresno/Palo Alto) from 1.5 (CI=1.1-1.9) in 1997 to 1.8 (CI=1.5-2.2) in 2000.

Park *et al.* (2005) utilized death certificate data from 22 states from 1982 to 1998 through the National Occupational Mortality Surveillance System to develop a case-control study. Cases were defined as having one of several neurodegenerative diseases, including PD. Controls were all decedents with no mention of neurologic disease, degenerative or otherwise and excluding certain exposures (e.g. solvents). Priority occupations associated with neurodegenerative disease (n=87) were identified from a previous study with statistically significant elevated proportionate mortality ratios (PMRs). Of 2,614,346 deaths identified from 1992 to 1998 among participating states, 33,678 were due to PD. The following occupational groups had a significant association with PD: biological scientists [mortality odds ratio (MOR)=2.04, CI= 1.39-2.92]; post-secondary teachers (MOR=1.61, CI=1.39-1.85); teachers (primary/secondary) (MOR= 1.30, CI= 1.18-1.43; clergy (MOR= 1.79, CI= 1.58-2.02); other religious workers (MOR= 1.70, CI= 1.27-2.21. PD risk was significantly elevated across all three farming specifications in all age groups, but the strongest association was for non-horticultural farmers (MOR=1.16, CI=1.11-1.22), with much higher risk below age 65 (MOR=2.23, CI=1.47-3.26). Of all of the occupations, there was the strongest significant relationship with PD among farmers under age 65. This was either due to early onset of PD attributable to farming exposures or the excess cases that might not be discernible above the greatly increased background cases at older ages. MORs were adjusted for age, race, sex, region, and SES. In other occupations where pesticide usage might be expected (e.g., farm workers, horticultural specialty workers), significant elevations were not observed although horticultural specialty workers did show a non-significant excess of PD (MOR=1.65, CI=0.92–2.71). This analysis based on death certificate data was limited by lack of work history and potential misclassification of cause of death. Thus, in many respects, morbidity data are preferred to analyze the association between PD and many exposures.

Galanaud *et al.* (2006) conducted a case-control study where 292 cases were chosen from those enrolled in a French insurance system for agricultural workers who submitted an application for PD health care coverage among ages 18-75. Controls were chosen from the same insurance group, but without signs of PD and matched with cases by age and geography. Exposure was assessed by interview with a questionnaire. Data were gathered on smoking status, pesticide exposure, and alcohol consumption. There was a decreased risk of PD with smoking, (OR=0.6, CI=0.4–0.9). PD risk among those who had never smoked but did have professional pesticide use was 2.0 (CI=1.1–3.6); the risk was not statistically significant for those who ever smoked (OR= 0.9, CI=0.5--1.8).

Wastensson *et al.* (2006) analyzed cases of PD in a group of paper mill workers in Sweden exposed to the fungicide diphenyl. Subjects were identified from company files and trade union cards

where job titles and time of employment were noted. Initially, 506 exposed workers who had worked in production of diphenyl-impregnated paper anytime from 1954 to 1970 were identified. However, only 284 were still living in Sweden in August 2002. The resulting cohort of workers still alive and under the age of 80 years (255 workers) included only those with job titles or other designations indicating that they had worked in the large hall where the production of diphenyl-impregnated paper took place. Given that the number of expected prevalent cases in the exposed group was estimated to be 0.9 (versus the five cases found), this resulted in a relative risk of 5.6 (CI=1.8-13, p=0.002). The authors also looked at PD risk among the deceased workers (n=222). Nine cases of PD were found in the exposed group compared to the 4.3 cases of PD expected from data on the lifetime risk of developing PD in the general population (RR=2.1, CI=0.96-4.0).

Frigerio *et al.* (2006) recruited 149 PD cases (90 men) and 129 controls from the Rochester Epidemiology Project from 1975 through 1996. Each case was individually matched by age (± 1 year) and sex to a general population control who resided in Olmsted County, MN, and who was free of PD, other parkinsonism, or tremor of any type in the index year (year of onset of PD in the matched case). All subjects who farmed for five or more years were asked additional detailed questions about the main type of crops, the use of pesticides (including herbicides, insecticides, and other pesticides), and the names of the specific products used. In this study, the use of pesticides by male farmers was more common in cases than in controls; however, because of limited power, the difference was not statistically significant. Exposure to pesticides related or unrelated to farming was associated with PD in men (OR=2.4, CI=1.1-5.4; p=0.04). This association remained statistically significant after adjustment for education divided in four quartiles (aOR=2.8, CI=1.2-6.5, p=0.02) or smoking ever vs. never (aOR=2.5, CI=1.1-5.7, p=0.03).

Ascherio *et al.* (2006) prospectively examined whether individuals exposed to pesticides are at higher risk for PD than those not exposed. The study population was comprised of participants from the Cancer Prevention Study II (CPS II) Nutrition Cohort, a longitudinal investigation of US men and women initiated in 1992 by the American Cancer Society. In 1982, as part of the original CPS II mortality study, participants completed a four-page survey that included questions on occupation and exposure to broad chemical categories, including pesticides/herbicides. If exposed, participants were asked to report the duration of exposure in years. Follow-up surveys were conducted in 1997, 1999, and 2001. The 143,325 individuals who returned the 2001 survey and did not have a diagnosis or symptoms of PD at baseline (1992) were included in the analyses. Individuals exposed to pesticides had a 70% higher incidence of PD

than those not exposed (aRR=1.7, CI=1.2-2.3; p=0.002). The relative risk for pesticide exposure was similar in farmers and non-farmers.

Kamel *et al.* (2006) used data obtained from licensed private pesticide applicators and spouses participating in the Agricultural Health Study to evaluate the relation of self-reported PD to pesticide exposure. Individuals applying for certification to use restricted-use pesticides in Iowa or North Carolina were enrolled in this cohort study. Cohort members, who were enrolled in 1993-1997, provided detailed information on lifetime pesticide use. At follow-up in 1999-2003, 68% of the cohort was interviewed. Cases were defined as participants who reported physician-diagnosed PD at enrollment (prevalent cases, n=83) or follow-up (incident cases, n=78). Cases were compared with cohort members who did not report PD (n=79,557 at enrollment and n=55,931 at follow-up). Incident PD was associated with cumulative days of pesticide use at enrollment (highest quartile vs. lowest, OR=2.3, CI=1.2-4.5; p-trend = 0.009). Although positive, there was not a statistically significant association with personally applying pesticides more than half the time (OR=1.9, CI=0.7-4.7). Associations between the risk for PD and exposure to specific pesticides are discussed in other relevant sections. Prevalent PD was not associated with overall pesticide use.

Dick *et al.* (2007a) conducted a case-control study of 959 prevalent cases of parkinsonism (767 with PD) and 1989 controls in Scotland, Italy, Sweden, Romania and Malta. Cases with drug-induced or vascular parkinsonism or dementia were excluded. Subjects completed an interviewer-administered questionnaire about lifetime occupational and hobby exposure to solvents, pesticides, iron, copper and manganese. Lifetime and average annual exposures were estimated blind to disease status using a job-exposure matrix modified by subjective exposure modeling. Multiple logistic regression analysis was conducted with adjustments for age, sex, country, tobacco use, ever knocked unconscious and family history of PD. Analyses showed a positive but non-significant association between PD and any exposure to pesticides (aOR=1.25, CI=0.97 to 1.61) and low pesticide exposure vs. no exposure (aOR=1.09, CI=0.77-1.55). However, there was a statistically significant positive association between PD and high pesticide exposure vs. no exposure (aOR=1.39, CI=1.02 - 1.89).

Hancock *et al.* (2008) observed that the frequency of pesticide exposure (>10 days per year), exposure duration (>25 years), and cumulative days of exposure (>215 days) were statistically significantly associated with a 2-fold increased risk for PD in a dose-response manner (p<0.013). The authors conducted a case-control study using 319 cases and 296 controls who were relatives of the cases. Subjects were recruited from the Morris K. Udall PD Research Center of Excellence at Duke University Medical Center. Cases were confirmed by an in-person examination. Pesticide exposure was

assessed using a telephone questionnaire. Questions included: "Have you ever applied pesticides to kill weeds, insects, or fungus at work, in your home, in your garden, or on your lawn?" Individuals provided only a "yes/no" answer for this question, so separation of residential applications from occupational applications for analysis was not possible. If the answer was "yes," individuals were asked to list the name of any pesticides they remembered using. For each pesticide, individuals were asked the number of days it was used per year, whether it was currently being used, the years application started and stopped (if applicable), and whether protective gear, such as a mask, rubber gloves, or rubber boots, was used during application. The study examined the associations of pesticide application, well-water consumption, and farming residences/occupations with PD while controlling for age at examination, sex, cigarette smoking and caffeine consumption. By recruiting controls that were relatives of the cases, the authors argue that there is less likelihood for spurious association due to unmeasured genetic and environmental factors compared to a population-based sample. Cases were significantly more likely to report pesticide application than their unaffected relatives (OR=1.61, CI=1.13-2.29). Frequency, duration, and cumulative exposure were also significantly associated with PD in a dose-response pattern ($p < 0.013$). The association was modified by family history of PD, as only individuals with no family history of PD had an increased risk of PD associated with pesticide exposure (OR=1.8, CI=1.2-2.7). When examining associations of herbicide application with PD, there was no statistically significant positive association with PD for individuals who reported ever applying herbicides (OR=1.59, CI=1.00-2.54), as compared to individuals who never applied pesticides. However, there was significant positive association with PD for individuals who reported ever applying pesticides other than herbicides (OR=1.61, CI=1.09-2.38), as compared to individuals who never applied pesticides. When examining associations between insecticide application and PD, only individuals who reported ever applying insecticides were significantly more likely to develop PD (OR=1.83, CI=1.20-2.81). There was no statistically significant increased risk of PD among individuals who reported ever applying pesticides other than insecticides. In addition, specific insecticide classes (organochlorines and organophosphates) were found to be associated with PD and these are cited in the corresponding sections, below. Well-water consumption and living on a farm were not associated with PD. Because the association between pesticide exposure and PD only existed for those with a no family history of the disease, the authors recommended that pesticides should be considered as an effect modifier in future candidate gene studies.

Elbaz *et al.* (2009) conducted a case-control study of 224 cases on 557 controls. Cases were those with PD selected from the Mutualité Sociale Agricole (health insurance company for farmers

whether retired or active). Controls were from the same health insurance company but without PD. Pesticide exposure was evaluated in two phases including an expert evaluation. Analyses were performed by broad category (insecticide, herbicide, fungicide) and 29 pesticide families (this part of the analysis was restricted to men). The authors found an association between professional exposure to pesticides overall and PD (OR=1.8, CI=1.1-3.1) as well as a dose-response relationship between the length of exposure (p-trend=0.01 for ≤ 38 and > 38 years of exposure vs. no exposure) and PD). Associations between pesticides and PD were generally stronger in men with older onset PD than younger onset PD. Thus, genetics seemed to play a larger role in younger cases, whereas environmental exposures played a larger role in older cases. Professional pesticide use was associated with PD in a population characterized by a high prevalence of exposure. For gardening with non-professional pesticide use, there was a positive but not statistically significant risk of PD (OR= 1.4, CI= 0.9-2.3 and p=0.18).

Hristina *et al.* (2010) conducted a case control study in Belgrade from 2001-2005 to examine the association between residential and occupational exposure to pesticides and PD development. There were a total of 110 PD cases with 220 hospital controls which were matched by age, sex, and geography. PD cases, significantly more frequently than controls, spent some time during their life in rural settings ($p < 0.001$). The authors examined a number of variables for potential association with PD including occupation, type of agricultural work, and type of nonagricultural work, exposure to any pesticides, insecticides, fungicides, well water drinking, and spring water drinking, and occupational exposure to chemicals and metals. The final logistic regression analysis, in which an adjustment was made for smoking, included gardening (ever), exposure to insecticides (residential plus occupational), occupational exposure to dyes, occupational exposure to naphtha and its derivatives, well water drinking (ever), spring water drinking (ever) and service sector worker. All of these variables had a positive and statistically significant association with PD, with the exception of service worker. For exposure to insecticides through occupation and residence the aOR=3.22 (CI=1.32–7.87). The most frequently used insecticide was imidaklopid, followed by malathion, cipermetrin, and dimetoat. However, the relationship to individual insecticides was not examined nor was the quantity of exposure. For gardening at any time during life the OR was 5.51 (CI=3.04–10.01). Of note is that PD was not associated with agricultural occupation ($p=0.424$) and thus was not included in the final model. It was postulated that well water could serve as a vehicle for various chemicals' exposure, which can initiate the development of PD.

Sanyal *et al.* (2010) investigated the possible impact of environmental risk factors on idiopathic PD development in a case-control study performed in Eastern India between January 1st, 2006 and December 10th, 2009. PD patients (140 men, 35 women) and 350 non-PD age-sex matched controls were included in the study. Subjects were given a structured neurological examination and administered a questionnaire, which elicited detailed information on demographic data, pesticides, herbicides, family history, occupation, dietary and smoking habits. The multivariate analysis revealed that pesticide exposure was positively associated with PD development (OR=17.12, CI=4.97-58.84). Other factors associated positively with the risk of PD were family history of PD in first and second degree relative (OR=21.4, CI=6.36-70.12), exposure to chemicals other than pesticides and herbicides (OR=7.54, CI=2.72-20.89), rural living (OR=4.05, CI=2.53-6.49), and previous history of depression (OR=1.98, CI=1.18-3.29), whereas, smoking appeared to be a protective factor (OR=0.45, CI=0.26-0.79). Well water drinking for at least five years, though a significant risk factor on univariate analysis (OR=4.5, CI=2.1-9.9), could not be proved significant in multivariate analysis. Head trauma, vegetarian dietary habit, occupation involving physical exertion and exposure to domestic pets were not as significant risk factors for PD.

Moisan *et al.* (2011) conducted a cross-sectional study to examine the relationship between PD prevalence and farming type as a proxy for pesticide exposure. Examples of farming type included cattle ranching, general field cropping, and mixed cropping, with pesticide exposure varying greatly by farming type. Cases were identified using data from a French organization that reimburses current farmers and retired farmers for health insurance. PD prevalence was then estimated in five French districts. The authors split the exposures (farming type) into 5 quintiles, by density. For example, an area with the lowest density of cattle farms would be placed in the 1st quintile for that exposure category, whereas an area with the highest density of cattle farms would be placed in the 5th quintile. After adjustment for age, sex, district, and income, the risk of PD increased with the density of farms specialized in fruits and permanent crops (FSFPC) (OR_{5th vs. 1st quintile}=1.21, CI =1.02-1.43, p-trend=0.008). The association between PD prevalence and FSFPC was similar across all districts. The authors noted that 1) there was a high use of insecticides and herbicides in FSFPC and thus FSFPC pesticide applicators were potentially more exposed to pesticides than persons applying pesticides to other crops and 2) while cases were not confirmed by a neurologist, it was unlikely that diagnostic misclassification depended on farming type. Additionally, through a validation study, the authors noted that the sensitivity/specificity of the case definition did not depend on FSFPC density (p= 0.980).

Van der Mark *et al.* (2012) conducted a meta-analysis of 46 studies on PD and pesticide exposure, including 39 case-control, 4 cohort, and 3 cross-sectional studies. Exposure to pesticides was classified as ever vs. never exposed and by exposure assignment based on job title. The authors reported a statistically significant association between pesticide exposure and PD risk (summary risk ratio or sRR)=1.62, CI=1.40-1.88) and between PD and job title–based exposure assignment (sRR=2.5, CI= 1.5-4.1). Self-reported pesticide exposure was also associated with PD development (sRR=1.5, CI=1.3- 1.8). The authors indicate that conclusions are limited by subjective exposure assessment and recommended that future studies focus on more objective methods of pesticide exposure assessment.

Van Maele-Fabry *et al.*(2012)conducted a meta-analysis of 12 cohort studies of workers occupationally exposed to pesticides with an outcome that included parkinsonian disorders or associated diseases (Parkinson’s disease, parkinsonism). Only those results for PD are reported here. The method of exposure assessment varied among the 12 studies and five of the studies only included men. The small number of studies prevented conclusions about specific chemical class of pesticides likely to be related to PD. Studies restricted to banana, sugarcane and pineapple plantations showed an increased mRR of 2.05 (CI=1.23 – 3.42) for PD, as did studies where the PD diagnosis was confirmed by a neurologist mRR 2.56 (CI=1.46 – 4.48). This meta-analysis provided some support for the hypothesis that pesticide exposure increases the risk of PD.

Noyce *et al.* (2012) reported a statistically significant positive association between exposure to pesticides (or herbicides or insecticides) and PD using a systematic review and meta-analysis of the risk factors for PD. For 36 case-control studies combined, there was a positive association between pesticide exposure and the later diagnosis of PD (OR=1.77, CI=1.48 – 2.12); for two combined cohort studies, the relative risk (RR) was 1.78 (CI=1.30-2.42), and for all studies combined, the RR was 1.78 (CI=1.50-2.10). After the authors conducted the Egger test, which suggested evidence of publication bias ($p<0.001$), the trim and fill method was used to account for this bias. This resulted in diminished summary estimates for pesticide exposure (RR declined from 1.78 to 1.53; CI= 1.29-1.80).

Savica and co-authors (2013) investigated specifically how PD risk factors, including pesticide use, affected the development of PD in men and women separately. This study of 196 case-control pairs (392 individuals) revealed that in men but not in women, lack of coffee consumption (never vs. ever), history of head trauma, and pesticide use were independent but relatively rare risk factors for PD and subjects who had at least one of these three risk factors (composite exposure variable) had over five-times greater risk of PD development (OR=5.28, CI=2.67-10.43). However, the magnitude of the effect due to exposure to pesticides alone or specific type of pesticides used were not provided.

Pezzoli and Cereda (2013), who investigated the risk of PD associated with exposure to pesticides and solvents using meta-analyses of data from cohort and case-control studies, found a marginal and non-statistically significant association (RR=1.26, p=0.075) between exposure to pesticides and PD development, for five pooled cohort studies. For three cohort studies investigating the role of employment in agricultural jobs, the pooled estimate for risk of PD was RR=1.33 (p<0.001). In primary analyses including all case-control studies, the authors also found that PD was associated with exposure to any type of pesticides, herbicides, and insecticides. For pesticides, the OR was 1.76 (CI=1.56—2.04); for herbicides, the OR was 1.33 (CI=1.08-1.65); for insecticides, the OR was 1.53 (CI=1.12-2.08). However, no association was observed with fungicides, rodenticides, organochlorines, and organophosphates. The authors concluded that the literature supports the hypothesis that exposure to pesticides is a risk factor for PD.

One review article (Kieburtz and Wunderle, 2013) evaluated the level of evidence for the association between pesticide exposure and the development of PD among 4 studies by choosing one of two categories: 1) sufficient evidence of an association 2) limited suggestive evidence. For the sufficient evidence category, the articles needed to demonstrate a consistent positive relationship and that bias or chance can be ruled out with reasonable confidence. For the limited suggestive evidence category, the studies suggested a positive relationship, but bias or chance could not be ruled out with confidence. The authors concluded that for these 4 studies, there is limited suggestive evidence of an association between pesticide exposure and PD.

Lack of association

The studies which could find no association between general pesticide exposure and risk of PD are summarized here. In some cases, however, the studies were mixed with both positive and no associations.

Ho *et al.* (1989); surveyed eight old age homes in two areas of Hong Kong to find PD cases and controls. Initial pool was 561 subjects. Of these, 35 cases were identified through examination by trained doctor and independently checked by 2 other examiners. Controls (n=105) matched by age and sex were those that were free of PD and dementia. A person-to-person interview with a structured questionnaire was used to collect information on social characteristics, medical history, health practices, and environmental factors such as rural living, farming practices, exposure to pesticides and consumption of raw vegetables. Informants were used for those PD patients with dementia. Other variables identified included smoking, drinking alcohol, and tea drinking. Risk of PD among those living in rural areas over 40 years was 4.9 (CI=1.4-18.2). The risk was not statistically significant for 21-40 years

of residency (OR=2.1). Risk of PD among those farming over 20 years was OR=5.2 (CI=1.6-17.7). There was a positive but not statistically significant association between previous use of pesticides and herbicides and PD development (OR= 3.6, CI=1.0-12.9). Consumption of raw vegetables increased the risk for PD (OR=10.8, CI=2.4-40.0). There was a protective effect of drinking black or other tea seen, but this effect was not statistically significant. Smoking was also protective effect of (OR=0.6, CI=0.2-1.3), but it was not statistically significant. Patients with PD showed a much higher frequency of eating raw vegetables. A case control study carried out by Koller *et al.* (1990) included 150 PD cases, which were randomly selected from the Movement Disorder Clinic at the University of Kansas Medical Center. PD was confirmed by the presence of two or more cardinal signs of the disease and responsiveness to levodopa therapy. Controls (n=150) matched by age and sex were randomly recruited from the same clinic as the cases. All control subjects were examined and those with any parkinsonian signs were excluded. Exposure information was collected through a questionnaire administered by a trained interviewer. Exposure data collected included rural versus urban living, number of years spent farming, number of years drinking well water, and exposure to herbicides or pesticides. There was a statistically significant difference in the number of years that PD patients spent living in a rural environments compared to controls (OR=1.9, p=0.01). There was also an association between well water use and PD (OR=1.7, p=0.03), though that was dependent on rural living. There was no association between PD and pesticide use (OR=1.1, p=0.8).

A pilot case- control study by Wechler *et al* (1991) involved subjects (cases and controls) recruited from a neurology clinic in Seattle, (some cases were recruited from area Parkinson's support groups). The study included 34 cases of PD (no discussion of how PD was classified or verified) and 25 controls (average age 68.4 and 58.9 years, respectively). Exposure to various environmental factors and occupations was assessed through a self-administered questionnaire mailed to participants. The return rate of the questionnaires was 49% (59 of 121 distributed). A non-significant but elevated odds ratio was reported for males working in farming occupations (OR =3.1, CI=0.3-35.2). Analysis of occupational exposure to pesticides was limited to males, because too few females worked in the occupations of interest. However, analysis of pesticide exposure at home included both males and females. More home-use of 3 pesticides (Kleenup grass & weed killer, Orthotriox, and Pestkill) was observed among PD cases than controls, but the trend was not significant for any of those pesticides. No measures of association were reported for pesticide use. Although no clear trends were observed for pesticide exposure, the authors recommended that future studies should examine the association between PD and exposure to specific pesticides.

Another case-control study was carried out with 161 cases and 149 matched controls (Stern *et al.*, 1991). There were 2 groups of cases: young onset (<40 year olds; 46%) and old onset (>60 year olds; 53.7%). There were 60.4% males and 39.6% females; 98.0% Whites and 4.0% Blacks). The risk factors evaluated for their association with PD development included well water drinking, rural living, insecticide and herbicide exposure, head injury, smoking, education. Exposure was assessed by using a structured questionnaire. There was no statistically significant association between PD and exposure to insecticides (OR=0.5, CI=0.2-1.1) and herbicides (OR=0.9, CI=0.6-1.5). There was a non-statistically significant difference in the risk for PD between young/old onset groups given insecticide or herbicide exposure. Self-reporting of exposures in structured questionnaires may have introduced recall bias.

A case-control study of 19 families having two or more siblings with PD was conducted in Kansas to investigate possible risk factors for PD (Wong *et al.*, 1991). Nineteen sibling pairs examined by a neurologist for PD were analyzed. PD was defined as the presence of at least two of the cardinal signs of parkinsonism (i.e., tremor, bradykinesia, rigidity, and postural instability). Patients with atypical features and dementia were excluded. Two control groups were selected for comparison; 38 individuals who were age- and sex-matched to the PD patients were randomly selected from the general neurology and medicine clinics. Demographic data included lifetime histories of places of residence; sources of drinking water; occupations, such as farming; and exposure to herbicides and pesticides. A comparison of parkinsonian siblings with siblings with essential tremor revealed no differences in any risk factors for the years of shared environment. Rural living and drinking well water ($p=0.07$), but not farming and herbicide/pesticide exposure (for herbicide/pesticide use OR=1.0, $p=1.00$), were significantly increased in 38 parkinsonians compared with 38 normal control subjects.

Smargiassi *et al.* (1998) carried out a questionnaire-based case-control study to investigate the possible association between exposure to environmental factors, including occupational exposure to pesticides and herbicides and PD development. Cases included 86 patients with neurologist-confirmed idiopathic PD and 86 controls similar in sex and age in Northern Italy, recruited in outpatient specialist centers of the same University Hospital (glaucoma, psoriasis vulgaris, essential arterial hypertension and renal diseases). Exposure was defined as occupational or residential contact with a given factor for at least 10 consecutive years prior to the onset of PD. Subjects who never smoked were excluded. In this study, occupational exposure to pesticides and herbicides was not associated with the development of PD (OR=1.15, CI=0.56-2.36). However, the following risk factors were identified for PD: well water use (OR= 2.78, CI= 1.46-5.28) and occupational exposure to industrial chemicals (OR= 2.13, CI= 1.16-3.91), which included pesticides and herbicides. Among industrial chemicals, only organic solvents were

identified as significant risk factors for PD (OR= 2.78, CI= 1.23-6.26). Smoking habit was negatively associated with PD (OR= 0.41, CI= 0.22-0.75), confirming the "protective" role of tobacco smoking suggested by many studies.

Werneck and Alvarenga (1999) conducted a case-control study in the city of Rio de Janeiro, Brazil to investigate the relationship between exposure to potential risk factors (herbicides and pesticides [unspecified], occupational exposure to chemicals, ingestion of drugs with secondary PD effects, rural life, water well source, family history, cranial trauma, and cigarette smoking) and development of PD. 92 subjects (41 men & 51 women, average age 70.55 yrs) diagnosed with PD between Jan 30, 1996-Feb 1, 1997 were randomly selected at the Neurology Dept of IASERJ Central Hospital; 110 controls (47 men & 63 women, average age 68.38 yrs) selected in the same hospital and matched with by sex and age (+/- 2 yrs); all controls underwent examination to exclude those showing any signs of parkinsonism or dementia; a questionnaire was used to investigate potential risk factors. Contact with herbicides and insecticides ("Have you regularly inhaled or handled herbicides and insecticides? For how long?") for a minimum period of 15 years was required for enrollment in the study. No significant association was found between PD and general herbicide/pesticide exposure (OR=2.49, CI=0.53-13.14). Six cases (6.36%) and 3 controls (2.72%) had contact with herbicides and pesticides; four individuals used them during rural residency (all had PD) and two PD cases were positively associated with family history (OR=14.5, CI=2.98-91.38), the use of drugs with secondary PD action (OR=11.01, CI=3.41-39.41), and with exposure to chemical agents (OR=5.87, CI=1.48-27.23). PD was inversely associated with cigarette smoking (OR=0.39, CI=0.16-0.95). Univariate analysis showed cigarette smoking to be a protective factor for PD development.

Kuopio *et al.* (1999) studied the environmental risk factors of PD in Finland, particularly the factors related to rural environment, in a study in 1992. The population of 196,864 people, including urban and rural areas comprised this community-based case-control study. The case subjects (n= 123; 63 males and 60 females; 96 urban and 27 rural) were matched with control subjects (n=246; 126 males and 120 females; 192 urban and 54 rural). Analyses were carried out by conditional logistic regression model. The use of DDT had been the same in both groups (OR=1.04, CI=0.68-1.60, p=0.855). The use of pesticides (regular use OR=0.65, CI=0.33-1.29, p=0.221; occasional use OR=1.23, CI=0.74-2.04, p=0.431; for both OR=1.02, CI=0.63-1.65, p=0.935), herbicides (regular use OR=0.79, CI=0.38-1.66, p=0.539; occasional use OR=1.71, CI=0.90-3.23, p=0.101; and for both OR=1.40, CI=0.79-2.48, p=0.245) , or mercury-containing pickling solutions (regular use OR=1.37, CI=0.53-3.53, p=0.58; occasional use OR=1.47, CI=0.58-3.70, p=0.420; for both OR=1.58, CI=0.74-3.42, p=0.240), considering regular and

occasional use separately and both regular and occasional use together, was not associated with increased risk of PD. Only three case subjects and five control subjects recalled the use of paraquat-containing products.

Tuschen *et al.* (2000) conducted a prospective, hospital-based cohort study to examine the possible association between agricultural and horticultural work and the subsequent morbidity from PD. Fixed cohorts of 2,273,872 men and women aged 20-59 years on 1 January 1981 and identified in the Central Population Register of Denmark were followed. All first-time hospitalizations with PD as the principal diagnosis during the 13 years until 31 December 1993 were recorded. Standardized hospitalization ratios (SHR) were calculated using all gainfully employed persons as the standard and by multiplying the ratio by 100. Statistically significantly high risks for PD were found for farmers (79 cases, SHR=130, CI=103-163) and for all men in agriculture and horticulture (109 cases, SHR=134, CI=109-162). A consistent, but non-significant pattern of high risks for PD was found among other occupational groups known to be potentially exposed to pesticides (e.g., agriculture or horticulture workers); however, no data on exposure to pesticides per se were collected nor evaluated.

Herishanu *et al.* (2001) PD cases (n=93) and age- and sex-matched controls (n=93) recruited in medical facilities in S. Israel were administered a validated exposure questionnaire. A multivariate logistic regression was applied for data analysis. Ninety-three consecutive unselected urban patients diagnosed between 1989-1995 as suffering from PD and treated at the outpatient PD clinic of Soroka University Medical Center in Beer-Sheva (the only general hospital in the entire region serving approximately 600,000 inhabitants) were compared to 93 age- (\pm two years) and sex-matched controls. The latter were recruited from the outpatient dermatology, neurology and internal medicine clinics of this hospital. Inclusion criteria for PD were progressive disorder and presence of at least two of the cardinal signs of tremor, rigidity, bradykinesia, postural instability, and good response to l-dopa. Additional criteria were absence of significant cognitive impairment and lack of an alternative etiology known to cause secondary parkinsonism. Controls presenting extrapyramidal signs, or evidence of other neurodegenerative diseases, such as Alzheimer's disease or essential tremor, were excluded from this study. All subjects (patients and controls) resided in Beer-Sheva or in other towns of the Negev. The risk of PD due to pesticide exposure was increased but not statistically significant (OR=6.81, CI=0.75-6.44, $p < 0.1$). Multivariate logistic regression analysis showed history of work in construction was the strongest predictor of PD risk, followed by exposure to pesticides. Smoking was found to be a strong protective factor for PD development.

Petrovich *et al.* (2002) conducted a prospective cohort study with 30 years of follow-up on the island of Oahu, HI. Male subjects of Japanese ancestry (n=8006) were enrolled in the Honolulu Heart program which began in 1965. Diagnosis of PD was identified through hospitalization records, death certificates, and review of medical records for all patients with PD from a local neurologist's office. After 1991, the diagnosis of PD was based on a complete reexamination of the entire cohort. Exposure was assessed by occupational exposure histories including questions on type of plantation work and length of time working on plantations. Information on confounding variables including age, smoking pack years, and coffee consumption was also collected. Workers who had over 20 years of work on a plantation were 1.9 times (CI=1.0-3.5) more likely to have PD compared to men with no plantation work. However, self-reported pesticide exposure was not significantly associated with PD. Study data implicate occupational pesticide exposure as a likely factor in PD in those study subjects who had worked on plantations for more than a decade.

Abbott *et al.* (2003) examined data from a 1965 cohort of 8,006 Japanese-American men who were interviewed about environmental, life-style, and physical attributes at selected examinations to study the relationship between these factors and the incidence of PD. All subjects were PD-free at the start of follow-up for clinical PD. Independent effects of each factor on the risk of PD were examined through the use of proportional hazards regression models. Results were not statistically significant ($p=0.101$) for exposure to pesticides and risk of PD (no ORs were reported). Among non-drinkers of coffee, risk of PD was 3-times higher in men who were exposed to pesticides for more than 3 years (63.4/10,000 person-years) as compared to men with no exposure to pesticides (21.4/10,000 person-years, $p=0.044$). Risk of PD in nonsmokers also seemed to increase susceptibility to pesticides for exposures beyond 3 years versus men who were not exposed (27.4 versus 11.8/10,000 person-years, $p=0.053$). The authors reported that their results indicated cigarette smoking and coffee drinking reduced the susceptibility to PD associated with exposure to pesticides.

Exposure to pesticides was not associated with increased risk for PD in a retrospective case-control study conducted by Nuti and co-authors (2004) in Tuscany, Italy. Other environmental factors studied for their potential association with PD development were residency in rural areas and well-water drinking. PD patients (n=190; 106 males and 84 females) were identified as cases for the study. A population of 190 controls (106 males and 84 females) was recruited. There was no significant difference in mean age and sex between cases and controls. However, controls were more likely to be cigarette smokers. The results of this study did not show any statistically significant difference between PD patients and controls for time spent in rural or industrial residence, in well water drinking or in the

exposure to herbicides and pesticides. The ORs for exposure to pesticides and the risk for PD were as follows: OR=0.82 for 1-10 years of exposure duration, OR=1.00 for 11-20 years of exposure duration, OR=0.90 for 21-30 years of exposure, and OR= 1.2 for \geq 30 years of exposure. Although the findings of this study did not show any statistically significant association between the risk for PD development and residency in rural area, well-water drinking, and exposure to pesticides these findings indicated a protective effect of cigarette smoking on PD development.

Firestone *et al.* (2005) Confirmed PD cases (n=250) and 388 age-, sex-, clinic location-, and enrollment period-matched controls were administered a structured interview to assessed exposure to pesticides. PD case patients were identified between 1992 and 2002 at the Group Health Cooperative (GHC) in western Washington State or the University of Washington. To ensure complete ascertainment, subjects were identified using a combination of provider referrals and computerized databases containing diagnostic coding and pharmacy information. Pesticides were grouped into organophosphates, any pesticide, and not exposed and exposure was categorized into low/med/high for analyses. Only lifelong well-water consumption had a statistically significant association with the risk of PD (OR=1.81, CI=1.02-3.21). Odds ratios for occupational exposure were not significant but suggested a gradient that paralleled an expected increase in pesticide exposure by occupation (pesticide worker: OR=2.07, CI=0.67-6.38; crop farmer: OR=1.65, CI=0.84-3.27; animal and crop farmer: OR=1.10; CI=0.60-2.00; and dairy farmer: OR=0.88, CI=0.46-1.70). The authors found no evidence of risk from home-based pesticide exposure. For example, any pesticide exposure resulted in an OR of 0.95 (CI=0.66-1.37). Risk for herbicides was elevated but not statistically significant (OR=1.09, CI= 0.77-1.53).

Brown and co-authors (2006) conducted a literature review of epidemiological studies with pesticide exposure as one of the risk factors for PD development. Of the 38 case-control studies reviewed, all but 10 were already reviewed as original articles in this report. The 9 studies not included, reporting either positive or no association between risk of PD and exposure to pesticides, are summarized here. Fungicide exposure was not found to be a significant risk factor for PD, nor was exposure to rodenticides (Behari *et al.* 2001). Falope *et al.*, (1992) reported an OR of 1.0 (CI=0.33-3.06) for the risk of PD and exposure to pesticides. Chaturvedi *et al.* (1995) examined pesticide exposure and fertilizer exposure and reported an OR of 1.81 (CI=0.92-3.36). Taylor *et al.* (1999) found that pesticide exposure was not a significant risk factor for PD after adjustment for confounding variables (OR=1.02, CI=0.9-1.2). The relationship between exposure duration and PD risk was investigated by Nelson *et al.*, 2000 and a positive association was observed with high doses of pesticides compared with low doses. There was also a positive correlation with duration of exposure to, and high doses of, herbicides and

insecticides. Preux *et al.* (2000) found an OR of 1.34 (CI=0.85-2.1) and exposure to pesticides assessed through question “Have you ever been exposed to pesticides?” Vidal *et al.*, 2002 had an OR of 1.7 (CI=1.1-2.8). Duzcan *et al.* (2003) asked “Have you been exposed to pesticides for more than 20 days during a year for at least 10 years?” and the OR was 1.96 (CI=1.31-6.69) indicating a statistically significant association with PD. Kamel *et al.* (2001) did not find a significant association with paraquat exposure, although risk was elevated: OR= 1.5 (CI=0.7-3.0); for maneb, OR=1.6 (CI=0.7-4.1).

A case-control study examining the association between exposure to pesticides, well water use, and rural exposures and the risk for PD was carried out by Wright and Keller-Byrne (2005). Residents of eastern Missouri and southwestern Illinois were enrolled in the study in 1995. Cases (n=102) were recruited from PD support groups and matched with 133 controls. A self-administered questionnaire was used to assess exposure history. Occupational pesticide use was not associated with PD (aOR=1.2, CI=0.3-4.8) after adjustment for smoking, rural living, familial PD, and other variables. However, exposure to 30 or more years of well water was associated with an increased risk of PD (aOR=8.7, CI=1.5-52). An elevated risk of PD was detected for any well water exposure during the first 40 years compared to no well water exposure (OR=7.3; CI=2.3-22.6) and for any exposure during childhood (first 20 years of life) compared to less than 20 years of exposure during childhood (aOR=8.3; CI=2.5-27.6). Among primary well water users in the first 20 years, the risk of PD was 11 times greater compared to subjects with no childhood exposure to well water (aOR=11.1, CI=2.1-51.8). There was also an exposure-response relationship for well water use; for each five-year increase of exposure before the age of 20, the risk of disease increased by 90% (OR=1.9, CI=1.3-2.8). Multivariate regression analyses adjusted for marital status, smoking, rural living, farm living, familial PD, familial essential tremor, head trauma, orchard employment, residential and occupational pesticide use.

Hofmann *et al.* (2006) investigated mortality among a cohort of workers from a Costa Rican banana plantation. The cohort included 40,959 individuals who worked on the plantation between 1972 and 1979. Their employment records were linked with the Costa Rican Mortality Registry to determine PD diagnosis as the cause of death through 1999. Although exposure to the nematocide, 1,2-dibromo-3-chloropropane (DBCP) was the primary interest for this study, the study could not distinguish between exposure to DBCP and exposure to other agricultural chemicals, including pesticides such as organophosphates, paraquat, and other pesticides. There were a total of 6 reported PD deaths during the follow-up period. Compared to the local population, male banana plantation had an elevated, but non-significant increased risk of mortality from PD (SMR= 2.39, CI=0.88-5.20) that was based on small numbers (n=6). Relative to national mortality, the SMR for PD was lower for the banana plantation

workers, but above the null value (SMR=1.11, CI=0.41-2.41). No deaths from PD were reported for female workers. The lack of precise exposure information and the likely misclassification of cause of death in the Mortality Registry were the major limitations of this retrospective mortality study.

Cho *et al.* (2008) conducted a case-control study with participants recruited from Seoul National University Hospital which was comprised of 235 PD patients, 133 multiple system atrophy (MSA) patients, and 77 normal control subjects, according to the criteria of the United Kingdom Parkinson's Disease Society (UKPDS) brain bank, but without the criterion of a positive family history. Environmental factor data were collected by face-to-face interviews using a structured questionnaire and included detailed information on duration of participation in farming, farming type, tap-water and well-water drinking, number and severity of pesticide intoxications, number and severity of carbon monoxide (CO) intoxications, and smoking history. Data on exposure to pesticides included the annual frequency of pesticide spraying and the duration of farming. The pesticides poisoning data included the number and severity of acute poisoning episodes. The combined variable of (farming years)*(annual frequency of pesticide spraying) resulted in an OR of 1.1 (CI= 1.0–1.2, $p<0.086$) for results greater than 30. The final logistic regression model demonstrated that smoking more than 10 pack-years was protective (OR=0.31). After adjusting for age, binary logistic regression analysis showed that drinking rural well water for more than 10 years significantly increased the risk of developing PD (by 2.4 for each 10-year period). The development of PD was not correlated with a history of direct pesticide exposure, but it was weakly correlated with a longer history of farming and more frequent use of pesticides. Farming greater than five years had an OR of 1.2 (CI=1.0-1.3, $p=0.041$). The spraying of pesticides more frequently than once per year had an OR of 1.1 (CI=1.0-1.2).

Peterson *et al.* (2008) investigated the association of dietary exposure to PCBs and methylmercury with PD in the Faroe Islands, where, the prevalence of PD is twice as high as would be expected, according to the authors. The OR for occupational exposure to pesticides (OR=6.00, CI=0.62-57.7) suggested an increased, but non-significant risk for PD, where pesticide exposure was classified as ever/never. However, only 2% of the population was exposed to pesticides. Hexachlorocyclohexane (β -HCH), which is an organochlorine pesticide, was also examined. This is addressed in the following section. Whale meat consumption in adulthood was also associated with PD. For those who ate whale meat at least twice per month, there was an increased risk compared to those who ate whale meat less than twice per month (OR=6.5, CI=3.02–14.1). Blubber consumption in adulthood (same frequency) also increased risk (OR=5.61, CI=2.5–12.8).

Firestone *et al.* (2010) conducted a case-control study of self-reported exposure histories among 404 PD cases and 526 controls, matched for age and sex. Newly diagnosed, idiopathic PD cases were identified between 1992 and 2006 at the Group Health Cooperative (GHC) and the University of Washington. A panel of neurologists confirmed PD diagnoses by medical chart review, requiring at least two of the four cardinal signs of PD (bradykinesia, resting tremor, cogwheel rigidity, and postural reflex impairment), one of which had to be bradykinesia or resting tremor. In women, the risk estimate was increased but not statistically significant for the general category of pesticides (aOR=3.9; CI=0.39-39.4), with adjustment for age, ethnicity, and smoking, though the number of exposed subjects was quite small. The risk estimate was also increased for women exposed to category of solvents (aOR=1.7, CI=0.98-3.04) and again it was not statistically significant. In men, none of the risk estimates were increased. For farming and related occupations, although none of the estimates was statistically significant, the trend paralleled an expected increase in pesticide exposure by occupation (i.e., a pesticide worker has a higher expected exposure than a crop farmer, who has a higher expected exposure than an animal farmer), and risk estimates were generally higher for women than men. The study also replicated the commonly observed inverse association between smoking and PD. The authors concluded that the risk of PD was not significantly affected by farming work, metal work, or exposure to pesticides, metals, or solvents. The authors also examined specific pesticides (malathion, parathion, diazinon, DDT, 2,4-D, and paraquat) which are addressed in subsequent sections.

Skeie *et al.* (2010) conducted a case-control study of 212 incident PD patients and 172 age- and gender-matched controls residing in four counties in Norway with a population of one million residents. Health outcomes studied were based on the Unified Parkinson Disease Rating Scale (UPDRS) and classified into tremor dominant (TD), postural instability and gait difficulties (PIGD), and intermediate (IND), based on symptoms. Several environmental risk factors were studied including occupation, smoking, alcohol, coffee consumption, and pesticide exposure. Pesticide exposure was self-reported and not quantified. Self-reported pesticide exposure, whether occupational or private use, was similar in patients and controls (18.0% vs. 17.1%, $p=0.46$). Agricultural work was associated with a higher risk of PD (OR=1.75, CI=1.03-3.0, $p=0.009$). There were no differences identified with other occupations. In the regression model which included intake of alcohol, coffee, and smoking, only coffee ($p=0.007$) and alcohol intake ($p=0.021$) remained significant whereas smoking was no longer significant. Thus, from this paper it seems as though only coffee intake reduces the risk of PD in general while associations to alcohol and smoking differ between PIGD and TD-PD patients.

Rugbjerg *et al.* (2011) conducted a case-control study focusing on recall bias in assessing pesticide exposure. A total of 403 cases were identified through a prescription plan database; patients were included as cases if they had at least one prescription for an anti-parkinsonian drug during the period of 1995-2002. All potential cases were confirmed by an initial screening phone interview about chronic diseases, anti-parkinsonian drugs taken, and the reason for their use. Those taking the drugs for known or suspected PD had an in-person physical assessment employing a checklist and record of symptoms, reviewed by a neurologist with a specialty in movement disorders. Controls (n=405), which were identified through stratified random sampling of the British Columbia (BC) Ministry of Health Services client registry (representing 97.5% of the population), were matched to cases by birth year, gender, and geographic region. Subjects were interviewed about job history, medical/personal habits history, and beliefs about the disease risk factors. Each participant's job history was reviewed by an occupational (also called industrial) hygienist who was blind to case status to determine whether potential exposures of interest commonly associated with an occupation were reported. This occupational hygiene interview was conducted with those reporting pesticide exposures above background. The risk estimates for subcategories of pesticides tended to follow the same pattern: the highest risk estimates were for self-reports; the hygiene review resulted in reductions in risk estimates; and there were slightly higher risk estimates for exposures through pesticide spraying. For example, self-reported exposure to pesticides was associated with an OR of 1.76 (CI=1.15–2.70) among 40-69 year-old participants. When exposures were based on the hygiene interview, the risk decreased to a non-statistically significant OR of 1.51 (CI=0.85–2.69). None of the ORs for pesticide subcategories were statistically significant, except self-reported insecticide exposure (OR=1.80, CI=1.03-3.15). In a second model looking at the relationship between agricultural work and PD (36 cases and 17 controls), those reporting agricultural jobs had a statistically increased risk of PD (OR=2.47, CI=1.18–5.15) when adjusted for gender, birth year, and smoking. However, when hygiene-reviewed exposures were added, the elevated and statistically significant OR for agricultural work remained (OR=2.47, CI=1.18–5.15), but the risk due to pesticide exposure was no longer elevated (OR=0.83, CI=0.43–1.61). This study provides little support for pesticide exposure as a cause of PD. The relationship to agricultural jobs suggests that farming exposures other than those to pesticides, e.g., solvents, fuels, exhaust, dust, microorganisms, traumatic injuries, or endotoxins, should be considered as risk factors for PD.

Parron *et al.* (2011) conducted an ecological study using averaged prevalence rates of PD in selected Andalusian (South Spain) health districts categorized into areas of high and low environmental pesticide exposure. The pesticide exposure categories were based on the number of hectares devoted

to intensive agriculture and pesticide sales per capita. A total of 17,429 cases were collected from computerized hospital records (minimum dataset) between 1998 and 2005. In a bivariate analysis, a statistically significantly increased risk (prevalence rate ratio) was found for PD development (OR= 1.30, CI=1.22-1.39, $p<0.001$) when comparing prevalence rates of high pesticide exposure areas relative to low exposure areas. Males living in high pesticide use areas had a small but significantly increased risk for PD (OR=1.21, CI=1.11-1.32, $p<0.001$) as compared to males living in areas of low pesticide use. Similar to males, females showed statistically increased risk (OR=1.40, CI=1.28-1.52, $p<0.001$). A stepwise multiple logistic regression analysis of the PD development adjusted for exposure to pesticides, gender and age, showed no age differences between the populations residing in high relative to low exposure to pesticides (OR=1.06, CI=1.05-1.06, $p<0.001$); however, males had an increased risk for PD development (OR=1.18, CI=1.10-1.28, $p<0.001$) but the OR for exposure to pesticides was 0.94 (CI=0.87-1.010, $p=0.096$). Therefore, although living in areas with high pesticide use was a statistically significant risk factor for PD in bivariate analysis, this association could not be confirmed significant in multivariate analysis. The authors postulated that these contradictory results may in part be explained by 1) districts of low environmental exposure to pesticides having a greater use of herbicides but located in inland areas, where residents were more likely to drink well water from groundwater aquifer and 2) methodological differences among the studies.

Feldman et al. (2011) found no association between occupational pesticide exposure and PD development in a prospective cohort study of males from the Swedish Twin Study, which followed 14,169 Swedish men for up to 43 years. The authors assessed exposure to 14 chemical and biological compounds, including pesticides, through a job exposure matrix (JEM) based on occupation. The JEM assessed the probability of occupational exposure to chemical and biological compounds in four classes. Class 0 represented a very low probability of exposure (less than 1/10 of persons within the occupational family exposure) while class 3 represented a high probability of exposure (more than 2/3 exposed). Hazard ratios (HR) with 95% confidence intervals adjusted for age, smoking and education were used to estimate the relative risk of disease associated with exposure. For the risk of PD, the reported hazard ratios were 0.8 (CI=0.3-2.2) for class 1 probability of pesticide exposure vs. unexposed and 0.9 (CI=0.5-1.4) for the higher or class 2 probability of pesticide exposure. The authors concluded that there was no association between Parkinson's disease or Parkinsonian disorders and occupational exposure to pesticides and other chemicals examined. There was, however, a significant association with inorganic dust exposure.

A positive but non-statistically significant association was found between the risk for PD and exposure to pesticides among gardeners Kenborg *et al.* (2012) in a cohort study in Denmark. 3124 male members of the Danish Union of Gardeners who were said to have been regularly exposed to a mixture of pesticides during their active working life were recruited for the study. Standardized hospitalization rate ratios (SHR) for PD were calculated as the ratio of two numbers: the numerator (“observed”), which was the actual number of hospital admissions over a specified time period, and the denominator (“expected”), which was the number of hospital admissions that would be expected if patients under the care of that facility experienced hospital admissions at the national rate for patients with similar characteristics. Twenty-eight gardeners were hospitalized with a primary diagnosis of PD, while 24.5 were expected for the general population. The subjects were followed from 1977-2008 for a primary diagnosis of PD and were compared to the general Danish population (using the national insurance database) and matched by age, gender, and calendar period specific rates. The estimated SHR was 1.14 (CI=0.76-1.65). Despite the non-statistically significant findings, the authors concluded that their study indicated a weak but dose-related association between exposure to pesticides and risk for PD and recommended the need for larger studies.

Steenland and co-authors (2013) recruited elderly patients (ages 65 and over) in facilities where free medical exams were provided. Patients were screened with a three-tier tool and evaluated for PD and Alzheimer’s disease. Linear and logistic regression analyses were applied. The authors reported an adjusted OR (aOR) of 2.57 (CI=0.91-7.26, p=0.07) for occupational exposure to pesticides with adjustments for age, sex and education.

Organochlorine

Organochlorine (OC) pesticides include DDT, aldrin, dieldrin, heptachlor, chlordane, and lindane, among others. Lindane is also known as gamma-hexachlorocyclohexane (HCH). Technical-grade HCH was used as an insecticide in the United States and typically contained 10-15% gamma-HCH as well as the alpha (α), beta (β), delta (δ), and epsilon (ϵ) forms of HCH.⁵ β -HCH is a by-product of the manufacturing process of the insecticide γ -HCH (lindane) and may be present in lindane; however, pure β -HCH is not used as a pesticide.

Of the 12 studies analyzing an association between OC pesticide exposure and PD development, 7 provided statistically significant evidence to support this association and 5 studies did not have

⁵ (<http://www.atsdr.cdc.gov/substances/toxsubstance.asp?toxid=138>).

statistically significant evidence. Out of the 7 studies reporting increased risk of PD due to OC pesticide use, statistically significant ORs ranged from 1.4 to 5.8. Most of the statistically significant exposures studied were related to lindane, but one study also included dieldrin.

In one study, the consumption of traditional foods such as whale blubber increased the exposure to β -HCH, which is persistent in the marine environment. The half-life of this chemical is approximately 8 years. Risk factors for PD previously mentioned still apply (e.g. smoking reducing the risk of PD). Collectively, the studies support a multifactorial etiology of PD, influenced by family history of PD, head trauma, and pesticide exposure. The limitations of the studies included exposure misclassification, recall bias, small sample size, and competing risk factors that may not have been taken into consideration.

Positive association

The studies which, for the most part, found a positive and statistically significant association between OC pesticide exposure and risk of PD are summarized here. In some cases, however, the studies were mixed with both positive and no associations.

The previously described case-control study by Seidler and co-authors (1996) found that PD patients were more likely than control subject to have used OC pesticides; however, the adjusted OR was statistically significant only in comparison to the regional control (aOR=5.8, CI= 1.1-30.4) but not for the neighborhood control (aOR=1.6, CI=0.4-6.2). Hancock *et al.* (2008), which was previously described, also examined the risk of exposure to two classes of insecticides: OC and organophosphates. There was an increased risk of PD associated with OC pesticide exposure (OR=1.99, CI= 1.09-3.64). Elbaz *et al.* (2009) found in this previously described study that in men, insecticides were associated with PD (OR=2.2, CI=1.1-4.3) and OC pesticides (mainly lindane) in particular (OR=2.4, CI=1.2-5.0).

The Faroe Island study (Peterson *et al.*, 2008), also previously described, looked specifically at β -HCH, which is persistent in the marine environment, was found at increased levels among the Faroese due to consumption of traditional foods such as whale blubber. Current β -HCH levels among the Faroese were associated with an increased risk of PD (OR=1.4, CI=1.1–2.0). Richardson *et al.* (2009) conducted a case- control study in which 50 PD patients, 43 controls, and 20 patients with Alzheimer's disease (AD) who had been seen at the University of Texas Southwestern Medical Center on June 10, 2002 and Dec. 31, 2007 had serum levels of 19 OC pesticides measured. The researchers identified β -HCH, which was the main OC compound present, age, and sex (being male) as predictor variables for PD status. β -HCH was detectible in the largest number of PD patients (76%, detectible range 0.12/1.80 ng/mL) of any of the pesticides tested. The OR for PD among those with β -HCH levels was over four

times higher compared to controls (OR=4.39, CI= 1.67 – 11.6). There was no statistical difference observed with any of the other pesticides. The authors noted that β -HCH levels remained constant over time: there was no difference in β -HCH levels in samples taken at the two time points (6/10/2002 and 12/31/2007). This is largely because the half-life of β -HCH is approximately 8 years, which means there could be a decreased clearance of β -HCH in the PD patients. Future studies with DNA samples and demographic data on environmental factors such as smoking would provide a way to determine whether β -HCH levels were the result of genetic polymorphisms in one or more enzymes involved in the metabolism of β -HCH or environmental factors.

Weisskopf *et al.* (2010) used Finnish Mobile Clinic (FMC) Health Examination Survey data to form a nested case-control study within this survey population. Serum samples were collected during 1968 to 1972 and analyzed in 2005-2007 for OC pesticides. Incident PD cases were identified through the Social Insurance Registry and confirmed by review of medical records. There were 101 cases and 349 controls matched for age, sex, municipality, and vital status. Locations of cases were rural, semi-rural and industrial communities. Nine persistent OC pesticides were investigated. Only five OC pesticides were found at levels higher in the FMC than the United States National Health and Nutrition Examination Survey (NHANES) population, so these were the pesticides of focus chosen by the authors. These included β -hexachlorocyclohexane (β -HCH), p,p'-dichloro-diphenyl-dichlorethylne (p,p'-DDE), hexachlorobenzene (HCB), and dieldrin. Cases and controls were 20-79 years old at baseline. Because of strong confounding by cigarette smoking among smokers, analyses were restricted to never smokers (n=68 cases, n= 183 controls). Only dieldrin had a significant association with PD among never smokers (aOR=1.95, CI=1.26-3.02, p=0.003), when adjusted for age, sex, region, smoking, triglycerides, cholesterol and other pesticides. Increasing dieldrin concentrations were associated with increased but non-statistically significant ORs (OR per inter quartile range =1.28, CI=0.97-1.69, p=0.08). Drawbacks to the study include that there was a one-time-only measurement of pesticides in serum and this measurement happened decades before the development of PD. Thus, variations in exposure to the pesticides after FMC blood collection are not captured and could introduce measurement error since past exposures do not necessarily predict future exposures. Five pesticides were at higher concentrations in FMC than NHANES because FMC blood was collected before bans on these pesticides in Finland. Based on chemical properties, toxicokinetics, and use patterns, the authors determined that cyclodienes, including dieldrin, are among the more likely candidates to contribute to the development of PD. Dieldrin use was banned in Finland in 1969.

Richardson *et al.* (2011) conducted a case-control study (n=283) to investigate the association between β -HCH levels in blood serum and PD development. Four cohorts representing two discrete time periods (2001-2003 vs. 2006-2008) at two sites were matched by age and by gender; however, the study did not control for smoking. The reported adjusted odds ratio (OR) was from 1.02 to 1.12 for risk of PD per 1 ng/mg increase in β -HCH serum levels across the four cohorts and OR=1.03 (CI=1.00–1.07, p = 0.031) in the pooled analysis. The OR in subjects with β -HCH levels above the inter-quartile range of 39.08 ng/mg cholesterol was 2.85 (CI= 1.8-4.48; p value < 0.001). The authors concluded that elevated levels of serum β -HCH were associated with increased risk for PD development.

Lack of association

The studies which, for the most part, could find no association between OC pesticide exposure and risk of PD are summarized here. In some cases, however, the studies were mixed with both positive and no associations. All of the studies which follow were already described in the general pesticides section, but analyses on OCs specifically are highlighted here.

Hertzman *et al.* (1994) did not find a statistically significant association between PD development and exposure to OC as a group of pesticides in a case-control study carried out in a horticultural region of British Columbia. The authors estimated exposure through an occupational exposure questionnaire.

The review article by Brown *et al.* (2006) included one paper related to OC exposure that was not included in our review. Kamel *et al.* (2001) found an elevated increase for OC pesticide exposure, but it was not a statistically significant (OR=1.8 , CI=0.9- 3.2).

A later paper by Kamel and colleagues (2006) evaluated the risk of PD due to exposure to the OC pesticides aldrin, dieldrin, chlordane, DDT, heptachlor, lindane, and toxaphene. Exposure was estimated through a self-administered questionnaire. However, there were no statistically significant odds ratios for either prevalent or incident PD and any of these pesticides (e.g., aOR=1.4 for incident PD and lindane, CI=0.8-2.5). The OR was adjusted for age, state, and type of participant (applicator or spouse).

Rugbjerg *et al.* (2011) did not find a statistically significant association with self-reported OC pesticide exposure after adjustments for gender, birth year (5-year age groups), and smoking (cumulative pack-years) (aOR=1.23, CI=0.53, 2.85).

Pezzoli and Cereda (2013) investigated the risk of PD associated with exposure to pesticides and solvents using meta-analyses of data from cohort and case-control studies. The authors included prospective cohort and case-control studies providing risk and precision estimates. No association was observed with exposure to OC pesticides.

Organophosphorus

Commonly used organophosphorus (OP) pesticides (also called organophosphates) have included parathion, malathion, methyl parathion, chlorpyrifos, and diazinon, among others. Of the 8 studies analyzing an association between OP pesticide exposure and PD development, 3 studies provided statistically significant evidence to support this association and 5 studies did not have statistically significant evidence. Out of the 3 studies reporting increased risk of PD due to OP pesticide use, statistically significant ORs ranged from 1.89 to 2.5. There was only one study which found a statistically significant association between a specific OP pesticide (chlorpyrifos) and PD. The other two studies examined OP pesticides in general. Although other studies examined malathion, parathion, and diazinon, these were no statistically significant associations with PD. Risk factors for PD previously mentioned still apply (e.g. smoking reducing the risk of PD). Collectively, the studies support a multifactorial etiology of PD, influenced by family history of PD, head trauma, and pesticide exposure. The limitations of the studies included exposure misclassification, recall bias, small sample size, and competing risk factors that may not have been taken into consideration.

Positive association

The studies which, for the most part, found a positive and statistically significant association between OP pesticide exposure and risk of PD are summarized here. In some cases, however, the studies were mixed with both positive and no associations. With the exception of Amanpreet *et al.* (2008), all of the studies have already been described in the general pesticides section.

The case-control study conducted in Germany by Seidler *et al.* (1996;) found that PD patients were more likely than control subject to have used alkylated phosphates/carbamates; however, the adjusted OR was statistically significant only for comparison with regional control subjects (aOR=2.5, CI=1.3-4.6). There was no relationship in comparison to neighborhood controls (aOR=1.8, CI=0.9-3.3). Hancock *et al.* (2008) examined the risk of exposure to two classes of insecticides: organochlorines and organophosphates. Exposure to organophosphorous pesticides was associated with an increased risk of PD (OR=1.89, CI=1.11-3.25).

Amanpreet *et al.* (2008) conducted a case-control study in Texas in order to identify the risk of PD associated with specific pesticide and chemical products. The authors identified cases (n=100) and controls (n=84) from a cohort of PD patients from a neurology practice. Inclusion criteria (age >=50, location of residence in East Texas, diagnosed with PD by standard clinical/lab diagnostic criteria by a neurologist specializing in movement disorders etc.) were identical for cases and controls; the only difference between the two was that controls had no history of PD. A questionnaire gathered

information on demographics, lifestyle activities, family medical history, occupational history, spraying herbicides/pesticides, and use of specific pesticides. There was an increased risk of PD associated with the use of chlorpyrifos products (OR=2.0, CI=1.02-3.8). The authors note that chlorpyrifos are widely used as pesticides in agriculture, as well as in residential settings for termite treatments and lawn care. Although the risk of malathion and parathion exposures were also examined, malathion had an elevated risk that was not statistically significant (OR=1.3, CI=0.7-2.4) and parathion did not have an elevated risk (OR=0.7, CI=0.2–2.5). The small sample size of this case-control study likely limited the ability to find statistically significant results.

Lack of association

The studies which, for the most part, could find no association between OP pesticide exposure and risk of PD are summarized here. In some cases, however, the studies were mixed with both positive and no associations. All of the studies which follow were already described in the general pesticides section, but analyses on OPs specifically are highlighted here.

Firestone *et al.* (2005) demonstrated that the odds ratio for OP were elevated but not statistically significant (OR=1.07, CI=0.46-2.49). The authors reported that the odds ratios for OP paralleled the World Health Organization hazard classifications, with parathion (OR=8.08, CI=0.92-70.85) much higher than diazinon (OR=1.04, CI=0.35-3.06) or malathion (OR=1.01, CI=0.37-2.72), but none of which were statistically significant. Kamel *et al.* (2006) examined the association of several OP pesticides and prevalent and incident PD. While there was an elevated risk for prevalent PD from exposure to chlorpyrifos, it was not statistically significant (OR=1.2, CI=0.7-2.1). There was also an elevated risk for incident PD associated with malathion exposure but this was not statistically significant either (OR=1.2, CI=0.6-2.1).

Firestone *et al.* (2010) examined occupational exposures to specific pesticides; the only increased risk estimate was for men exposed to parathion (OR=5.8, CI=0.66-50.8), the most potent of the OP reported. However, this was not statistically significant. There were no statistically significant associations for parathion, malathion, or diazinon, either.

Rugbjerg *et al.* (2011) did not find a statistically significant association between OP pesticide exposures that had been reviewed by an industrial hygienist and PD (OR=0.74, CI=0.20–2.78). The authors note that since the self-reported risk estimates were uniformly higher, this could suggest that recall bias is at play.

Pezzoli and Cereda (2013) investigated the risk of PD associated with exposure to pesticides and solvents using meta-analyses of data from cohort and case-control studies. The authors included

prospective cohort and case-control studies providing risk and precision estimates. No association was observed with exposure to OP.

Botanical

Botanical pesticides include rotenone, pyrethrins, and neem, among others. Of the 3 studies analyzing an association between botanical pesticide and PD development, all provided statistically significant evidence to support this association. Rotenone was the only botanical pesticide for which ORs were reported. These ORs ranged from 1.7 to 10.9. The highest OR was associated with exposure from gardening in the past year. In the same study, any rotenone use had only a slightly lower risk (OR=10.0). Risk factors for PD previously mentioned still apply (e.g. smoking reducing the risk of PD). Collectively, the studies support a multifactorial etiology of PD, influenced by family history of PD, head trauma, and pesticide exposure. The limitations of the studies included exposure misclassification, recall bias, small sample size, and competing risk factors that may not have been taken into consideration.

Positive association

The studies which, for the most part, found a positive and statistically significant association between OP pesticide exposure and risk of PD are summarized here. In some cases, however, the studies were mixed with both positive and no associations.

Amanpreet *et al.* (2008), which was previously described, also examined rotenone exposure and PD risk. The risk of PD for use of organic pesticides such as rotenone in the past year in gardening was 10.9 (CI=2.5-48.0). For any rotenone use, the risk was slightly lower (OR=10.0, CI=2.9-34.3).

Tanner *et al.* (2011) conducted a case control study nested in the Agricultural Health Study (AHS), which assessed lifetime use of pesticides that either inhibit mitochondrial complex I or cause oxidative stress and the relationship with PD. The AHS cohort members included private pesticide applicators and their spouses. The controls were matched to cases by age, sex, and state (Iowa or North Carolina) at a ratio of approximately 3 controls per case. Diagnosis of PD was determined by agreement of two neurologists after independent review. Pesticide use was assessed by computer-assisted telephone interviews. Based on 110 PD cases and 358 controls, PD was associated with the use of a group of pesticides that inhibit mitochondrial complex I (OR= 1.7, CI=1.0–2.8), which includes rotenone (OR=2.5, CI=1.3–4.7).

Kamel *et al.* (2006) did report an OR for rotenone and prevalent PD (OR=1.7), but no CI was reported. This OR was based on anywhere from 4-10 cases (specific number not cited for rotenone). It is likely that an association was not detected due to the small sample size, but without a CI, the direction of the association cannot be determined.

Quaternary Ammonium

The primary pesticide in this category includes paraquat. Of the 8 studies analyzing an association between quaternary ammonium pesticide exposure and PD development, 3 studies provided statistically significant evidence to support this association and 5 studies did not have statistically significant evidence. Out of the 3 studies reporting increased risk of PD due to quaternary ammonium pesticide use, statistically significant ORs ranged from 1.36 to 3.01. The highest OR was associated with paraquat exposure among traumatic brain injury patients. The risk of PD for paraquat exposure alone ranged from 1.36 to 2.5. Paraquat was the only quaternary pesticide evaluated in these studies. Risk factors for PD previously mentioned still apply (e.g. smoking reducing the risk of PD). Collectively, the studies support a multifactorial etiology of PD, influenced by family history of PD, head trauma, and pesticide exposure. The limitations of the studies included exposure misclassification, recall bias, small sample size, and competing risk factors that may not have been taken into consideration.

Positive association

The studies which, for the most part, found a positive and statistically significant association between quaternary ammonium exposure and risk of PD are summarized here. In some cases, however, the studies were mixed with both positive and no associations. All of the studies which follow, with the exception of Costello *et al.* (2009) and Lee *et al.* (2012) were already described in the general pesticide section, but analyses on paraquat specifically are highlighted here.

Costello *et al.* (2009) conducted a study of 377 PD cases and 341 controls to examine the association between pesticide exposure and PD. Logistic regression was used to control for age, sex, and race. GIS modeling of the pesticide use registry (PUR) provided estimates of actual pesticide exposure, which is an improvement over memory responses from surveys. The combined exposure to maneb (dithiocarbamate) and paraquat resulted in an increased risk of PD (OR=1.74, CI=1.13-2.73).

Tanner *et al.* (2011) examined the use of a group of pesticides that cause oxidative stress and found an increased risk (OR=2.0; CI=1.2–3.6). Paraquat, which falls into this group, was also associated with an increased risk (OR=2.5; CI=1.4–4.7).

Lee, *et al.* (2012; A case-control study involving 357 incident idiopathic PD cases and 754 population controls in Central California (Fresno, Kern and Tulare counties) investigated PD risk due to both traumatic brain injury (TBI) and exposure to pesticides (specifically paraquat). The authors reported a two-fold increase of PD risk for PD patients reporting a TBI (aOR=2.00, 95% CI 1.28-3.14) and a moderate (but weaker) PD risk for exposure to paraquat alone (aOR = 1.36, 95% CI 1.02 – 1.81). However, the risk for developing PD increased three-fold (aOR= 3.01, 95% CI 1.51-6.01) in TBI patients

exposed to paraquat. The authors concluded that exposure to these two risk factors may act together to increase PD risk in a more than additive manner.

Lack of association

The studies which, for the most part, could find no association between paraquat exposure and risk of PD are summarized here. In some cases, however, the studies were mixed with both positive and no associations.

Hertzman *et al.* (1994), as previously reported, did not find a statistically significant association between PD development and exposure to paraquat among men in a case-control study carried out in a horticultural region of British Columbia (OR=1.25, CI=0.34- 4.63).

Kamel *et al.* (2006) reported an elevated OR for paraquat and prevalent PD and it was of statistically significant (OR=1.8, CI= 1.0, 3.4). There was not an association with incident PD and paraquat.

Elbaz *et al.* (2009) previously described, also looked at paraquat exposure; however, no statistically significant association was found (for men over age of 65, OR=1.4 (CI=0.6–3.1). The authors indicate that there may be several reasons an association was not found: 1) paraquat is mainly used as a nonselective herbicide to kill weeds around fields, thus resulting in lower exposure levels compared to other herbicides 2) if gene-environment interactions are involved, paraquat may be associated with PD only among susceptible individuals and 3) toxicological studies have suggested that maneb (dithiocarbamate fungicide) and paraquat act synergistically.

Wang and co-authors (2011) conducted a case-control study (362 incident PD case and 341 controls) among residents of Central Valley, California to investigate association between residential and occupational exposure to ziram, maneb, and paraquat and the risk for PD. GIS-based models were used to estimate both residential and occupational exposure to ziram, maneb, and paraquat from 1974 – 1999. Pesticide Regulation Pesticide Use Report (PUR) data were combined with land use maps and geocoded address information to estimate exposure within a 500-m radius around occupational/residential addresses. The PD risk was estimated for ambient exposures to each pesticide separately and in combinations for both occupational and residential exposures. The combined exposure to maneb, ziram, and paraquat showed a stronger association with PD development than exposure to these individual pesticides alone; those results are described in the carbamate section, below. There was no association between exposure to paraquat and PD for residential exposure alone (OR= 0.77, CI= 0.50-1.17); there was also no statistically significant association for workplace exposure only (OR=1.07,

CI =0.59 – 1.96), but there was a weak statistically significant association for both residential and occupational exposure combined (OR=1.50, CI= 1.03 – 2.18).

A retrospective cohort study conducted in UK reported no evidence of increased mortality from PD (as the underlying or contributing cause of death) among paraquat workers Tomenson and Campbell (2011). The study population was a cohort of workers in Widnes, UK who manufactured paraquat in any of four plants between 1961 and 1995. The cohort group was comprised of 926 males and 42 females as of 6/30/2009. However, females were excluded from the final analysis because of their small numbers. Paraquat exposure was assessed qualitatively for 729 male workers based on their highest exposure to 11 substances including paraquat. Approximately 300 of the 729 workers had high or medium exposure to paraquat. Workers with high exposures were engineering maintenance workers in two plants, and process operators and plant supervisors from all plants. By 06/30/2009, there had been one death from PD as the underlying cause of death, but zero deaths which mentioned PD as a contributing cause among 307 workers. Standardized mortality ratios (SMRs) were calculated using local and national mortality rates. The SMR using local mortality rates was 31 (CI= 1-171) and the SMR for local mortality rates was 32 (CI= 1– 176) for local mortality. The authors concluded that there was no evidence of increased mortality (underlying and mentioned cause) from PD. The following were some limitations of this study: 1) PD could only be ascertained if underlying or contributing cause of death was recorded on the death certificate and 2) a full quantitative paraquat exposure assessment was not conducted. However, the authors argued that the latter was not a limitation because the one worker who died of PD was assessed as having a medium exposure to paraquat and that the level of exposure had declined drastically between 1979 and 1993.

Carbamate or Dithiocarbamate

This type includes the carbamate pesticide methomyl and dithiocarbamate pesticides maneb, zineb, ziram, and mancozeb. Of the 4 studies analyzing an association between carbamate or dithiocarbamate pesticide exposure and PD development, 2 studies provided statistically significant evidence to support this association and 2 studies did not have statistically significant evidence. Out of the 2 studies reporting increased risk of PD due to dithiocarbamate pesticide use, statistically significant ORs ranged from 1.74 to 8.09. The study with the OR of 1.74 examined the residential exposure to a mixture of maneb, a dithiocarbamate, and paraquat, described in the previous section. This OR was adjusted for occupational exposure. The risk of PD was higher among younger individuals (under age 60) exposed to maneb and paraquat compared to those diagnosed at age 60 and over. The highest OR was among those individuals exposed to a combination of maneb and paraquat residentially and occupationally. In

this case, the workplace exposures estimates were higher than residential. This study also indicated that exposures were especially high for those workers diagnosed with PD before the age of 60. Risk factors for PD previously mentioned still apply (e.g. smoking reducing the risk of PD). Collectively, the studies support a multifactorial etiology of PD, influenced by family history of PD, head trauma, and pesticide exposure. The limitations of the studies included exposure misclassification, recall bias, small sample size, and competing risk factors that may not have been taken into consideration.

Positive association

The studies which, for the most part, found a positive and statistically significant association between carbamate exposure and risk of PD are summarized here. In some cases, however, the studies were mixed with both positive and no associations. All of the studies which follow were already described in the general pesticides section, but analyses on carbamates specifically are highlighted here.

Costello *et al.* (2009) enrolled 368 incident PD cases and 341 population controls from the Central Valley of California in a case-control study between 1998-2007. The authors developed and validated an exposure assessment tool based on geographic information systems that integrated information from California Pesticide Use Reports (PUR) and land-use maps to estimate historical exposure to agricultural pesticides in the residential environment. They generated estimates for maneb and paraquat exposures incurred between 1974 and 1999. Combined exposure to both pesticides within 500 m of the home increased PD risk by 75% (OR=1.75, CI=1.13- 2.73), compared to no exposure to these pesticides. After adjustment for occupational pesticide exposure, the OR remained essentially the same (OR=1.74, CI=1.11-2.72). Cases under the age of 60 years at the time of diagnosis were at much higher risk when exposed to both maneb and paraquat in combination (OR=4.17, CI=1.15- 15.16) as compared to no exposure during the period of 1974–1989.

A case-control study by Wang and co-authors (2011), reported that the combined exposure to maneb, ziram, and paraquat at both workplace and residential settings resulted in a substantially greater increase of the risk for PD than exposure to these individual pesticides alone. The study showed that combined exposure to ziram, maneb and paraquat at workplaces increased risk of PD three-fold (OR=3.09, CI=1.69 – 5.64), while combined exposure to ziram and paraquat (without maneb) resulted in an 80% increased risk (OR=1.82, CI =1.03 – 3.21). Workplace exposure estimates were higher than residential and those were especially high for those workers diagnosed with PD at a younger age (before age 60) and exposed to the combination of maneb and paraquat both occupationally and residentially (OR=8.09, CI=2.31 – 33.19). The risk of PD for combined exposure to ziram and paraquat was 5.98 (CI=1.95 – 18.32). The authors suggested that the different mechanisms by which the individual

chemicals contribute to dopaminergic neuron death may act together to dramatically increase the risk of PD. This was the first epidemiologic study reporting 1) effects of ziram and PD risk 2) ambient exposure to pesticides at work being associated with a greater risk of PD than residential exposure alone; and 3) workers who were also exposed at home had the greatest risk of developing PD.

Lack of association

The studies which, for the most part, could find no association between carbamate exposure and risk of PD are summarized here. In some cases, however, the studies were mixed with both positive and no associations. Both studies have already been described previously.

Hertzman *et al.* (1994) did not find a statistically significant association between PD development and exposure to dithiocarbamates (including ferbam) among men in a case-control study carried out in a horticultural region of British Columbia (OR=1.06, CI=0.48, 2.37). Kamel *et al.* (2006) found an elevated but non-statistically significant OR for carbamate and incident PD (OR=1.7, CI=0.7, 3.7). There was no association between prevalent PD and carbamate.

2. Pesticide Exposure with Genetic Interactions

Recent research of PD pathogenesis has revealed that certain environmental-genetic interactions may contribute more to the PD development than environmental or genetic factors alone. Multiple epidemiological studies investigated the effect of pesticide exposure on genetically susceptible individuals/populations or gene-environment relationship for the development of PD. This means that specific genotypes may either enhance or diminish the impact of pesticide exposure on the development of PD. The studies are summarized here by the type of pesticides evaluated for potential interaction with the genetic makeup of exposed individuals. The studies presenting positive associations between pesticide exposure and risk of PD are summarized first.

Pesticides (General)

Of the 11 studies analyzing an association between general pesticide exposure and PD development with possible genetic interactions, 6 provided statistically significant evidence to support an interaction effect between pesticide exposure and genes in modulating the risk of PD and 5 studies did not have statistically significant evidence that there was an interaction between genes and pesticide exposure in PD risk. Out of the 6 studies reporting an increased risk of PD due to general pesticide exposure interacting with genes, statistically significant ORs ranged from 3.17 to 4.9. Genotypes which were implicated include CYP2D6, MDR1, MnSOD, NQ01, and NOS1 SNPs. These genotypes have been

associated with metabolism of pesticides or other chemicals. Of the 5 studies that did not have evidence of an interaction between genes, pesticide exposure and PD, the genotypes included *SNCA*, *GST*, *MAO-A*, and *MDR1*. For 2 studies, the association between PD and pesticide exposure alone could not be determined. Collectively, the studies indicate that genotypes may interact with pesticide exposure to increase the risk of PD. The limitations of the studies included exposure misclassification, recall bias, small sample size, and competing risk factors that may not have been taken into consideration in statistical analyses.

Positive association with interaction

The studies which, for the most part, found a positive and statistically significant association between general pesticide exposure and PD development with genetic interactions are summarized here. In some cases, however, the studies were mixed with both positive and no associations.

Hubble *et al.* (1998) conducted a study among PD patients with dementia (PD+D) and the controls were those without dementia (PD-D). The authors sought to examine the interaction between non-genetic variables and three candidate gene markers: poor debrisoquine metabolizer allele (CYP 2D6 29B+), monoamine oxidase B allele 1, and apolipoprotein E ϵ 4 allele. Subjects with PD were recruited from the outpatient clinic of the Parkinson's Disease Center at the University of Kansas Medical Center. PD was defined by the presence of 2 of 3 cardinal features reported by the authors including tremor, rigidity and bradykinesia and sustained responsiveness to levodopa therapy. Patients with a history or clinical features suggestive of atypical parkinsonism and patients who developed clinically obvious cognitive deficits prior to the motor deficits of PD were excluded. The determination of dementia was based on the Mattis Dementia Rating Scale (DRS). Controls were subjects with total DRS scores above 136, no single DRS subtest score more than 2 standard deviations below the mean and no clinical suspicion of mild dementia. Pesticide exposure was reported by the patient through questionnaire. Exposure was defined as pesticide use for more than 20 days within any calendar year. The initial model looked at genetic and non-genetic variables in a stepwise fashion. There was not a relationship between CYP 2D6 29B+ and PD + D ($p=0.659$). When pesticide exposure was combined with different variables to assess interactions, the results were mixed. The only significant relationship was with the poor metabolizer gene (CYP 2D6 29B+). There was an approximately three-fold increased risk of PD+D given pesticide exposure and presence of the poor metabolizer gene (OR=3.17, CI=1.11–9.05). The authors suggested that pesticide exposure and CYP 2D6 29B+ interact to increase the risk of PD+D. There was no relationship with pesticide exposure combined with the monoamine oxidase B allele 1 or the apolipoprotein E ϵ 4 allele.

Drozdziak *et al.* (2003) recruited 107 PD cases and 103 matched controls of Polish origin from Western Pomerania, Poland. A total of 107 unrelated patients with idiopathic PD (56 males, 51 females) aged 24–82 years were enrolled in the study as cases. All patients were examined by consultant neurologists. PD was diagnosed if at least two of the four main signs of the disease reported by the authors (i.e. tremor at rest, rigidity, hypokinesia, and postural reflex impairment) were observed. There is a hypothesis about association between the 3435TT genotype of the *MDR1* gene and early onset PD). The risk of PD development in patients exposed to pesticides versus non-exposed subjects was significantly increased in heterozygous (3435CT) patients (OR=2.9, CI=1.2–7.1, $p<0.01$), non-statistically significant in homozygous (3435TT) patients (OR=1.3, CI=0.4–4.4) and approximately five times higher for both groups together (3435CT + 3435TT) (OR=4.9, CI=1.7–14.6). In summary, this study revealed an association between the *MDR1* gene polymorphism and PD in subjects exposed to pesticides. Consequently, the authors indicate that prevention or treatment methods could be implemented in those individuals with the 3435T allele who are exposed to pesticides. These methods could include elimination of exposure to pesticides or, if this is not possible, provide antioxidants to reduce the effects on brain cells.

Elbaz *et al.* (2004) performed a case–control study of PD in a population characterized by a high prevalence of pesticide exposure and studied the joint effect of pesticide exposure and cytochrome P450 D6 (CYP2D6*4) gene activity with PD. Subjects (190 cases and 419 controls) were selected among those enrolled in a French health insurance for agricultural workers. As part of the study protocol, they were examined by a neurologist with experience in movement disorders. Whenever it was impossible to directly examine the patient, the patient’s treating neurologist was contacted to obtain clinical information. PD was defined as the presence of parkinsonism (presence of at least two cardinal signs: rest tremor, bradykinesia, rigidity, impaired postural reflexes) after exclusion of other causes of parkinsonism. Controls were recruited among all Mutualité Sociale Agricole affiliates who made requests to be reimbursed for health care expenses. A maximum of three controls were matched to each case for age, sex, and region of residency. Cases between ages 18-75 had submitted coverage for PD. Pesticide exposure was assessed by occupational health physicians using an individual “expert evaluation procedure.” Occupational pesticide exposure combined with low CYP2D6*4 (two alleles) activity resulted in increased risk for PD (OR=4.74, CI=1.29–17.45, $p=0.02$). When occupational exposure and gardening exposure were combined, the risk was slightly lower for the poor metabolizers (two alleles of CYP2D6*4) (OR=3.28, CI=1.16–9.27). Thus poor metabolizers may have an increased risk of PD with pesticide exposure. Without pesticide exposure, there is no increased risk.

In another case-control study of a Taiwanese population Fong *et al.* (2007), the association between exposure to pesticides and the PD risk was examined in conjunction with genotypes implicated in pesticide metabolism: manganese-containing superoxide dismutase (*MnSOD*, allele-9 T>C) and NAD(P)H:quinone oxidoreductase 1 (*NQO1*, allele 609 C>T). PD patients (n=153) and controls (n=155) were matched for age, sex, and origin. PD risk was statistically significantly associated with exposure to pesticides (OR=1.69, CI=1.07-2.65, p=0.023) and this association remained significant after adjustment for age, sex, and cigarette smoking (aOR=1.68, CI=1.03–2.76, p=0.023). Considering genetic factors, there were no significant differences in frequencies of both genotypes of *MnSOD* and *NQO1* polymorphisms between PD patients and the control subjects (p>0.05). However, the difference in genotype distribution was significant among subjects who had been exposed to pesticides, with aOR of 2.49 (CI=1.18–5.26, p=0.0072) for *MnSOD* C allele and aOR of 2.42 (CI=1.16–4.76, p=0.0089) for *NQO1* T allele, respectively. Furthermore the combined *MnSOD/NQO1* variant genotype was significantly associated with a 4.09-fold increased risk of PD (CI=1.34–10.64, p=0.0052), among subjects exposed to pesticide. This study provided evidence that susceptible variants of *MnSOD* and *NQO1* genes may interact with occupational pesticide exposure to increase PD risk in southwestern Taiwanese.

Hancock *et al.* (2008) examined the joint effect of exposure to pesticides and nitric oxide synthase genes, which may create nitric oxide that contributes to neurodegeneration in PD. For the pesticide exposure portion of the study, families with PD were selected. Individuals enrolled through the Udall Center were administered a structured telephone questionnaire to collect detailed environmental risk factor data on demographics, health and habits, and pesticide and other chemical exposures. The pesticide exposure section assessed whether participants applied pesticides at work or in their home, garden, or lawn. Only first-hand exposures were considered. Participants who reported applying pesticides were asked to list the name of any pesticide they remembered using, the number of days it was used, and the years application started and stopped (if applicable). The reported pesticides were classified into specific chemical classes, but sample size was not sufficient to examine gene-environment interactions. Instead, the reported pesticide chemicals were classified more broadly into functional groups (e.g., insecticides or herbicides). Those cases who reported applying these prior to the age at disease onset were considered ever exposed. Gene-environment interaction analyses focused on 163 cases and 178 relatives and other controls from 168 sporadic PD families with environmental risk factor data available. Interactions were found between pesticides (insecticides, herbicides) and the *NOS1* SNPs (single nucleotide polymorphisms) rs12829185 (OR=3.12, CI=1.71–5.71), rs1047735 (no OR given), and rs2682826 (OR=3.52, CI=1.78–6.95). The authors indicate that findings support *NOS1* and *NOS2A* as

genetic risk factors for PD and show that these genes might modify the effects of established environmental factors for PD.

Zschieidrich *et al.* (2009) conducted a large case-control study to investigate the potential relationship between *MDR1* variants and PD. *MDR1* variants were determined in 599 Europeans (415 PD patients and 184 healthy controls recruited between 1999 and 2006). The population was further stratified by ethnicity, age at PD onset (≤ 45 years vs. >45) and exposure to pesticides. For 86 German patients and 54 German controls from the population-based study, data on private use of pesticides were obtained through a structured interview (answer options “never,” “very rarely,” “occasionally,” and “regularly.” Occupational exposure to pesticides in these individuals was estimated by using a job exposure matrix (JEM). Patients were classified as “exposed” when either answering “occasionally” or “regularly” in the interview or when obtaining a JEM score of >10 . Genomic DNA was extracted from venous blood samples. To test for the potential interaction between the c.3435C/T variant and pesticide exposure with respect to PD risk, a case-only analysis was performed. The case-only analysis revealed a statistically different genotype distribution at the c.3435C/T between PD patients exposed to pesticides compared to those non-exposed (OR=4.74; CI=1.01-22.3; $p=0.047$). The study demonstrated evidence that genetic variants of the *MDR1* might act as a modulator of PD risk in patients exposed to pesticides. Thus variants in *MDR1* could be another example of a susceptibility factor increasing the risk for PD in conjunction with an environmental factor, such as pesticide exposure. However no ORs were provided for the relationship between pesticide exposure and PD without variants of *MDR1*. The authors indicate that the results suggest another link between genetic variants of detoxifying enzymes, pesticide exposure, and PD.

Positive association without interaction

The studies which could find no evidence to support a gene-environment interaction affecting the association between general pesticide exposure and the risk of PD are summarized here.

Fong *et al.* (2005) conducted a case-control study to investigate the association between paraoxonase I (*PNO1*) polymorphism, pesticide exposure and risk of PD in Taiwanese population. Patients with idiopathic PD ($n=125$; 69 women and 56 men) were recruited for the study from a general hospital between July 2002 and June 2004. Unrelated controls ($n=162$; 90 women and 72 men) matched with the patients on age and sex were recruited from the outpatient clinic with diagnoses of back pain or cervical spondylosis. History of exposure to environmental factors and data on other factors were collected using a questionnaire filled out during a face-to-face interview. These included data on years of farming, drinking water sources, occupational exposure to pesticides, duration of pesticide exposure,

and age at the initial pesticide exposure. A positive exposure was defined as an occupational or residential contact with a given factor for at least 12 months prior to the onset of PD. Buccal mucosa cells were collected from each PD and control subjects to determine *PNO1* polymorphism status. There was a statistically significant association between the risk of PD and exposure to pesticides (OR=1.72, CI=1.07-2.75, p=0.025) and the risk of PD did not increase for those who had ever used well water. The relative risk of PD was 2.14 with ≥ 36 years of exposure to pesticides (OR=2.14, CI=1.23-3.71, p=0.005). There was no difference between the patients and the controls in the genotype of *PNO1* (p=0.504). This study revealed the strong relationship between long-term (≥ 36 years) exposure to pesticides and the development of PD. No significant differences were observed in the distribution of *PNO1* genotypes between PD patients and controls.

Brighina *et al.* (2008) conducted a case-control study in order to investigate the possible joint effects of *SNCA REP1* genotypes and pesticide exposure and the risk of PD. The study involved 833 case-control pairs (472 case-unaaffected sibling pairs and 361 case-unrelated control pairs). Cases (patients with PD) who resided in MN or in one of the surrounding four states (WI, IO, SD, or ND) were recruited prospectively from the Dept. of Neurology at the Mayo Clinic (Rochester, MN) after June 1, 1996. Controls included unaaffected siblings of cases or unrelated population control subjects, who screened negative for PD or were confirmed not to have PD by clinical assessment. Cases included more men than the control group (63.6% vs. 57.0%); the median age at PD onset was 61.9 years; case and controls were primarily white and of European descent. Genomic DNA was extracted from leukocytes (venous blood samples were obtained from all cases and controls). Pesticide exposure data were obtained by telephone interview using a structured risk factors questionnaire. Exposure to pesticide was classified as "overall" (occupational, including farming; residential, including gardening; or both) and was categorized by indication for use (herbicides, insecticides, or fungicides) and by chemical group of the active ingredients. Analyses were 1) adjusted for age and sex and 2) performed for subjects overall and stratified by family history of PD, age at study, and sex. The authors observed a statistically significantly increased risk of PD with increasing *SNCA REP1* bp length (OR=1.18 for each score unit; CI=1.02-1.37; p=0.03). Pesticide use (ever/never) was not associated with an increased risk of PD in the sample overall (OR=1.11, CI= 0.89-1.38, p=0.37); however, pesticide use was associated with PD in younger subjects only (≤ 59.8 years, OR=1.80, CI=1.12-2.87, p=0.01). For the functional pesticides subgroups (herbicides, insecticides, or fungicides), only herbicides use was associated with PD and only in younger subjects (OR=2.46, CI=1.34-4.52, p=0.004). This may be related to the fact that in this study population, the frequency of pesticide exposures considered separately were significantly greater in younger than in

older subjects. Patients with PD were more likely than controls to use pesticides belonging to the chlorophenoxy acid or esters chemical class, which are used as herbicides (OR=1.52, CI=1.04-2.22, p=0.004). Furthermore, 2,4-D was the most commonly reported chlorophenoxy herbicide by the study subjects. No other subclass was significantly associated with PD, though there were 44 different chemical subclasses of pesticides identified. Also, the number of chemical subclasses reported was greater in younger than older subjects. In multivariate analysis, both *SNCA REP1* score and pesticide exposures were statistically significantly associated with PD in younger subjects, but there were no pairwise interactions; the latter finding suggests other genetic loci confer susceptibility to PD in families. After restricting the susceptibility analysis to the youngest quartile of subjects, the best-fitted model included *SNCA REP1* genotype score and herbicides; both *SNCA REP1* genotype score (OR=1.65, CI=1.16-2.35) and herbicides (OR=2.39, CI=1.29-4.41) were significant. The study findings suggest that *SNCA REP1* genotype and exposure to herbicides have independent effects on risk of PD, primarily in younger individuals.

Kiyohara *et al.* (2010) examined the relationship of the seven GST polymorphisms (*GSTM1* deletion, *GSTT1* deletion, *GSTP1* rs1695, *GSTO1* rs4925, *GSTO1* rs11191972, *GSTO2* rs156697 and *GSTO2* rs2297235) and risk of PD, with special reference to the interaction with self-reported use of pesticides (home pesticide use, occupational pesticide use, and either home or occupational pesticide use) or cigarette smoking. This case-control study involved 238 PD patients as cases and 370 controls recruited from a Japanese population. PD patients were recruited at hospitals in Fukuoka Prefecture, Kyushu Island (southern Japan), Osaka, Kyoto, and Wakayama Prefecture. Eligible cases were those within 6 years of PD onset and presented at one of the 11 collaborating hospitals between April 1, 2006 and March 31, 2008. Hospital controls were recruited from among patients without a previous diagnosis of a neurodegenerative/malignant disease. Controls were not matched to cases. Genomic DNA was extracted from buccal samples. An unconditional logistic regression model was developed for each polymorphism (presence or absence of a null allele or a number of less active alleles) and was used to predict PD status. There were three measures for biologic interactions (additive scale): 1) the relative risk, 2) attributable proportion, and 3) synergy index. No statistically significant ORs, adjusted for age, sex, smoking status, pesticide exposure, and region of residence, were found for the GST polymorphisms, indicating that none of the GST polymorphisms was associated with PD. Cigarette smoking was statistically significantly associated with decreased risk for PD, however, no interaction was observed with any of the GSTs studied. Self-reported pesticide use was not associated with increased risk of PD (home pesticide use: p=0.19; occupational pesticide use: p=0.74; either home or occupational

pesticide use: $p=0.16$). There was no evidence of interaction between self-reported pesticide use and either GST polymorphism. The study assessed interactions between the *GST01* rs4925 and *GST02* rs156697 SNPs and pesticide use, specifically. The results suggest that the tested GST polymorphisms did not have an important role in PD susceptibility in the studied Japanese population. The power of the study to detect an interactive effect between GSTP polymorphisms and pesticide use was low due to a small number of pesticide users.

Unknown association without interaction

The studies which found no evidence to support a genetic interaction with the risk of PD, but did not specifically address the contribution of pesticide exposure to PD risk alone are summarized here.

A case-control study of 959 prevalent cases of parkinsonism (767 with PD) and 1989 controls was carried out across five European centers by Dick and co-authors (2007b). Occupational hygienists estimated the average annual intensity of exposure to solvents, pesticides and metals, (iron, copper, manganese) and were blind to disease status. There was a modest but significant association between the *MAO-A* polymorphism in males and the risk of PD (G vs T; aOR=1.30, CI =1.02-1.66). Also, the gene-environment interactions analysis for all cases (parkinsonism and PD) yielded little evidence of interaction effects between environmental and genetic factors; none of the analyses conducted being significant at the 5% level. However, the authors noted that a number of interactions may be worthy of further study. There were possible interaction effects between *GSTM1* null genotype and solvent exposure (which were stronger when limited to PD cases only). For polymorphism *PON55* in combination with pesticide exposure, there was an aOR of 4.4 (CI=0.88 to 22.3, $p=0.07$) adjusted for age, sex, country, ever used tobacco containing product, ever knocked unconscious and first degree family history of PD.

Kiyohara *et al.* (2013) recruited 606 subjects (238 PD cases and 368 controls) from various hospitals in Japan. Prevalent cases were within 6 years of the onset of PD and presented at one of the hospitals included in the study. The study aimed to determine the impact of the *MDR1* C3435T polymorphism on PD risk alone or in combination with environmental factors (smoking status, alcohol use, pesticide use). Subjects with the *TT* genotype of the *MDR1* C3435T polymorphism showed a non-significantly increased risk of PD (OR=1.49, CI=0.85-2.25) compared with those with the *CC* genotype. A gene-environment interaction was suggested only for alcohol consumption, with a combination of at least one T allele and ever drinking alcohol conferring significantly higher risk of PD (OR=1.83, CI=1.07-3.15, $p=0.029$), compared with the *CC* genotype and never drinking alcohol.

Organochlorine

Organochlorine (OC) pesticides include DDT, aldrin, dieldrin, heptachlor, chlordane, and lindane, among others. Lindane is also known as gamma-hexachlorocyclohexane (HCH). Technical-grade HCH was used as an insecticide in the United States and typically contained 10-15% gamma-HCH as well as the alpha (α), beta (β), delta (δ), and epsilon (ϵ) forms of HCH

(<http://www.atsdr.cdc.gov/substances/toxsubstance.asp?toxid=138>). Only one OC study examined the gene-OC pesticide exposure interaction. While there was a positive association between OC exposure and the risk of PD, the gene *ABCB1* did not modify this risk.

Positive association without interaction

The studies which could find no evidence to support a gene-environment interaction affecting the association between general pesticide exposure and the risk of PD are summarized here.

Dutheil *et al.* (2010) conducted a case-control study to examine the association between PD and two polymorphisms in the gene *ABCB1*, as well as the interaction between *ABCB1* polymorphism and OC insecticides. Patients enrolled in the French health system for agricultural workers (Mutualité Sociale Agricole) with PD (n=207) were examined by a neurologist and were matched to controls (n=482). Participants were classified as never users of pesticides, users of pesticides for gardening, and professional users of pesticides. Detailed information on lifelong pesticides use was obtained for professional users by occupational health physicians. Pesticides were coded using a pesticide dictionary and were grouped into various categories of pesticides. However, the authors focused on OC pesticides because in previous studies they were found to display the most robust association with PD and a dose-effect trend. Males with OC insecticide exposure (confirmed non-gardening exposure) were found to have an increased risk of PD (OR=2.2, CI=1.1-4.5.), which was statistically significant. The occurrence of two *ABCB1* polymorphisms (C3435T, G2677) did not differ between cases and controls (p=0.43 and p=0.97, respectively). Although statistically not significant, the risk of PD among those men who had OC exposure and were homozygous carriers of variant G2677 (A,T) was 3.5 (CI=0.9-14.5).

Organophosphorus

Commonly used organophosphorus (OP) pesticides (also called organophosphates) have included parathion, malathion, methyl parathion, chlorpyrifos, and diazinon, among others. Of the 2 studies analyzing an association between OP pesticide exposure and PD development with possible genetic interactions, both provided statistically significant evidence to support an interaction effect between OP pesticide exposure and genes in modulating the risk of PD. Both studies examined the *PON1-55* genotype and one study examined the *PON1-192QQ*. There was an interaction effect with chlorpyrifos

(OR=2.61) for the MM genotype compared to unexposed heterozygous carriers in one study and in another the OR was 2.45. There was also increased risk for chlorpyrifos exposure for individuals with the *PON1-192QQ* genotype (OR=1.95). The highest risk for PD was when there was chlorpyrifos and both genotypes (OR=3.28). Diazinon exposure in combination with the MM genotype also increased risk (OR=5.30). The *PON1* gene appears to play a mediating role in the etiology of PD among individuals exposed to chlorpyrifos, diazinon, and potentially other pesticides which are detoxified by *PON1* enzymes.

Positive association with or without interaction

The studies which, for the most part, found a positive and statistically significant association between general pesticide exposure and PD development with and without genetic interactions are summarized here.

Manthripragada *et al.* (2010) examined the association between PD and exposure to diazinon, chlorpyrifos, parathion as well as the influence of the *PON1-55* MM variant genotype on PD risk. From 2001 to 2008, 351 cases and 363 controls were recruited from three rural California counties. Pesticide exposure was assessed by self-reporting and a geographic information system (GIS). The authors found an increased risk of PD among carriers of *PON1-55* MM exposed to OP pesticides (OR=2.2, CI=1.1-4.5) compared to the heterozygous genotype and those not exposed. Carrying the MM genotype and having been exposed to chlorpyrifos increased the risk of PD nearly three times compared to unexposed wildtype/heterozygous *PON1-55* carriers (OR=2.61, CI=1.25–5.44). Those who were exposed to chlorpyrifos but did not carry the homozygous variant genotype experienced a moderate increase in PD risk (OR= 1.48, CI=1.04–2.12). For chlorpyrifos exposure among those individuals with an onset of PD at age 60 or younger and MM genotype carriers, there was also an increased risk (OR=5.3, CI=1.7-16). For diazinon exposure among individuals with carriers of the MM genotype, there was a doubling of risk compared with individuals with the wildtype or heterozygous genotype and no diazinon exposure (OR=2.24, CI=1.12–4.48). Those who were exposed to any diazinon but did not carry the homozygous variant genotype exhibited little to no increased risk of PD (OR=1.18, CI=0.83–1.68). For those highly exposed to diazinon, the authors found an even larger increase in risk for MM carriers (zero/low versus high: OR=5.30, CI=1.71-16.4), and strong interaction on the multiplicative scale (OR_{interaction}=4.59, CI = 1.37–15.4). The authors did not find any increase in PD risk with parathion exposure, even among those who were *PON1-55* MM genotype carriers.

Lee *et al.* (2013) found increased risk of PD among *PON1-55* MM carriers with exposure to chlorpyrifos (aOR=2.45, CI=1.24-4.83); no statistically significant interaction was observed between

exposure to diazinon and parathion (for diazinon, aOR=1.84, CI=0.97-3.47; for parathion, aRO=1.78, CI=0.89-3.60). The risk of PD among *PON1-192QQ* carriers with exposure to chlorpyrifos was also increased (aOR=1.95, CI=1.13-3.37) and again, it was not statistically significant for exposure to diazinon (aOR=1.53, CI=0.90-2.61) and parathion (aOR=1.65, CI=0.95-2.89). The highest risk of PD was found among those with *PON1-55 MM-192QQ* and exposure to chlorpyrifos (OR=3.28, CI=1.02-10.58).

Quarternary Ammonium

The primary pesticide in this category includes paraquat. Of the 3 studies analyzing an association between quaternary ammonium pesticides and PD development with possible genetic interactions, 2 provided statistically significant evidence to support an interaction effect between quaternary ammonium exposure and genes in modulating the risk of PD. These studies looked at *DAT* and *GSTT1* genotypes. There was an interaction effect with maneb and paraquat exposure and PD (OR=4.53) with individuals who had two or more *DAT* susceptibility genes compared to those with zero susceptibility genes. There was also increased risk of PD among those highly exposed to paraquat with a *GSTT1* homozygous deletion (OR=11.1). One study did not find an interaction with *SNCA* and it was unknown if there was an association between PD and paraquat exposure without *SNCA*.

Positive association with interaction

The studies which found evidence to support a gene-environment interaction affecting the association between quaternary ammonium pesticide exposure and risk of PD are summarized here.

Ritz *et al.* (2009) conducted a population-based case-control study of 324 incident PD patients and 334 population controls. Subjects were recruited in three California counties. Cases were recruited by referral and confirmed by UCLA movement disorder specialists. Exposure to pesticides was assessed using telephone interview and GIS modeling. Participants also provided blood or buccal samples for genetic analyses. The study looked at the association between exposure to paraquat and maneb and PD alone, as well as the gene-environment hypothesis that dopamine transporter (*DAT*) genetic variants interact with pesticide exposure to increase the risk of PD. High residential exposures to both paraquat and maneb between 1974 and 1999 increased the risk of PD more than two-fold (aOR=2.32, CI=1.23–4.40), and occupational exposure increased the risk of PD by approximately 50% in males (aOR=1.56, CI=0.95–2.56), but the latter association was statistically non-significant. The authors found that high exposure to maneb and paraquat increased PD risk almost 3-fold but not significantly, in subjects who carried one *DAT* susceptibility allele (OR=2.99, CI=0.88-10.21) and as much as a 4.5-fold in carriers of two or more susceptibility alleles (OR=4.53, 95% CI=1.70–12.1). In subjects with little or no residential

exposure to these pesticides, there was no increase in risk with susceptibility allele carrier status or increasing number of susceptibility alleles.

Goldman and Samuel (2012) recruited participants from the Agricultural Health Study (prospective study of licensed pesticide applicators, mostly farmers) and their spouses for the Farming and Movement Evaluation case-control study. PD risk was the outcome measured. Subjects from Iowa and North Carolina with PD (n=87) and 343 matched controls were recruited. Paraquat use was determined by interview. PD diagnosis was confirmed by in-person examination. Subjects were genotyped for homozygous deletion of *GSTM1* and *GSTT1*. These are glutathione transferase genes, which detoxify a wide range of xenobiotic compounds. The hypothesis was that a deficient enzyme might enhance the neurotoxicity of paraquat. Analyses were restricted to men because no women used paraquat. The general risk of PD (without reference to genotype) for those who ever used paraquat was statistically significantly increased (aOR=2.6, CI=1.3-5.0) with adjustment for age, sex, state (Iowa or North Carolina), and smoking status. Men with functional *GSTT1* and paraquat use had a statistically insignificant PD risk (aOR=1.5, CI=0.6-3.6); homozygous deletion of *GSTT1* (*GSTT1*0*) and paraquat exposure had an OR of 11.1 (CI=3.0-44.6, p=0.027). PD risk with *GSTT1*0* and no paraquat exposure was not significant: aOR=1.1 (CI=0.4-2.4). Greater total years of paraquat use was strongly associated with increased risk of PD in those with *GSTT1*0* (p trend= 0.001).

Unknown association without interaction

The studies which found evidence to support a genetic interaction with the risk of PD, but did not specifically address the contribution of pesticide exposure to PD risk alone are summarized here.

Gatto *et al.* (2010) used a population-based case-control study to examine if the risk of PD depended on the combined presence of α -synuclein (*SNCA*) gene variations and pesticide exposures. Abnormal aggregation of the α -synuclein protein, a major component of Lewy bodies and a hallmark of PD, is believed to be important in the molecular pathogenesis of the disease. Several single nucleotide polymorphisms (SNPs) and haplotypes in the *SNCA* promoter have also been shown to be associated with the risk of sporadic PD, and increasing REP1 length is associated with an increased risk of PD, as well as possibly with a decrease in age of PD onset. PD cases (n=333) were recruited from three rural California counties and matched to 336 population controls from the same area. Cases were confirmed by University of California, Los Angeles (UCLA) movement disorder specialists. This study was unique in that it estimated long-term pesticide exposure from agricultural applications in rural counties of California's Central Valley using a geographic information system approach that integrates unique state-mandated pesticide use reports (PUR) and land use data. The authors summed intensity of pesticide

applications (pounds per acre applied within a 500-meter buffer zone) at residences across a 26-year period (1974 to 1979) to calculate the cumulative total intensity of ambient paraquat exposure for each subject. Exposure to paraquat at or above the median value in the control population was considered high and below the median (including no exposure) was considered low/no exposure. There were no statistically significant findings regarding the interaction between pesticide exposure and *SNCA* variations on the risk of PD. However, there were some positive, non-significant associations. For example, among those who were homozygous or heterozygous for the *REP1 263* genotype and high paraquat exposure, the aOR=1.45 (CI=0.59–3.59) compared to those without low or no paraquat exposure, as adjusted age, sex, education, race, smoking status, and family history of PD (positive/negative in a first-degree relative).

Carbamate or Dithiocarbamate

This type includes the carbamate pesticide Methomyl and dithiocarbamate pesticides maneb, zineb, ziram, mancozeb. There was one study which examined the exposure of maneb and paraquat among individuals with the *DAT* susceptibility gene. The risk for PD increased with exposure to these two pesticides among individuals with two or more *DAT* susceptibility genes (OR=4.53).

Positive association and/or interaction

There was one study that found an evidence to support a gene-environment interaction affecting the association between dithiocarbamates pesticide exposure and risk of PD and it is summarized here.

Ritz *et al.* (2009) study, which was described in the previous section, addressed the combination of maneb and paraquat exposure, with a resulting increased OR among those carrying *DAT* susceptibility alleles. High exposure to maneb and paraquat in carriers of one susceptibility allele increased PD risk three-fold (OR=2.99, CI=0.88-10.2), but this was not statistically significant. In carriers of two or more alleles, the PD risk increased four-fold (OR=4.53, CI=1.70-12.1). Similar results were obtained for occupational exposure to maneb and paraquat.

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Data Abstraction Form

Tracking Information:

Reviewer initials: _____ Article Number: _____

Primary author last name: _____

Year article/report published: _____

Title of Article/Report:

Publication details [Journal or report name, volume (issue), pages]:

Study Type (check one):

Published peer-reviewed journal article

Government report

Public health authority

Other (describe)

Report Details

Brief description, including the study population (race/ethnicity, age, sex) and outcome measures: (i.e. cancer)

Measure of Association (i.e. odds ratios, rate ratio)

Statistically Significant Finding? (cite p-value or confidence intervals for controls versus exposed, i.e.).

Study Recommendations (if any)?

Appendix I: Pesticide active ingredients looked up for Pesticides and Parkinson's Committee public group. Sources include NSPIRS registration data, EPA registration information, Agency for Toxic Substances and Disease Registry, and the Compendium of Pesticide Common Names.

Name	Type	Chemical Class	Registration Status	Some Potential NM Sites Include	Other Sites Include	Notes
Maneb	Fungicide	Dithiocarbamate	Inactive	n/a	n/a	Last uses CANCELED in 2010
Heptachlor	Insecticide	Cyclodiene	Inactive	n/a	n/a	Most uses CANCELED in 1974; final cancelation in 1999
Endrin	Insecticide	Cyclodiene	Inactive	n/a	n/a	Most uses CANCELED in 1986; final cancelation in 1991
Aldrin	Insecticide	Cyclodiene	Inactive	n/a	n/a	CANCELED by USDA in 1970; EPA lifted in 1972 for limited use; final use canceled 1987
Dieldrin	Insecticide	Cyclodiene	Inactive	n/a	n/a	CANCELED by USDA in 1970; EPA lifted in 1972 for limited use; final use canceled 1987

Name	Type	Chemical Class	Registration Status	Some Potential NM Sites Include	Other Sites Include	Notes
Benomyl	Fungicide	Benzimidazole fungicide; carbamate acaricide	Inactive	n/a	n/a	CANCELED in '80s-'90s
Zineb	Fungicide	Dithiocarbamate	Inactive	n/a	n/a	CANCELED in 1991
Lindane	Insecticide	Organochlorine	Inactive	n/a	n/a	Most uses CANCELED by 1999; all uses canceled by 2006. Pharmaceutical use remains.
Ziram	Fungicide	Dithiocarbamate	Active Not restricted 2 registered in 2013	Pecans, ornamentals	Berries, many fruits, flowers	Fungal diseases rare in NM
Rotenone	Piscicide, insecticide	Botanical	Active Restricted 2 piscicides registered in 2013	Aquatic sites for control of undesirable fish		
Propargite	Miticide	Sulfite	Active All are restricted 4 registered in 2013	Potatoes, corn, beans, peanuts, cotton, alfalfa	Berries, citrus, mint, hops, carrots, many fruits	

Name	Type	Chemical Class	Registration Status	Some Potential NM Sites Include	Other Sites Include	Notes
Diazinon	Insecticide	Organphosphorus	Active Many are restricted 9 registered in 2013	Lettuce, onions, nursery plants, dairy cattle, beef cattle	Grapes, many veggies, berries, melons, fruits	
Mancozeb	Fungicide	Dithiocarbamate	Active Not restricted 44 registered in 2013	Potatoes, onions	Melons, cucurbits, mangos, applies, citrus, cranberries	Fungal diseases rare in NM
Endosulfan	Insecticide/miticide	Cyclodiene	Active Restricted 3 registered in 2013	Pecans, cotton, wheat, ornamentals 3 registered in 2013	Cucurbits, many vegetables, peaches, cherries, tobacco	
Permethrin	Insecticide/miticide	Pyrethroid	Active Most are not restricted 556 registered in 2013	Many sites; many are nonagricultural. Homes, yards, garden dusts, dogs & cats, buildings, lettuce, pecans, many vegetables, ornamentals, etc.	Berries, many fruits, flowers, many vegetables, nuts, etc.	

Name	Type	Chemical Class	Registration Status	Some Potential NM Sites Include	Other Sites Include	Notes
Cypermethrin	Insecticide/miticide	Pyrethroid	Active Most are not restricted 80 registered in 2013	Many sites; many are nonagricultural. Foggers, roach sprays, pecans, onions, cotton, lettuce	Other greens, broccoli, cauliflower, garlic	
Chlorpyrifos	Insecticide/miticide	Organophosphorus	Active Some restricted, some not 46 registered in 2013	Alfalfa, cotton, onions, pecan, cattle, wood products, industrial sites, ant dens, vector control	Citrus, soybeans, tobacco, many fruits, many vegetables	
Methomyl	Insecticide/miticide	Carbamate	Active Some restricted, some not. 9 registered in 2013	Many food crops, restaurants, sod, food processing plants	Fruits, vegetables, grains, tobacco, mint, barns	
2,4-D	Herbicide	Phenoxyacetic	Active Not federally restricted 383 registered in 2013	Many agricultural & nonagricultural sites		

Name	Type	Chemical Class	Registration Status	Some Potential NM Sites Include	Other Sites Include	Notes
Paraquat	Herbicide, desiccant	Quaternary ammonium	Active Restricted 12 registered in 2013	Many agricultural sites. Pecans, alfalfa, cotton, peanuts, grapes, lettuce	Berries, citrus, greens, melons, many fruits, many vegetables, soybeans, tobacco, grains, ornamentals, sod, conifers	

Appendix II: Results of PubMed Search on July 2nd, 2013
((Parkinson's Disease) AND pesticide) OR insecticide, herbicide, rodenticide, fungicide

Items 1 - 200 of 1547 ([Display the 200 citations in PubMed](#))

1. [The Parkinson's disease protein DJ-1 binds metals and protects against metal induced cytotoxicity.](#)
Bjorkblom B, Adilbayeva A, Maple-Grodem J, Piston D, Okvist M, Xu XM, Brede C, Larsen JP, Moller SG.
J Biol Chem. 2013 Jun 21. [Epub ahead of print]
PMID: 23792957 [PubMed - as supplied by publisher] **Free Article**
[Related citations](#)
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Emmrich JV, Hornik TC, Neher JJ, Brown GC.
FEBS J. 2013 Jun 21. doi: 10.1111/febs.12401. [Epub ahead of print]
PMID: 23789887 [PubMed - as supplied by publisher]
[Related citations](#)
3. [Dopaminergic neurotoxicity of S-ethyl N,N-dipropylthiocarbamate \(EPTC\), molinate, and S-methyl-N,N-diethylthiocarbamate \(MeDETC\) in Caenorhabditis elegans.](#)
Caito SW, Valentine WM, Aschner M.
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PMID: 23786526 [PubMed - as supplied by publisher]
[Related citations](#)
4. [Cellular Localization of Dieldrin and Structure-Activity Relationship of Dieldrin Analogues in Dopaminergic Cells.](#)
Allen EM, Florang VR, Davenport LL, Jinsmaa Y, Doorn JA.
Chem Res Toxicol. 2013 Jun 27. [Epub ahead of print]
PMID: 23763672 [PubMed - as supplied by publisher]
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Williams CA, Lin Y, Maynard A, Cheng SY.
Cell Mol Neurobiol. 2013 Jun 7. [Epub ahead of print]
PMID: 23744253 [PubMed - as supplied by publisher]
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Pezzoli G, Cereda E.
Neurology. 2013 May 28;80(22):2035-41. doi: 10.1212/WNL.0b013e318294b3c8.

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Martins JB, Bastos Mde L, Carvalho F, Capela JP.
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Yuan YH, Sun JD, Wu MM, Hu JF, Peng SY, Chen NH.
Neurochem Res. 2013 Aug;38(8):1553-60. doi: 10.1007/s11064-013-1055-7. Epub 2013 May 7.
PMID: 23645222 [PubMed - in process]
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9. [Sodium butyrate improves locomotor impairment and early mortality in a rotenone-induced Drosophila model of Parkinson's disease.](#)
St Laurent R, O'Brien LM, Ahmad ST.
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Lee PC, Rhodes SL, Sinsheimer JS, Bronstein J, Ritz B.
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Sci Rep. 2013;3:1635. doi: 10.1038/srep01635.
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Tien LT, Kaizaki A, Pang Y, Cai Z, Bhatt AJ, Fan LW.
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Chorfa A, Bétemps D, Morignat E, Lazizzera C, Hogeveen K, Andrieu T, Baron T.
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Chung SJ, Armasu SM, Anderson KJ, Biernacka JM, Lesnick TG, Rider DN, Cunningham JM, Ahlskog JE, Frigerio R, Maraganore DM.
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Bernstein AI, O'Malley KL.
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National Water-Quality Assessment Program

**Estimation of Annual Agricultural Pesticide Use for
Counties of the Conterminous United States, 1992–2009**



Scientific Investigation Report 2013–5009

**Cotton bud and
blue sky**



Wheat crop



Growing corn



Soy bean field



Estimation of Annual Agricultural Pesticide Use for Counties of the Conterminous United States, 1992–2009

By Gail P. Thelin and Wesley W. Stone

National Water-Quality Assessment Program

Scientific Investigations Report 2013–5009

U.S. Department of the Interior
U.S. Geological Survey

U.S. Department of the Interior
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U.S. Geological Survey
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U.S. Geological Survey, Reston, Virginia: 2013

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FOREWORD

The U.S. Geological Survey (USGS) is committed to providing the Nation with reliable scientific information that helps to enhance and protect the overall quality of life and that facilitates effective management of water, biological, energy, and mineral resources (<http://www.usgs.gov/>). Information on the Nation's water resources is critical to ensuring long-term availability of water that is safe for drinking and recreation and is suitable for industry, irrigation, and fish and wildlife. Population growth and increasing demands for water make the availability of that water, measured in terms of quantity and quality, even more essential to the long-term sustainability of our communities and ecosystems.

The USGS implemented the National Water-Quality Assessment (NAWQA) Program in 1991 to support national, regional, State, and local information needs and decisions related to water-quality management and policy (<http://water.usgs.gov/nawqa>). The NAWQA Program is designed to answer: What is the quality of our Nation's streams and groundwater? How are conditions changing over time? How do natural features and human activities affect the quality of streams and groundwater, and where are those effects most pronounced? By combining information on water chemistry, physical characteristics, stream habitat, and aquatic life, the NAWQA Program aims to provide science-based insights for current and emerging water issues and priorities. From 1991 to 2001, the NAWQA Program completed interdisciplinary assessments and established a baseline understanding of water-quality conditions in 51 of the Nation's river basins and aquifers, referred to as Study Units (http://water.usgs.gov/nawqa/studies/study_units.html).

National and regional assessments are ongoing in the second decade (2001–2012) of the NAWQA Program as 42 of the 51 Study Units are selectively reassessed. These assessments extend the findings in the Study Units by determining water-quality status and trends at sites that have been consistently monitored for more than a decade, and filling critical gaps in characterizing the quality of surface water and groundwater. For example, increased emphasis has been placed on assessing the quality of source water and finished water associated with many of the Nation's largest community water systems. During the second decade, NAWQA is addressing five national priority topics that build an understanding of how natural features and human activities affect water quality, and establish links between sources of contaminants, the transport of those contaminants through the hydrologic system, and the potential effects of contaminants on humans and aquatic ecosystems. Included are studies on the fate of agricultural chemicals, effects of urbanization on stream ecosystems, bioaccumulation of mercury in stream ecosystems, effects of nutrient enrichment on aquatic ecosystems, and transport of contaminants to public-supply wells. In addition, national syntheses of information on pesticides, volatile organic compounds (VOCs), nutrients, trace elements, and aquatic ecology are continuing.

The USGS aims to disseminate credible, timely, and relevant science information to address practical and effective water-resource management and strategies that protect and restore water quality. We hope this NAWQA publication will provide you with insights and information to meet your needs, and will foster increased citizen awareness and involvement in the protection and restoration of our Nation's waters.

The USGS recognizes that a national assessment by a single program cannot address all water-resource issues of interest. External coordination at all levels is critical for cost-effective management, regulation, and conservation of our Nation's water resources. The NAWQA Program, therefore, depends on advice and information from other agencies—Federal, State, regional, interstate, Tribal, and local—as well as nongovernmental organizations, industry, academia, and other stakeholder groups. Your assistance and suggestions are greatly appreciated.

William H. Werkheiser
USGS Associate Director for Water

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Conversion Factors

Inch/Pound to SI

Multiply	By	To obtain
Area		
acre	4,047	square meter (m ²)
acre	0.4047	hectare (ha)
acre	0.4047	square hectometer (hm ²)
acre	0.004047	square kilometer (km ²)
square mile (mi ²)	259.0	hectare (ha)
square mile (mi ²)	2.590	square kilometer (km ²)
Mass		
pound, avoirdupois (lb)	0.4536	kilogram (kg)

SI to Inch/Pound

Multiply	By	To obtain
Area		
square kilometer (km ²)	247.1	acre
square kilometer (km ²)	0.3861	square mile (mi ²)

Abbreviations

ACU	Agricultural Chemical Use
CDL	Cropland Data Layer
CRD	Crop Reporting District
DPR	California Department of Pesticide Regulation
DPR-PUR	Department of Pesticide Regulation-Pesticide Use Reporting (California)
EPest	Estimated pesticide use
FR	Fruitful Rim
GIS	Geographic Information System
NASS	National Agricultural Statistics Service
NAWQA	National Water Quality Assessment Program
NPUD	National Pesticide Use Data
PUR	Pesticide Use Reporting
RE	Relative error calculated as: (EPest–NASS) /NASS
USDA	U.S. Department of Agriculture
USEPA	U.S. Environmental Protection Agency
USGS	U.S. Geological Survey
WARP	Watershed Regressions for Pesticides

Estimation of Annual Agricultural Pesticide Use for Counties of the Conterminous United States, 1992–2009

By Gail P. Thelin and Wesley W. Stone

Abstract

A method was developed to calculate annual county-level pesticide use for selected herbicides, insecticides, and fungicides applied to agricultural crops grown in the conterminous United States from 1992 through 2009. Pesticide-use data compiled by proprietary surveys of farm operations located within Crop Reporting Districts were used in conjunction with annual harvested-crop acreage reported by the U.S. Department of Agriculture National Agricultural Statistics Service (NASS) to calculate use rates per harvested-crop acre, or an 'estimated pesticide use' (EPest) rate, for each crop by year. Pesticide-use data were not available for all Crop Reporting Districts and years. When data were unavailable for a Crop Reporting District in a particular year, EPest extrapolated rates were calculated from adjoining or nearby Crop Reporting Districts to ensure that pesticide use was estimated for all counties that reported harvested-crop acreage. EPest rates were applied to county harvested-crop acreage differently to obtain EPest-low and EPest-high estimates of pesticide-use for counties and states, with the exception of use estimates for California, which were taken from annual Department of Pesticide Regulation Pesticide Use Reports.

Annual EPest-low and EPest-high use totals were compared with other published pesticide-use reports for selected pesticides, crops, and years. EPest-low and EPest-high national totals for five of seven herbicides were in close agreement with U.S. Environmental Protection Agency and National Pesticide Use Data estimates, but greater than most NASS national totals. A second set of analyses compared EPest and NASS annual state totals and state-by-crop totals for selected crops. Overall, EPest and NASS use totals were not significantly different for the majority of crop-state-year combinations evaluated. Furthermore, comparisons of EPest and NASS use estimates for most pesticides had rank correlation coefficients greater than 0.75 and median relative errors of less than 15 percent. Of the 48 pesticide-by-crop combinations with 10 or more state-year combinations, 12 of the EPest-low and 17 of the EPest-high totals showed significant differences ($p < 0.05$) from NASS use estimates. The differences between EPest and NASS estimates did not follow consistent patterns related to particular crops, years,

or states, and most correlation coefficients were greater than 0.75.

EPest values from this study are suitable for making national, regional, and watershed assessments of annual pesticide use from 1992 to 2009. Although estimates are provided by county to facilitate estimation of watershed pesticide use for a wide variety of watersheds, there is a greater degree of uncertainty in individual county-level estimates when compared to Crop Reporting District or state-level estimates because (1) EPest crop-use rates were developed on the basis of pesticide use on harvested acres in multi-county areas (Crop Reporting Districts) and then allocated to county harvested cropland; (2) pesticide-by-crop use rates were not available for all Crop Reporting Districts in the conterminous United States, and extrapolation methods were used to estimate pesticide use for some counties; and (3) it is possible that surveyed pesticide-by-crop use rates do not reflect all agricultural use on all crops grown. The methods developed in this study also are applicable to other agricultural pesticides and years.

Introduction

Hundreds of millions of pounds of pesticides are applied to agricultural crops every year to control weeds, insect infestations, plant diseases, and other pests. Annually, the total amount of conventional pesticides (excluding sulfur, petroleum oil, chlorine, hypochlorites, and wood preservatives) applied to crops grown throughout the conterminous United States has increased from a low of about 698 million pounds in the early 1990s (http://www.epa.gov/opp00001/pestsales/07pestsales/historical_data2007_3.htm#table5_6, accessed November 16, 2011) to a high of over 800 million pounds in 1996 (*fig. 1*). From 1996 through 2007, there was a slight downward trend in the total amount of pesticides used, reflecting decreases in the use of herbicides, plant growth regulators, and other conventional pesticides. Most of these differences in pesticide use can be attributed to changes in crop-management practices, the development of new pesticides that are effective at reduced use rates, and the introduction of genetically modified crops (Young, 2006; Fernandez-Cornejo and McBride, 2000).

2 Estimation of Annual Agricultural Pesticide Use for Counties of the Conterminous United States, 1992–2009

Pesticides are important to crop management because they contribute to increased crop yields and improve the quality of crops. Pesticides applied to crops and soil, however, can be transported to surface water and groundwater, where they can degrade water quality. Pesticide concentrations in streams vary widely across the United States and are influenced by many factors, such as the amount and timing of pesticide applications and the soils, climate, and hydrology where they are applied (Gilliom and others, 2006). Nationally consistent information on the amount and geographic distribution of pesticide use, both current and historic, is essential for designing water-quality studies, interpreting water-quality data, assessing trends in pesticide use, and developing water-quality models that relate pesticide use to concentrations in the hydrologic environment.

Agricultural pesticide-use information is available from the U.S. Department of Agriculture (USDA) National Agricultural Statistics Service (NASS), but these data are reported as state totals for varying regions, crops, and years and, consequently, do not have sufficient geographic coverage, resolution, or temporal consistency to support studies at watershed or multicounty scales. California’s Department of Pesticide Regulation (DPR) collects detailed pesticide-use information from all licensed applicators in the State and publishes annual Pesticide Use Reports (DPR-PURs)

that include detailed pesticide-use information (California Department of Pesticide Regulation, 2010). Agricultural pesticide-use data also are available from proprietary sources, but extrapolation techniques, such as those described in this report, are needed so that these data can be used by the National Water Quality Assessment (NAWQA) Program to estimate pesticide use for all counties of the conterminous United States.

A previous U.S. Geological Survey (USGS) study focused on developing extrapolation methods to determine county-level estimates for the herbicide atrazine by using proprietary pesticide-use reports and county harvested-crop acreage (Thelin and Stone, 2010). As part of that approach, regional rates were developed by using data from multiple years, and atrazine estimates were calculated for most counties in the conterminous United States. Comparisons with other data sources indicated that this approach to regional extrapolation could over-estimate pesticide use for pesticides that are not widely used across all geographic regions or when pesticide-use practices changed. This report describes an approach to estimating pesticide use, referred to as EPest, that is based on previous efforts but has changes that limit the use of regional rates, that incorporate a refined version of crop growing regions, and that expand the method to 39 herbicides, insecticides, and fungicides used in agriculture (*table 1*).

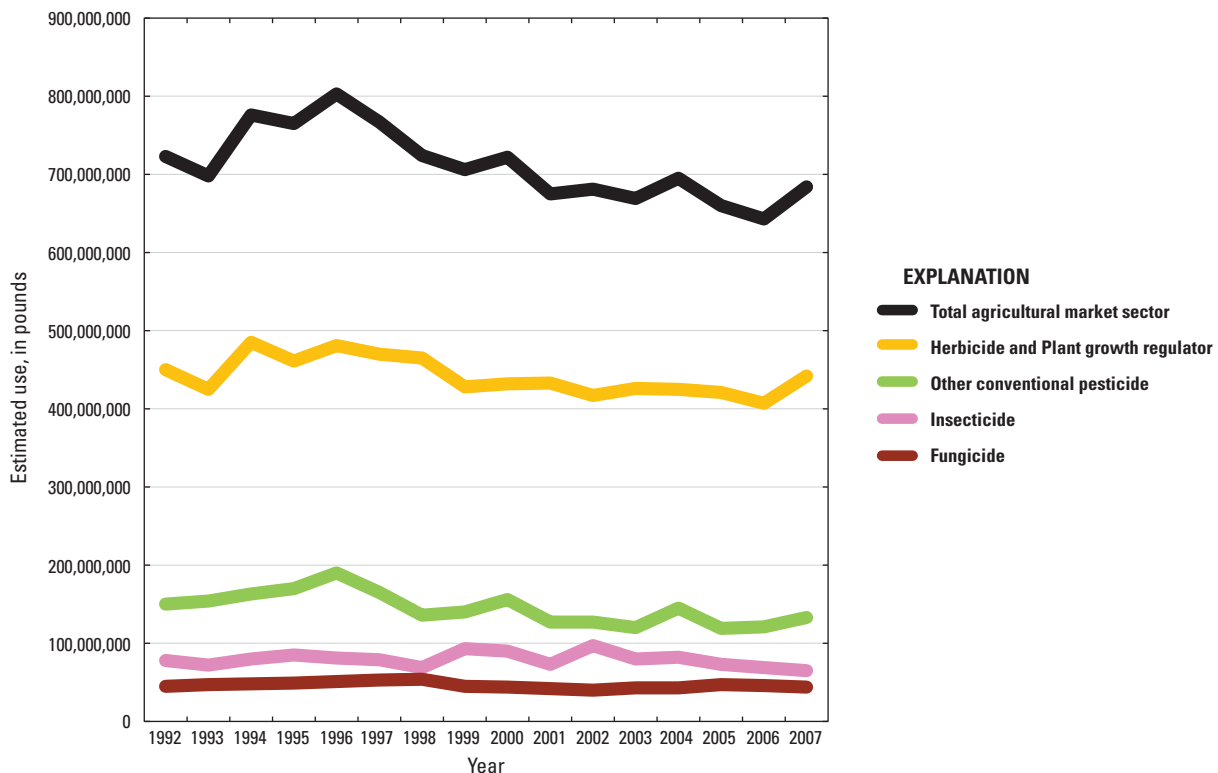


Figure 1. Trends in agricultural conventional pesticide use in the conterminous United States, 1992–2009.

Table 1. List of pesticide names and type, for which annual county pesticide-use estimates were calculated.

Pesticide name	Type
Acetochlor	Herbicide
Acifluorfen	Herbicide
Alachlor	Herbicide
Atrazine	Herbicide
Benomyl	Fungicide
Bentazon	Herbicide
Bromoxynil	Herbicide
Butylate	Herbicide
Carbofuran	Insecticide
Chlorimuron	Herbicide
Cyanazine	Herbicide
EPTC	Herbicide
Ethalfuralin	Herbicide
Ethoprophos	Insecticide
Fluometuron	Herbicide
Fonofos	Insecticide
Glyphosate	Herbicide
Linuron	Herbicide
Methomyl	Insecticide
Methyl parathion	Insecticide
Metolachlor	Herbicide
S-metolachlor	Herbicide
Metribuzin	Herbicide
Nicosulfuron	Herbicide
Norflurazon	Herbicide
Oryzalin	Herbicide
Oxamyl	Insecticide
Pebulate	Herbicide
Phorate	Insecticide
Propachlor	Herbicide
Propanil	Herbicide
Propargite	Insecticide
Propiconazole	Fungicide
Propyzamide	Herbicide
Terbacil	Herbicide
Terbufos	Insecticide
Thiobencarb	Herbicide
Triallate	Herbicide
Trifluralin	Herbicide

Purpose and Scope

The purpose of this report is to describe (1) a method to estimate annual pesticide-by-crop use rates (pounds applied per harvested-crop acre), referred to as EPest rates, for 39 pesticides; (2) the process that was followed to apply these rates to produce an EPest-low and EPest-high estimate of annual use for each county; and (3) how the estimates for selected pesticides and crops derived by these methods compare with estimates from other published sources. This method was developed by using pesticide-use estimates reported for Crop Reporting Districts (CRDs) to calculate annual pesticide-by-crop use rates and, from that, estimates of pesticide use for individual counties. The 39 selected pesticides represent some of the primary pesticides used throughout the nation on row crops and several orchard and

vegetable crops, and include 28 herbicides, 9 insecticides, and 2 fungicides. Most of these same pesticides were included in a Watershed Regressions for Pesticides (WARP) multi-compound model analysis (Charles Crawford, U.S. Geological Survey, oral commun., 2011).

The pesticides evaluated in this study represent a range of herbicides, insecticides, and fungicides that are used on a variety of row, fruit, nut, and specialty crops grown in different environmental settings. Several of these pesticides have had changes in use over time, providing an evaluation of method performance for a wide range of conditions. To assess the accuracy of EPest totals, state-level totals were compared with NASS use estimates for selected pesticides and crops for states and years for which NASS survey data were available.

Data Sources

Data sources used to develop EPest pesticide-by-crop use rates and annual pesticide-use estimates by county included the following: (1) proprietary pesticide-by-crop use estimates reported for CRDs; (2) USDA county harvested-crop acreage reported in the 1992, 1997, 2002, and 2007 Census of Agriculture (<http://www.agcensus.usda.gov/>), and NASS annual harvested-crop acreage data collected from crop surveys for non-census years (<http://quickstats.nass.usda.gov/>); (3) boundaries for CRDs and counties; (4) regional boundaries derived from USDA Farm Resource Regions; and (5) pesticide-use information from California DPR-PUR. Each of these sources is described in following sections.

Pesticide-Use Data

Proprietary data from GfK Kynetec, Inc. on the amounts of pesticides applied to individual crops by CRD are the primary source of information used in this study and are referred to as surveyed use data in the remainder of this report. The surveyed use data are based on agricultural pesticide use surveys of more than 20,000 farm operations distributed throughout the conterminous United States (AgroTrak Quality Management Plan, written commun., August 2011). Data from the Census of Agriculture on the size (in acres) and number of farms that grow individual crops and represent selected land uses, such as pasture, are used to stratify all farms in the United States by size and to allocate the number of farms that will be surveyed in each strata. The survey design allocates a greater proportion of the sample to larger farm operations so that a greater percentage of crop acreage is represented, with the goal of more accurate characterization of farm operations and pesticide-use patterns. Use estimates for over 400 pesticides that are applied to a variety of row, specialty, fruit, and nut crops are reported by multi-county areas, referred to as CRDs (*fig. 2*). Surveys of farm operations within each CRD are extrapolated to represent total pesticide use for that CRD, and then estimates for individual CRDs or groups of CRDs are expanded to estimate pesticide use for states.

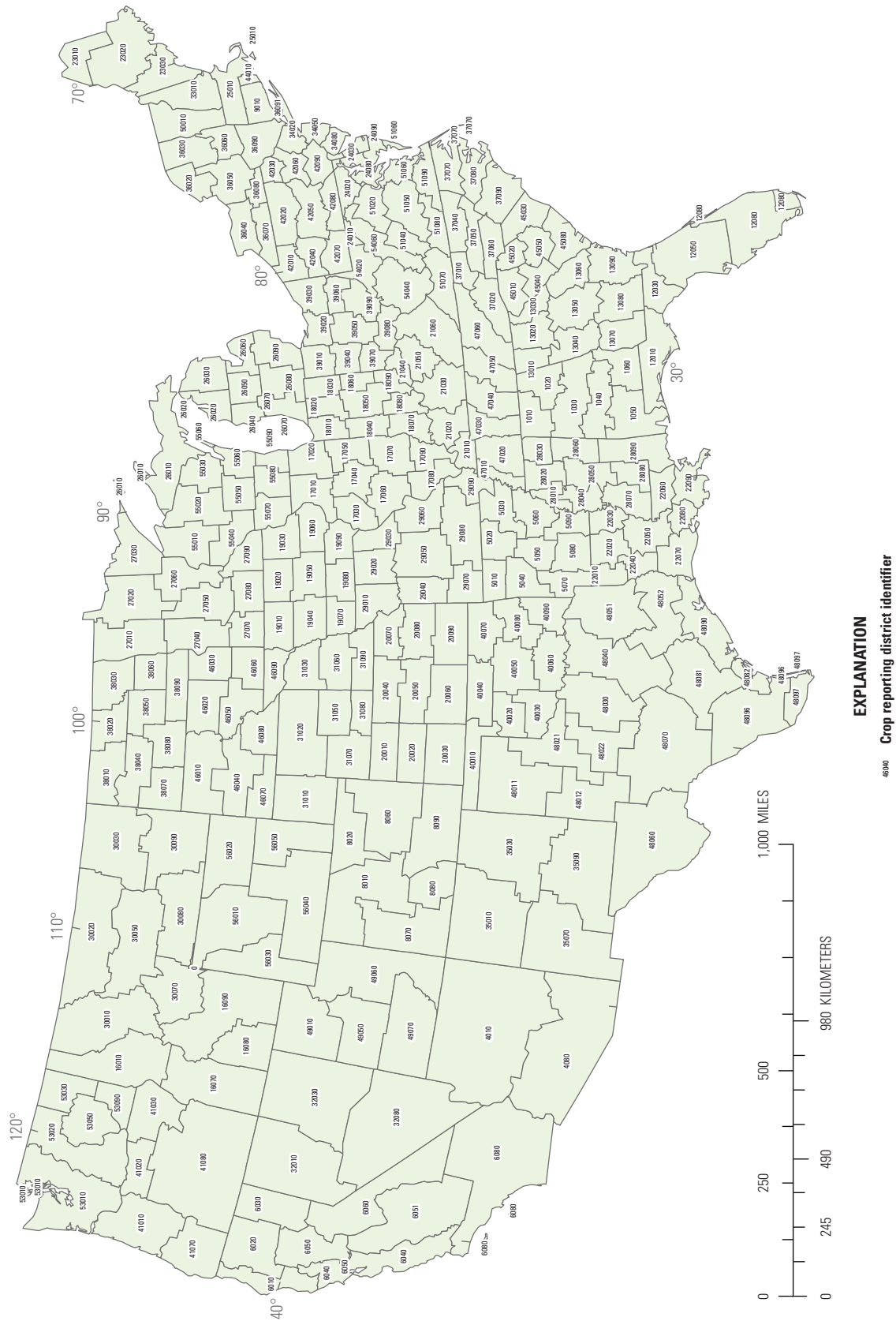


Figure 2. Crop Reporting Districts of the conterminous United States.

Harvested-Crop Acreage

The surveyed use data are based on planted-crop acres within a CRD, but NAWQA requires pesticide-use estimates at the county scale, including use estimates for pesticides that potentially were not surveyed. Therefore, the surveyed use data had to be disaggregated from CRDs to the individual counties. The USDA is the only uniform source of annual crop-acreage estimates for all counties in the United States. The USDA reports data on planted and harvested-crop acreage, but planted-acreage data are not available from the USDA for all of the individual crops with surveyed use data. Therefore, harvested acreage, rather than planted acreage, was used to develop annual pesticide-by-crop use rates. In taking this approach, it is recognized that use-rate estimates could be numerically greater than actual use rates on planted crops because not all planted acres are harvested. The emphasis of the method was to develop the best possible estimates of total use in a county, which required the use of the comprehensive data on harvested cropland. Annual harvested-crop acreage by county data from the USDA Census of Agriculture and NASS crop surveys were used in method development (1) to calculate the pesticide-by-crop use rates for each crop and CRD surveyed, and (2) to estimate pesticide use for all counties that report harvested acreage in the conterminous United States. Harvested-crop acreage was obtained from the Census of Agriculture for 1992, 1997, 2002, and 2007, and from NASS annual surveys for the years between censuses. *Table 2* lists the crops for which EPest use rates were developed and the USDA crop names for which acreage data were retrieved from the Census of Agriculture and NASS.

County-level harvested-crop acreage for the 76 crops and other non-crop agricultural-land uses, such as pasture and woodland, were obtained from USDA reports and used to produce harvested-crop acreage totals for all CRDs. However, additional processing was required in three cases: (1) the USDA did not report county acreage for a crop and year because of census nondisclosure rules that protect the identity of individual farm operations, (2) the USDA-NASS annual surveys did not collect data for a particular state or crop, or (3) the crop acreage was the total acreage for multiple categories of that crop. In cases when county acreage was not reported because of USDA nondisclosure rules or when a crop and state had not been surveyed by NASS, the county crop acreage was estimated through linear interpolation of acreage reports for the crop and county from consecutive years before and after the year of missing crop acreage. In order to produce acreage totals for EPest crop names that were composed of more than one USDA crop name, the subcategories for that crop were summed to produce total harvested acreage. For example, the county total for sorghum acreage was calculated by summing the acreage for the subcategories of sorghum:

sorghum for grain, sorghum for silage, and sorghum for syrup. Crop-acreage totals that comprised more than one crop name typically required crop acreage to be estimated through linear interpolation for some of the crop names because NASS crop surveys do not report all the same crop names as the Census of Agriculture. For example, NASS did not report acreage of corn for forage from 1992 through 2009. To estimate corn-for-forage acreage in non-census years, the acreage from two Censuses of Agriculture (prior and next) was interpolated to fill in the non-surveyed corn-for-forage acreage.

Geospatial Data

Two geospatial datasets were integral to the method used to calculate pesticide-by-crop use rates for surveyed and non-surveyed CRDs. These datasets included boundaries for CRDs and USDA Farm Resource Regions (<http://www.ers.usda.gov/Briefing/ARMS/resourceregions/resourceregions.htm>). CRD boundaries were used (1) to develop a table that listed the spatial relation of each CRD in the conterminous United States to its surrounding CRDs and (2) to determine the counties that were associated with each CRD so that estimates reported for CRDs could be disaggregated to counties. The second geospatial dataset was a modified version of the USDA Farm Resource Boundaries, which was used (1) to determine the Farm Resource Region for each CRD and (2) to develop regional use rates for individual crops when a CRD rate did not exist.

CRDs are defined as multi-county areas that share similar geographic attributes, including soil type, terrain, elevation, and climatic factors, such as mean temperature, annual precipitation, and length of growing season. There are 304 CRDs in the conterminous United States, and most states are divided into 9 CRDs; however, some states, such as Massachusetts and New Hampshire, contain only 1 CRD, whereas Texas has 15 CRDs.

A geospatial vector dataset of CRD boundaries was used to generate a table that enumerates the spatial relation between each of the individual CRDs and the CRDs surrounding each of these 'primary' CRDs. For each primary CRD, two concentric rings of CRDs were identified by using a Geographic Information System (GIS) proximity mapping function. CRDs that touched the primary CRD were designated as tier 1 CRDs, and CRDs that touched tier 1 CRDs were designated as tier 2 CRDs. Any CRD could be considered a primary, a tier 1, or a tier 2 CRD, depending on which CRD is central to the area of interest. *Figure 3*, for example, shows primary CRD 20060 (Kansas CRD 60) and the tier 1 and tier 2 CRDs that are associated with it. When CRD-level pesticide use data were not available, associated tier 1 and tier 2 CRDs were used to calculate pesticide-by-crop rates.

6 Estimation of Annual Agricultural Pesticide Use for Counties of the Conterminous United States, 1992–2009

Table 2. EPest crop name and corresponding U.S. Department of Agriculture (USDA) Census of Agriculture crop names.

EPest crop name	USDA, Census of Agriculture crop name(s)
Alfalfa	Alfalfa hay
Almonds	Almonds
Apples	Apples
Barley	Barley for grain
Beans and peas	Green lima beans; snap beans; green peas, excluding southern peas; peas, green southern
Berries	Strawberries
Bulb crops	Garlic; green onions; dry onions
Conservation Reserve Program (CRP), long-term acres	Land enrolled in conservation reserve or wetlands reserve programs
Canola, rapeseed	Canola, other rapeseed
Cherries	Sweet cherries; tart cherries
Citrus, other	Other citrus fruit
Cole crops	Broccoli
Corn	Corn for grain
Cotton	Cotton, all
Cropland for pasture	Cropland used for pasture or grazing
Cucurbits	Cucumbers and pickles; pumpkins; squash
Dry beans and peas	Dry lima beans; dry edible beans, excluding limas; dry edible peas; dry southern peas
Eggplant and peppers	Eggplant; peppers, bell; peppers, chile
Summer fallow	Summer fallow
Flax	Flaxseed
Grapefruit	Grapefruit
Grapevines	Grapes
Hay, other	Grass silage, haylage
Idle cropland, other	Idle cropland, other
Leafy vegetables, other	Celery; spinach
Lemons	Lemons
Lettuce	Lettuce all
Lots, farmsteads, other	Lots, farmsteads and other
Melons	Cantaloupes; watermelons
Nut trees, other	Hazel nuts (filberts); pistachios
Oats and rye	Oats for grain; rye for grain
Oranges	Oranges, all
Pasture/range	Pastureland and rangeland, other than cropland and woodland pastured
Peaches	Peaches, all
Peanuts	Peanuts for nuts
Pears	Pears, all
Pecans	Pecans
Potatoes	Potatoes
Prunes	Plums and prunes
Rice	Rice
Roots and tubers	Carrots
Sorghum	Sorghum for grain; sorghum for silage or green chop; sorghum for syrup
Soybeans	Soybeans for beans
Stone-like fruit, other	Apricots; avocados
Sugarbeets	Sugar beets for sugar
Sugarcane	Sugar cane for sugar
Sunflowers	Sunflower seed all
Sweet corn	Sweet corn
Tobacco	Tobacco
Tomatoes	Tomatoes
Other vegetables	Artichokes
Walnuts	Walnuts, english
Wheat, spring	Durum wheat for grain; other spring wheat for grain
Wheat, winter	Winter wheat for grain
Woodland	Total woodland

A geospatial dataset of USDA Farm Resource Regions was used to develop regional pesticide-by-crop use rates for CRDs that were not surveyed and for which a tier 1 or tier 2 rate was not available. In a previous atrazine study (Thelin and Stone, 2010), USDA Farm Production Regions were used to develop regional rates. These boundaries follow state boundaries and often combine large areas that can have different soils, topography, and agricultural practices. The Farm Production Region boundaries were replaced with USDA Farm Resource Regions because these boundaries take into account farm practices and physiographic, soil, and climatic traits (<http://www.ers.usda.gov/publications/aib760/aib-760.pdf>). Farm Resource Region boundaries conform to CRD boundaries. There are nine Farm Resource Regions, which were further subdivided in cases where the region was not contiguous. For example, the Fruitful Rim (FR) Region is located in parts of the West, Southwest, and Southeastern United States, so this large region was subdivided into four subregions: (1) FR-Northwest, including Washington and parts of Oregon and Idaho; (2) FR-West, including parts of California and Arizona; (3) FR-Texas, including Texas and

New Mexico; and (4) FR-Southeast, including Florida and parts of Alabama, Georgia, and South Carolina. Similarly, the Eastern Uplands, Northern Crescent, and Southern Seaboard were divided into eastern and western subregions (fig. 4).

Pesticide-Use Estimates for California

EPest-low and EPest-high estimates for California were not calculated by using the method described in this report; instead, county totals were obtained from the online DPR-PUR database (California Department of Pesticide Regulation, 2010). Since 1990, California has required reporting of all agricultural pesticide use. DPR-PUR includes information on the pesticide applied, location and time of application, and the agricultural crop treated. Annual pesticide-use estimates by crop were retrieved from the online DPR-PUR database and merged with the EPest-low and EPest-high county data after the estimation process was completed for the rest of the country.

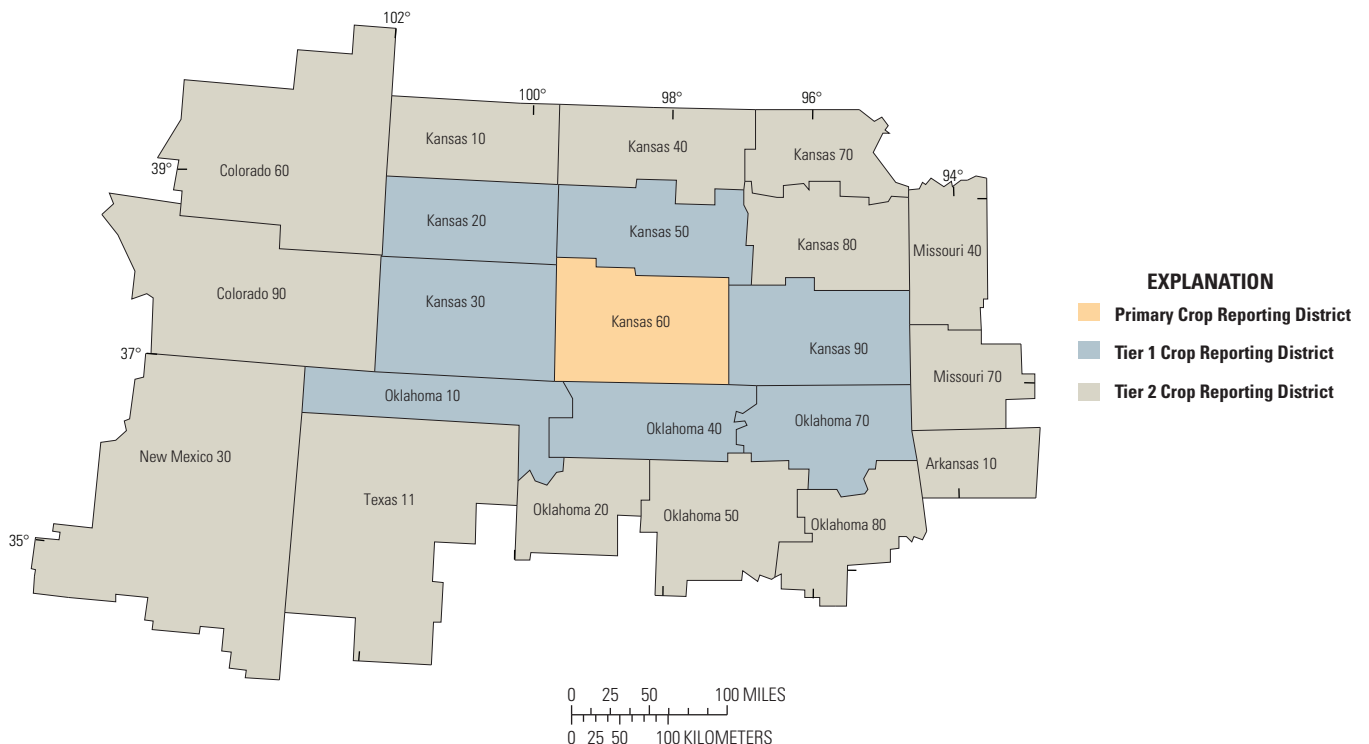


Figure 3. Crop Reporting District 20060 (Kansas CRD 60) and neighboring tier 1 and tier 2 Crop Reporting Districts.

Methods for Estimating Pesticide Use

The following sections describe methods developed to estimate agricultural pesticide use for counties in the conterminous United States, except those in California. In order to calculate estimates of pesticide use for counties, pesticide-by-crop use rates were developed for CRDs on the basis of surveyed use data and harvested-crop acreage from the USDA. The resulting pesticide-by-crop use rates are referred to as EPest surveyed-use rates, which are calculated by dividing the amount of pesticide applied to a crop in the CRD by harvested-crop acres. Not every CRD in the conterminous United States was surveyed; therefore, EPest

extrapolated rates were developed for unsurveyed CRDs by using surveyed rates from nearby CRDs or surveyed and extrapolated rates from CRDs in the same region. A surveyed or an extrapolated rate, depending on the CRD, was applied to county harvested acreage to estimate pesticide use on individual crops grown in each county of the conterminous United States, except California. The following sections describe (1) the method used to replace false zero values reported in the surveyed use data with inferred data, (2) how the EPest surveyed and extrapolated rates were developed, and (3) the decision process that was followed to assign these EPest rates to counties to produce EPest-high and EPest-low estimates of pesticide use for counties in the conterminous United States.

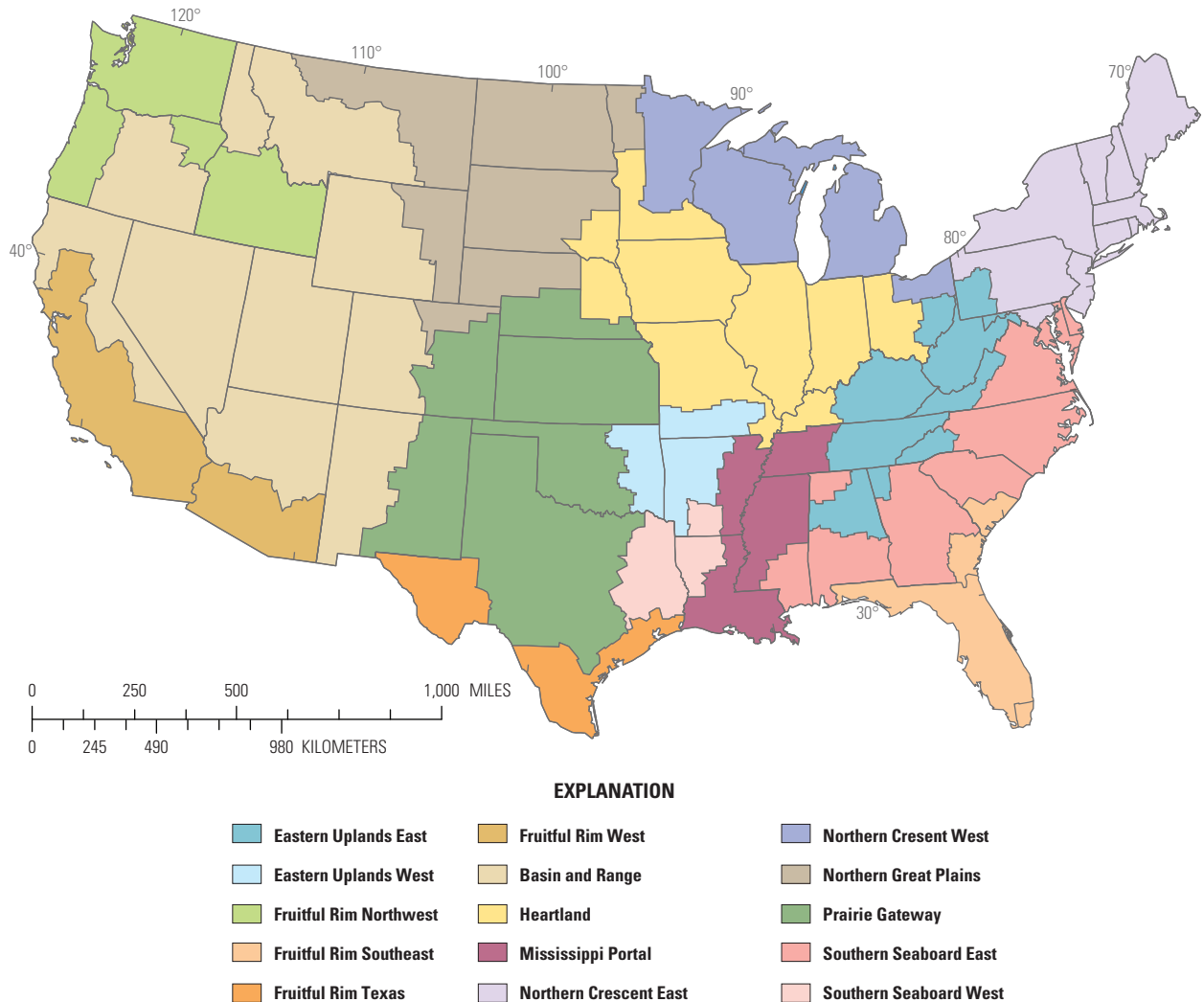


Figure 4. U.S. Department of Agriculture Farm Resources Regions (<http://www.ers.usda.gov/publications/aib760/aib-760.pdf>), as subdivided for calculating regional estimated pesticide-use rates.

Processing Zero Values

The surveyed-use data included the following elements: pounds of pesticide applied to a crop, number of crop acres treated, and overall pesticide-by-crop application rate. In some cases, a zero value was reported for one or more of the data elements because of rounding or truncating values of less than one; therefore, a new inferred value was calculated to replace the false zero values as follows:

1. When the pounds applied were reported as zero, but the number of acres treated was greater than zero, and an application rate was reported, then a value for the pounds applied was calculated by multiplying the number of acres treated by the pesticide-by-crop application rate reported for the surveyed CRD.
2. When the number of acres treated and the pounds applied were reported as zero for the surveyed CRD, but an application rate was reported, then it was assumed that the number of acres treated was equal to one, and the pounds applied were equal to the application rate for 1 acre as reported for the CRD.
3. When the pounds applied and application rate were reported as zero for the surveyed CRD, but the number of acres treated was greater than zero, a new application rate could not be calculated. In these cases, the lowest non-zero application rate in the surveyed-use data across all years, pesticides, crops, and CRDs, which was 0.001 pounds per acre annually, was used to estimate the pounds applied (0.001 pounds per acre multiplied by the number of acres treated).

EPest Crop-Use Rates for Surveyed CRDs

EPest surveyed rates for 1992 through 2009 were developed for each of the 39 pesticides included in this study by using surveyed-use estimates of pounds of pesticides applied to individual crops and the harvested acreage for these crops reported by USDA. The pesticide-by-crop use rates determined from surveyed-use data for CRDs are based on planted-crop acreage, but were adjusted to harvested acreage for EPest county-level pesticide-by-crop use rates. EPest surveyed pesticide-by-crop use rates were calculated by dividing the pounds of pesticide applied to a crop in a CRD by the harvested-crop acreage in the CRD to yield a use rate per harvested acre—for a specific crop this is referred to as an EPest surveyed pesticide-by-crop use rate. Use rates calculated by using harvested-crop acreage rather than planted acreage can result in a greater rate per acre because, typically, there are fewer harvested acres than planted acres as a result of crop failure. To avoid artificially high use rates caused solely by the difference between planted and harvested acres, the harvested-crop acreage for the CRD and associated counties was adjusted if the CRD harvested-crop acres were less than

the surveyed CRD planted-crop acres. Specifically, a county-CRD weighting factor for each crop and year was calculated by determining the percentage that each county's acreage contributed to the total acreage in the CRD. When the sum of the harvested-crop acreage for counties in the CRD was less than the planted-crop acreage for the CRD reported in the surveyed-use data, the weighting factor was used to adjust the harvested acreage for each county in the CRD to the survey-reported planted-crop acreage.

EPest Use Rates for Unsurveyed CRDs—Tier 1, Tier 2, and Regional Use Rates

EPest surveyed-use rates were applied to the harvested-crop acreage in all counties that were part of the surveyed CRDs. Some CRDs, however, were not surveyed for a particular year or combination of years, even though a pesticide could have been used there. For these CRDs, indirect estimates were derived. To ensure that pesticide-use estimates accounted for all acreage that could have been treated, extrapolated use rates were developed for individual pesticides and crops in unsurveyed CRDs through a set of decision rules (*fig. 5*).

The decision process included developing three types of extrapolated pesticide-by-crop use rates, referred to as tier 1, tier 2, and regional rates. How a use rate was estimated for an unsurveyed CRD depended on the availability of rates from surrounding tier 1 and tier 2 CRDs. For this purpose, the proximity table of CRDs, described previously, was searched to determine if a new rate could be calculated on the basis of rates from tier 1 or tier 2 CRDs. First, the tier 1 CRDs surrounding the unsurveyed CRD were searched, and if one or more surveyed pesticide-by-crop use rates existed, the median rate was used from these surveyed rates, called tier 1 EPest rate, to estimate pesticide-by-crop use for the counties in the unsurveyed CRD. If a tier 1 rate could not be established because there were no surveyed rates available, then tier 2 CRDs were searched to determine if three or more of the tier 2 CRDs had surveyed rates. If so, then the median value of these rates was used as the tier 2 EPest rate which was then applied to the counties in the unsurveyed CRD. Finally, if a tier 1 or tier 2 EPest rate could not be determined, then a regional rate was calculated for the modified USDA Farm Resource Region (described previously) and used for the CRD. Regional rates were the median of all non-zero EPest rates, including surveyed, tier 1, and tier 2 EPest from the same modified USDA Farm Resource Region. To reduce the influence of duplicate extrapolated EPest rates on the calculation of regional rates, duplicate extrapolated rates were removed prior to the calculation. *Figure 6* illustrates the process of establishing and assigning EPest extrapolated rates for counties in the Southern Seaboard Region-East by using *S-metolachlor* on corn as an example.

The Southern Seaboard-East region is composed of 36 CRDs from all or part of 8 states, including Alabama, Delaware, Georgia, Maryland, Mississippi, North Carolina, South Carolina, and Virginia (fig. 6). In 2007, there were surveyed-use data for *S*-metolachlor on corn in 17 of the 36 CRDs in the region. On the basis of the surveyed rates for the 17 surveyed CRDs, *S*-metolachlor use on corn was estimated for 180 of 388 counties in the region. There were an additional 208 counties in the region that had corn acreage, but a surveyed rate was not available, so EPest tier 1, tier 2, or regional rates were estimated as described in the following paragraphs.

Tier 1 *S*-metolachlor-corn rates were estimated for 11 CRDs in the example region and applied to 114 counties in these CRDs. South Carolina CRD 45030, labeled *A* in figure 6, is used to illustrate how a tier 1 rate is calculated from adjacent tier 1 CRDs. The tier 1 rate was developed for South Carolina CRD 45030 by using surveyed rates from three surrounding CRDs, which had EPest surveyed rates of 0.0095, 0.7093, and 1.123 pounds per harvested acre. There were two other CRDs adjacent to South Carolina 45030, but there were no surveyed rates available for them. In this example, the median of the three available EPest surveyed rates was 0.7093 pounds per harvested acre (North Carolina CRD 37090), and this rate was used as the tier 1 rate to estimate 2007 *S*-metolachlor use on corn in the nine counties that are part of South Carolina CRD 45030.

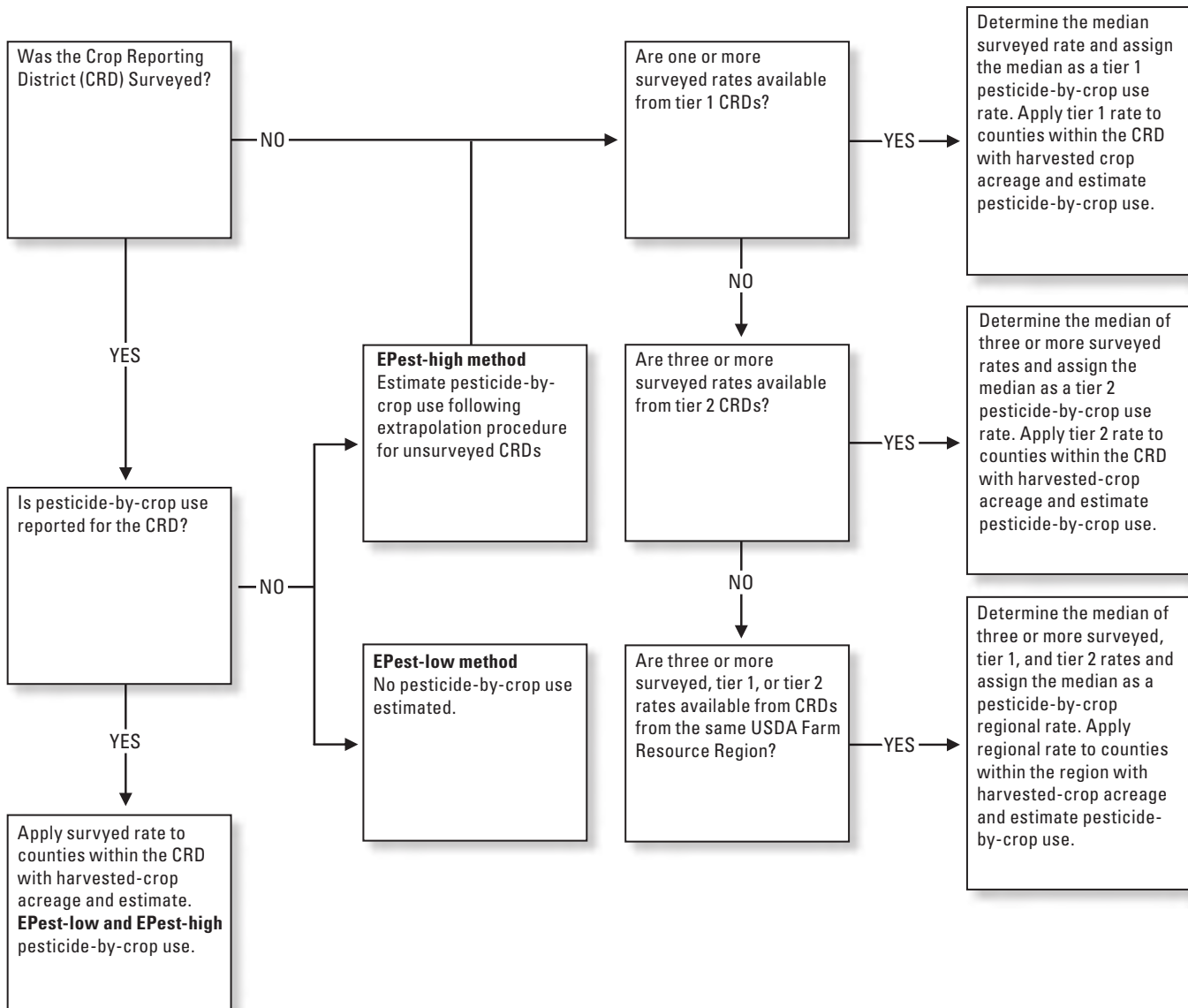


Figure 5. Summary of decision process followed to develop EPest rates.

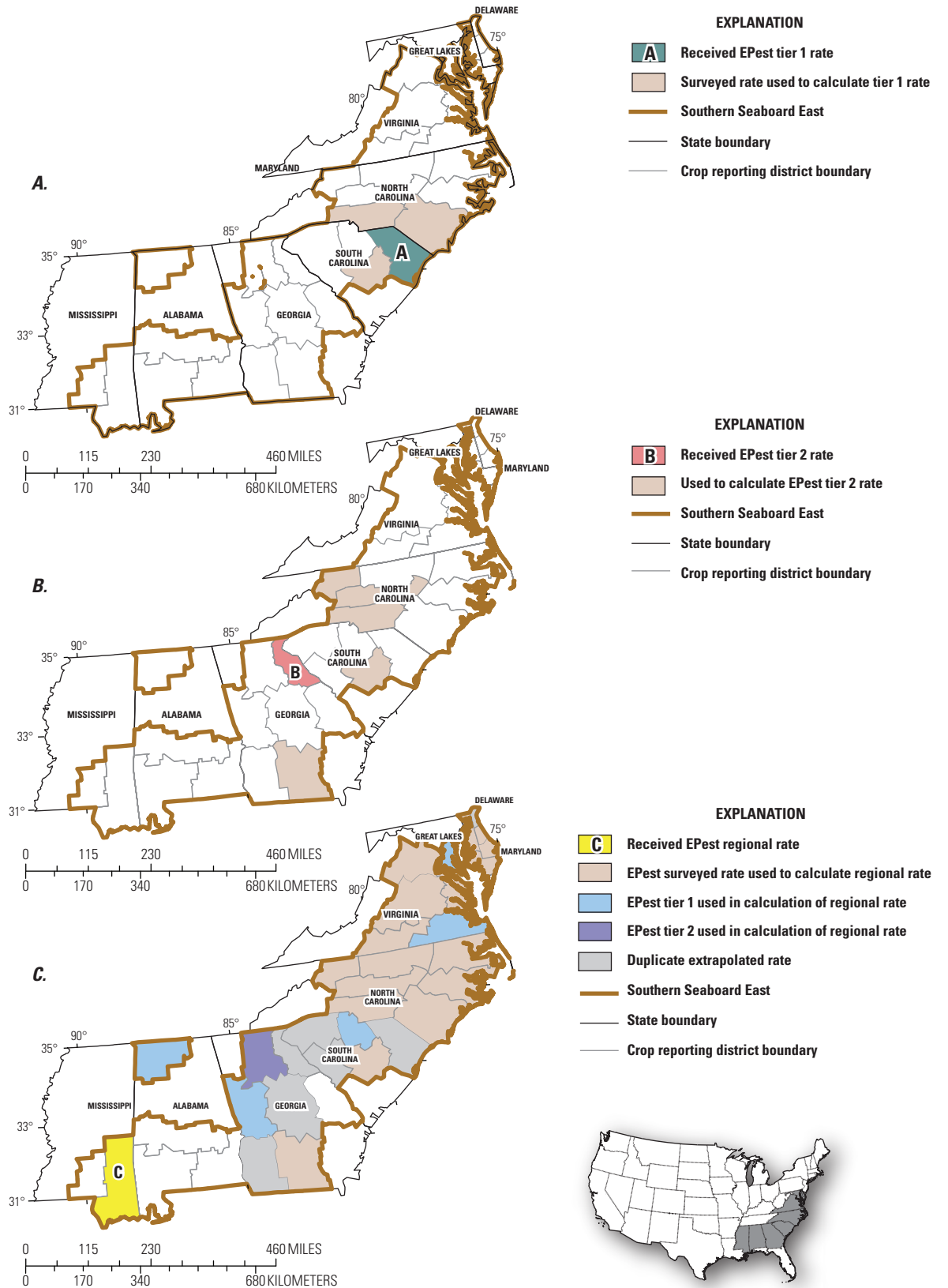


Figure 6. Methods for establishing extrapolated estimates for 2007 S-metolachlor use on corn in the Southern Seaboard-East region for (A) Epest tier 1 rate, (B) Epest tier 2 rate, and (C) Epest regional rate.

In the Southern Seaboard-East region, tier 2 *S*-metolachlor rates for corn were applied to 25 counties in two CRDs. Georgia CRD 13030, labeled *B* in *figure 6*, is an example of determining a tier 2 rate from surrounding CRDs. There were no EPest surveyed rates for *S*-metolachlor-corn from adjacent CRDs, so tier 2 CRDs were used. A minimum of three rates are required to determine a tier 2 rate, and there were five tier 2 CRDs that had surveyed annual rates of 0.1445, 0.3156, 0.7009, 0.9565, and 1.1229 pounds per harvested acre. The median of these five rates was 0.7009 pounds per harvested acre, which was assigned as the tier 2 rate used to estimate 2007 *S*-metolachlor use on corn for the nine counties in Georgia CRD 13030.

Finally, regional rates were calculated for 2007 *S*-metolachlor-corn in the Southern Seaboard-East region and applied to 6 CRDs and 69 counties. Mississippi CRD 28009, labeled *C* in *figure 6*, is used to illustrate how the regional rate was calculated from adjacent surveyed, tier 1, and tier 2 CRDs. There were 30 EPest rates available for the region, including 17 surveyed rates, 11 tier 1 rates, and 2 tier 2 rates. In the calculation of a regional rate, a minimum of three surveyed, tier 1, or tier 2 rates are required, and any duplicate extrapolated rates are dropped prior to calculating the median. In calculating the median regional rate, 7 duplicate rates were dropped, including 6 tier 1 rates and 1 tier 2 rate, so that 17 surveyed rates, 5 tier 1 rates, and 1 tier 2 rate were used to find the 2007 median rate of 0.3069 pounds per harvested acre of corn.

EPest-Low and EPest-High Estimates

Two variations on the method for estimating county pesticide use were developed to yield EPest-low and EPest-high estimates for counties in the conterminous United States other than California. Both methods incorporated surveyed and extrapolated rates to estimate pesticide use for counties, but EPest-low and EPest-high estimations differed in how they treated situations when a CRD was surveyed and pesticide use was not reported for a particular pesticide-by-crop combination (*fig. 5*). If use of a pesticide on a crop was not reported in a surveyed CRD, EPest-low reports zero use in the CRD for that pesticide-by-crop combination. EPest-high, however, treats the unreported use for that pesticide-by-crop combination in the CRD as unsurveyed, and pesticide-by-crop use rates from neighboring CRDs and, in some cases, CRDs within the same USDA Farm Resource Region are used to calculate the pesticide-by-crop EPest-high rate for the CRD.

Results

EPest-low and EPest-high totals were calculated from 1992 through 2009 for the 39 selected pesticides by using the methods described in this report. EPest-low totals, including California, were available for a low of 3,021 counties in 2008 to a high of 3,056 counties in 1992. The EPest-high method produced estimates for 3,049 counties in 2000 and 3,060 counties in 1994, including those in California. Pesticide-use estimates for counties in California are available from 1992 through 2009 for 35 of the 39 pesticides in this study. Use estimates are not available for the pesticides acetochlor, chlorimuron, propachlor, and terbufos because these pesticides were not used in California. For counties in California, there is a single county estimate, rather than a high and low estimate per pesticide by crop and year, which represents the sum of individual pesticide applications in a county reported by DPR-PUR (ftp://pestreg.cdpr.ca.gov/pub/outgoing/pur_archives).

EPest-low and EPest-high county pesticide-use totals for 1992–2009 are available from <http://water.usgs.gov/nawqa/pnsp/usage/maps/>. The county estimates represent the sum of individual pesticides used on all row, fruit, nut, and vegetable crops and selected agricultural land uses, such as summer fallow, pasture, and woodland. *Appendix 1* provides the annual EPest-low and EPest-high national totals for each of the 39 pesticides, the total pounds applied to individual crops, and the percentage of the national pesticide total each crop represents. With the exception of acetochlor, fonofos, propachlor, and *S*-metolachlor, annual estimates are available for 1992 through 2009. Acetochlor estimates are available beginning in 1994, when it was first registered for use, while estimates for fonofos and propachlor are reported for 1992 through 2005, and *S*-metolachlor estimates are available beginning in 1997.

EPest-low and EPest-high national use totals for each of the 39 pesticides are shown in *appendix 2* along with the amount and percentage of the total estimate that was derived from EPest surveyed, tier 1, tier 2, and regional rates, and from the DPR-PUR for California. Across all pesticides and years, the amount added to the EPest-low national total by extrapolated tier 1, tier 2, or regional rates, ranged from less than 1 percent for most compounds for one or more years to as much as 36 percent for terbacil use in 2003. A greater proportion of the EPest-high national total was derived from extrapolated rates, which ranged from less than 1 percent to as much as 94 percent for butylate use in 2007.

About 23 percent of the EPest-low and EPest-high annual national use totals were within 10 percent of one another and about 45 percent were within 25 percent of one another. EPest-high totals were more than double EPest-low totals for the pesticides alachlor, butylate, carbofuran, cyanazine, ethoprophos, linuron, methyl parathion, metolachlor, pebulate, propachlor, and terbacil for at least six of the years estimated. The extrapolated rates for surveyed CRDs used in EPest-high methods more than doubled the national total pesticide use for some years and pesticides for some specialty crops; for major crops, such as corn and alfalfa; and for some land uses, such as summer fallow, pasture and rangeland.

For the pesticides included in this study, EPest-low annual-use totals were less than or equal to EPest-high annual-use totals, as shown in *appendix 2*. However, EPest-low annual-use totals can be greater than EPest-high totals when the EPest-low pesticide-by-crop regional rate is greater than the EPest-high rate. EPest regional pesticide-by-crop rates are determined by using a minimum of three CRDs, and, typically, EPest-high regional rates were determined from a greater number of CRDs than EPest-low regional rates. In some cases, rates from additional CRDs can result in an EPest-high regional pesticide-by-crop rate that is less than the EPest-low regional rate. For example, if the EPest-low regional rate were determined from five rates—158, 54, 31.8, 9.68, and 5 pounds per acre—then the median would be 31.8 pounds of pesticide per harvested acre. The rates from these same five CRDs along with the EPest-high rates from any other CRDs in the region would be used to calculate the EPest-high regional rate. For example, if 158, 54, 31.8, 9.68, 9.05, 6.7, and 5 pounds of pesticide per crop acre were the rates used to determine the EPest-high regional rate, the EPest-high pesticide-by-crop regional rate would be 9.68 pounds of pesticide per harvested acre. Although these two rates were for the same counties in the region, the EPest-low total would be greater than the EPest-high use total.

In cases when a CRD was not surveyed, and a tier 1, tier 2, or regional rate was available, both EPest-low and EPest-high methods determined a pesticide-by-crop rate. In general, extrapolated rates for non-surveyed CRDs represented a greater percentage of use in more recent years because some pesticides were reported less frequently and some crops were not surveyed as extensively. EPest tier 1, tier 2, and regional rates have inherently greater uncertainty than rates for surveyed CRDs because a pesticide could have been applied to a localized area in response to a pest infestation, while the same crop grown in another part of the same region would not be managed in the same way, which can result in misrepresentative estimates of pesticide use. In addition, some EPest-high annual totals for pesticides that have been replaced or phased out, such as metolachlor and cyanazine, can be inaccurate because the EPest-high method assumes if a CRD was surveyed and an estimate for the pesticide was not reported, then an extrapolated rate could be used to estimate pesticide use.

Comparison of EPest National Estimates with Other Sources

National annual pesticide-use estimates developed by using EPest-low and EPest-high methods were compared with independently published estimates for seven herbicides. These comparisons were limited to acetochlor, alachlor, atrazine, EPTC, glyphosate, propanil, and trifluralin and to selected years because of limited data from the published sources. EPest totals for 1997, 2001, and 2007 were compared to (1) agricultural-use estimates published by the U.S. Environmental Protection Agency (USEPA; Kiely and others, 2004; Grube and others, 2011), (2) NASS-Agricultural Chemical Use (ACU) data (National Agricultural Statistics Service, 2008; hereinafter, referred to as NASS), and (3) National Pesticide Use Database (NPUD) estimates (Crop Protection Research Institute, 2006). NASS annual data were published as the “Total of Program States” in pounds per year and represent the amount of pesticide estimated for the states and crops that were surveyed for a specific year. Thus, the NASS national totals shown in these analyses are not intended to represent total use for all states or crops but are included as a point of reference. The USEPA estimates were reported as a range for each pesticide on agricultural crops as determined from a variety of public and proprietary data sources. Estimates for some pesticides and years were not available for each set of analyses, so comparisons were made for the years with the most complete data from each of the sources. Annual state estimates for the pesticides compared were available from EPest for 1992 through 2009; USEPA for 1997, 2001, 2003, 2005, and 2007; NPUD for 1992, 1997, 2002; and NASS for 1997, 2001, and 2006. In addition, NASS use estimates for propanil only were available for 2006. The NPUD estimates used in the 2001 analysis represent use for 2002, and the NPUD estimates were not included in the 2006–07 analysis. Lastly, the 2006–07 analysis did not include the USEPA use estimates for alachlor and EPTC.

Comparisons of EPest-low and EPest-high total use estimates with the USEPA, NASS, and NPUD data for 1997, 2001–02, and 2006–07 for the seven herbicides are shown in *figures 7A, 7B, and 7C*. With the exceptions of the EPest-low 2001 estimate for alachlor, the 2007 EPest-low and EPest-high estimates for propanil, and the 2007 EPest-high estimates for trifluralin, EPest and USEPA estimates differed from one another by less than 20 percent. NASS use estimates are not complete national estimates, so they were less than both EPest-low and EPest-high totals, and most 2006 NASS use estimates were a fraction of both USEPA and EPest totals because the number of the crops and states that were surveyed and reported by NASS was reduced in 2006. Overall, the comparisons illustrated in *figure 7* indicate a high level of agreement between EPest totals and both the USEPA and NPUD estimates, although none of these three sources of national estimates is known to be a better estimate of true use than the others.

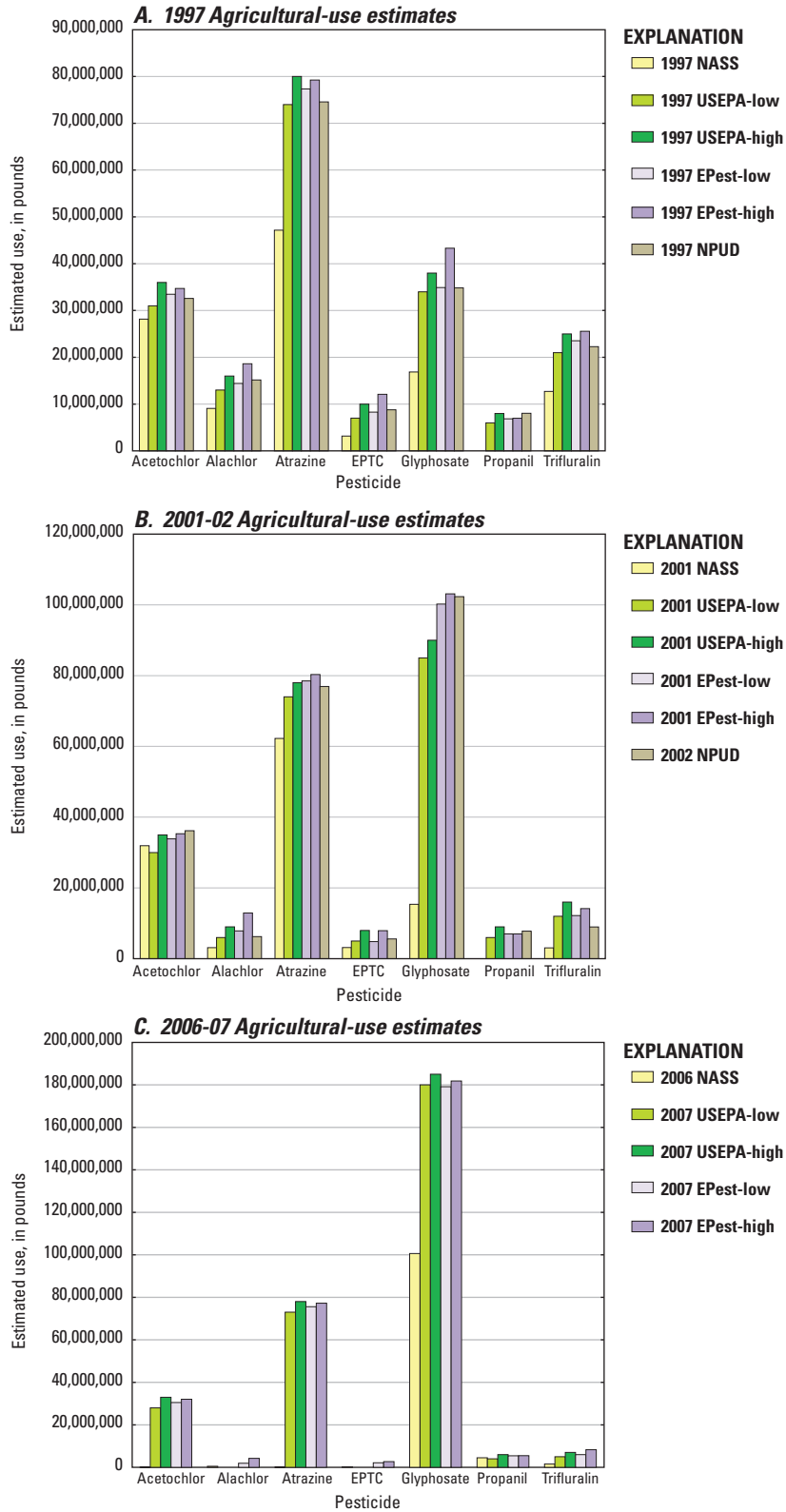


Figure 7. Comparison of EPEst-low and EPEst-high national total use of selected pesticides with national use estimates from other sources for (A) 1997 Agricultural-use estimates, (B) 2001–02 Agricultural-use estimates, and (C) 2006–07 Agricultural-use estimates. NASS, National Agricultural Statistics Service; USEPA, U.S. Environmental Protection Agency; EPEst, estimated pesticide use; National Pesticide Use Database.

Comparisons of EPest and NASS State Estimates

The national comparisons provide an aggregated assessment of how comparable EPest totals are to other published sources. In order to determine how well EPest use estimates represented regional and state level amounts and patterns of pesticide use, a second set of evaluations were made that compared EPest and NASS estimates for (1) state totals for individual pesticides and (2) state totals for individual pesticide-by-crop combinations. The comparisons between EPest and NASS state and state-by-crop estimates were the most controlled evaluations possible.

Comparison of State Total-Use Estimates

State-level comparisons were made for individual pesticides that have four or more estimates for combinations of states, crops, and years common to both EPest and NASS use estimates. Estimates for 33 pesticides and 34 states were compared for one or more years from 1992 through 2006. The pesticides included 24 herbicides, 8 insecticides, and 1 fungicide. Depending on the state and year, estimated state totals represented the sum of a pesticide used on one or more crops, including barley, corn, cotton, peanuts, rice, sorghum, soybeans, spring wheat, sunflowers, tobacco, and winter wheat. For each comparison, the difference between EPest and NASS use estimates was evaluated as the relative error (RE) for EPest relative to NASS estimates, or $(\text{EPest} - \text{NASS}) / \text{NASS}$, and RE was used to show the distribution of differences in state estimates for each pesticide (*fig. 8*). In *figures 8A* (EPest-low) and *8B* (EPest-high), positive RE values represent EPest totals that were greater than NASS use estimates and negative RE values represent EPest

totals that were less than NASS use estimates. Although differences between EPest and NASS estimates are expressed as proportional errors relative to NASS estimates in order to facilitate clear comparisons to publicly available NASS estimates, neither estimate can be considered a more certain estimate of true values than the other. The number of state-by-year combinations for each pesticide is indicated at the bottom of the plot (*fig. 8*). For the different pesticides, the number of state-by-year combinations used in the comparisons ranged from as few as 5 to as many as 443.

Of the 33 pesticides evaluated, less than one-third—10 EPest-low and 8 EPest-high— had median RE values significantly different from zero based on the 95-percent confidence interval on the median RE. For EPest-low, 6 of the 10 pesticides that were significantly different from NASS use estimates tended to have lower estimates compared to NASS (acifluorfen, bentazon, butylate, methomyl, methyl parathion, and propachlor), and the rest (atrazine, fluometuron, nicosulfuron, and propargite) tended to be greater than NASS. Compared to NASS use estimates, seven of the eight significantly different EPest-high totals tended toward overestimation (atrazine, fluometuron, fonofos, metribuzin, nicosulfuron, propargite, and trifluralin), and only one pesticide (methyl parathion) tended toward underestimation. The inter-quartile ranges for both sets of estimates generally were symmetrical for most pesticides, and there was a relatively small proportion of outlying individual values—generally fewer than 10 percent. Several pesticides showed wide confidence intervals around the median, and some had only a small number of estimates to compare, including propachlor and thiobencarb, among others.

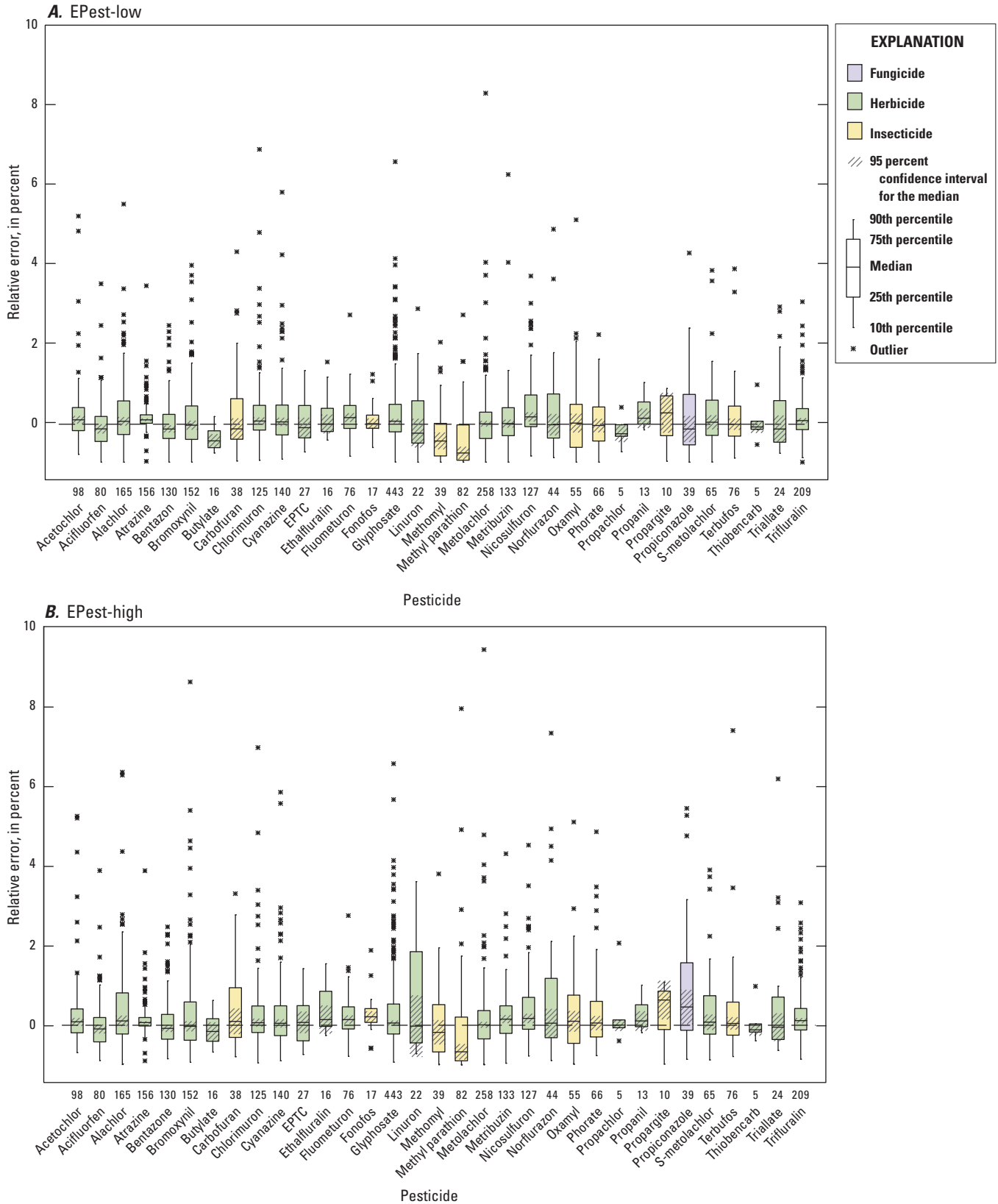


Figure 8. Distributions of relative error between EPest and National Agricultural Statistics Service (NASS) use estimates. Relative error expressed as $(EPEST - NASS)/NASS$. Estimated state totals represent the sum of use on one or more crops, including barley, corn, cotton, peanuts, rice, sorghum, soybeans, spring wheat, sunflowers, tobacco, and winter wheat. Numbers for each pesticide represent the number of state-by-year combinations compared.

Comparison of State Estimates for Individual Pesticide-by-Crop Combinations

EPest and NASS use estimates for individual states and crops were compared for selected years from 1992 to 2006, which are the most direct comparisons possible with the data available. The comparisons were limited to pesticide-by-crop combinations that had both EPest and NASS use estimates for at least 10 state-year combinations. This requirement allowed one or more crop comparisons for 29 pesticides, including 21 herbicides, 7 insecticides, and 1 fungicide, for one or more of the following crops: corn, cotton, rice, soybeans, spring wheat, and winter wheat. There were 17 pesticides compared for corn, 13 pesticides for cotton, 9 pesticides for soybeans, 4 pesticides for winter wheat, 4 pesticides for spring wheat, and a single pesticide for rice. Although NASS also reported pesticide-use estimates for other crops included in the all-crops state totals, such as sorghum, tobacco, peanuts, and barley, there were too few estimates for each of these crops to include them in the crop-specific comparisons.

The distribution of RE values for all available state-year combinations for each of the 47 pesticide-by-crop combinations are shown by crop (rice excluded) in *figures 9A–9E* for EPest-low totals and in *figures 10A–10E* for EPest-high totals. The figures show that the range of RE values for EPest-low totals for most pesticide-by-crop combinations was less than for EPest-high totals and contained fewer outliers, indicating that EPest-low totals tended to approximate NASS estimates more accurately than EPest-high totals.

Similarly, more than two-thirds (33 of 48) of EPest-low pesticide-by-crop combinations had median REs that were 15 percent or less, whereas just over half (26 of 48) of the EPest-high totals had median REs that were less than 15 percent or less (*tables 3 and 4*). Of the 15 EPest-low pesticide-by-crop combinations that had median REs that differed by 15 percent or more, 13 pesticide crop-combinations were less than NASS use estimates and 2 pesticide-by-crop combinations were greater than NASS use estimates (*table 3*). There were 21 EPest-high pesticide-by-crop combinations that had median REs greater than 15 percent, with 13 combinations greater than NASS use estimates and 8 that were less (*table 4*). These results were consistent with the aggregated state total comparisons presented previously, and overall, these comparisons indicated a reasonable agreement between EPest and NASS use estimates, with somewhat better agreement for EPest-low than high estimates. Nevertheless, some pesticide-by-crop combinations showed substantial differences in the estimates for specific states and years.

A combination of statistical tests were used to compare EPest and NASS use estimates for the pesticide-by-crop combinations. The Wilcoxon signed rank sum test (Conover, 1980; Lehmann, 1975) was used to further evaluate differences between magnitudes of EPest and NASS annual use estimates for each pesticide-by-crop combination with

sufficient state-year combinations. This non-parametric test evaluates whether the median difference between paired estimates is significantly different than zero, where significance was assigned to a probability (p) of less than 0.05 (two-tailed test). Comparisons that are not statistically significant can indicate agreement between estimates or also can indicate variability in the sample too great to establish significant differences. To help assess the degree of correlation between two ranked pairs of estimates, the Spearman rank correlation coefficient (r) was used, where values range from 0 to 1, and 1 indicates perfect agreement between estimates. The p -value from the Wilcoxon test, the Spearman correlation coefficient (r), the median RE, and the number of state/year combinations used in the evaluations of the comparisons to NASS use estimates are shown for each pesticide and crop combination in *table 3* for EPest-low and *table 4* for EPest-high.

The strongest agreement between estimates is indicated by statistically insignificant p -values, correlation coefficients approaching 1, and a low median and range for RE values. Pesticides evaluated in this study that met these criteria included acetochlor, cyanazine, and terbufos use estimates for corn, as well as chlorimuron and bentazon use estimates for soybeans. Some estimate comparisons had significantly different medians, but still showed strong correlation and a low RE value; examples include estimates for atrazine and metolachlor use for corn and trifluralin use estimates for cotton. Poor agreement between estimates was indicated by large RE values and low correlation coefficients for both significant and insignificant comparisons of medians. A small sample size can reduce the power of the tests, however, and smaller sample sizes were often associated with the lower correlation coefficients among these comparisons, particularly when RE values were greater than 0.15.

More than half of the comparisons of pesticide-by-crop combinations had RE values less than 0.15, and the majority of these comparisons were not significantly different and had correlation coefficients greater than 0.75. Of the 48 pesticide-by-crop combinations with 10 or more state-by-year combinations, 12 of the EPest-low pesticide-by-crop totals and 17 of the EPest-high totals significantly differed ($p < 0.05$) from the NASS use estimates. Of the comparisons with significant differences, two-thirds or more of the pesticide-by-crop combinations had correlation coefficients greater than 0.75, especially when comparisons had RE values of 0.15 or less. Comparisons that did not have significant differences tended to have lower RE values than comparisons that had significant differences. Nevertheless, about a quarter of all the comparisons had RE values greater than 0.15, but did not have significant differences. All of these had sample numbers less than 40, and most had fewer than 20 samples for comparison. Also, most had correlation coefficients less than 0.75, which demonstrates the importance of having a sample number large enough to achieve a good comparison of estimates.

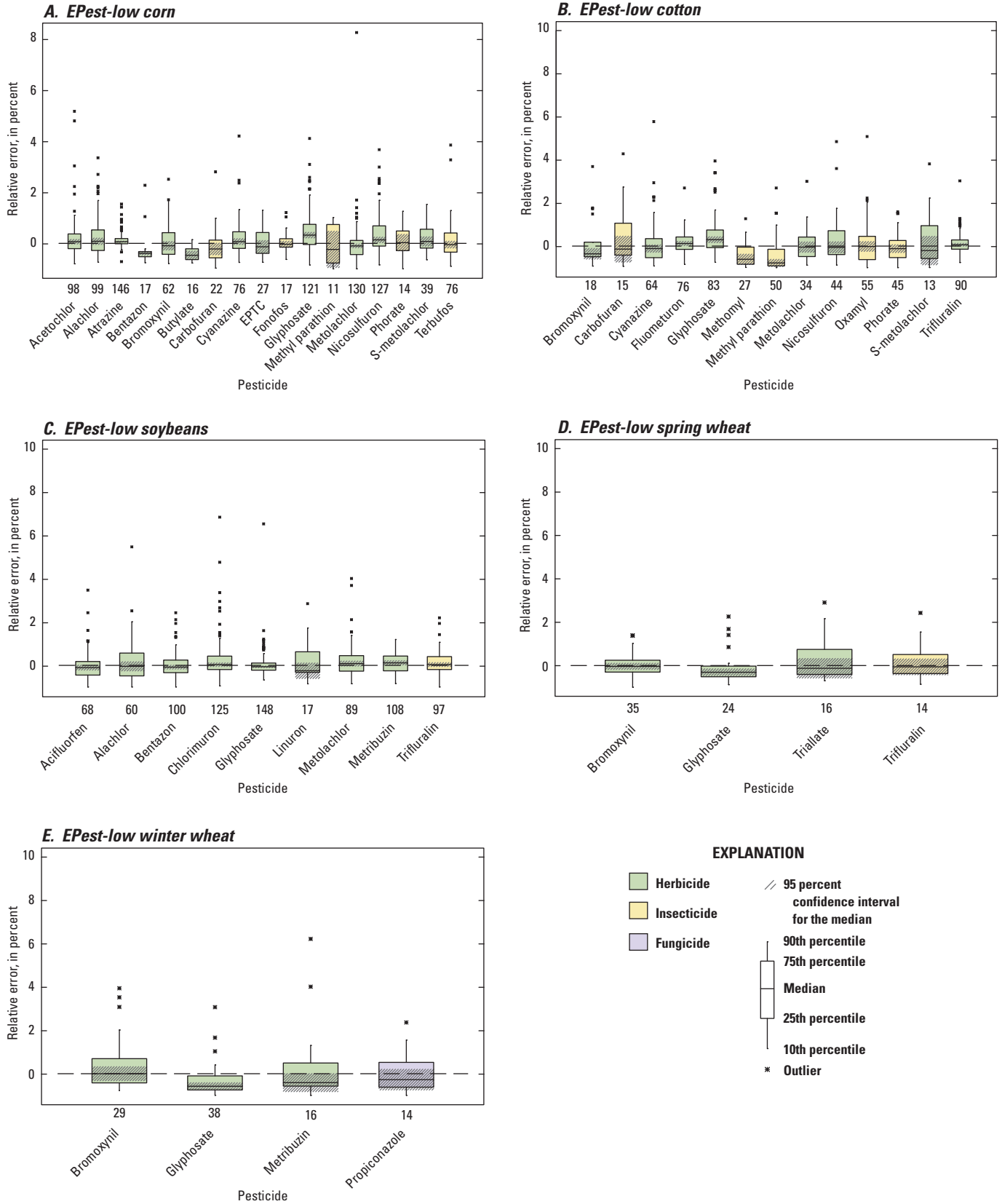


Figure 9. Distribution of relative error between EPest-low and National Agricultural Statistics Service (NASS) use estimates for (A) corn, (B) cotton, (C) soybeans, (D) spring wheat, and (E) winter wheat. Relative error expressed as (EPEST - NASS)/NASS.

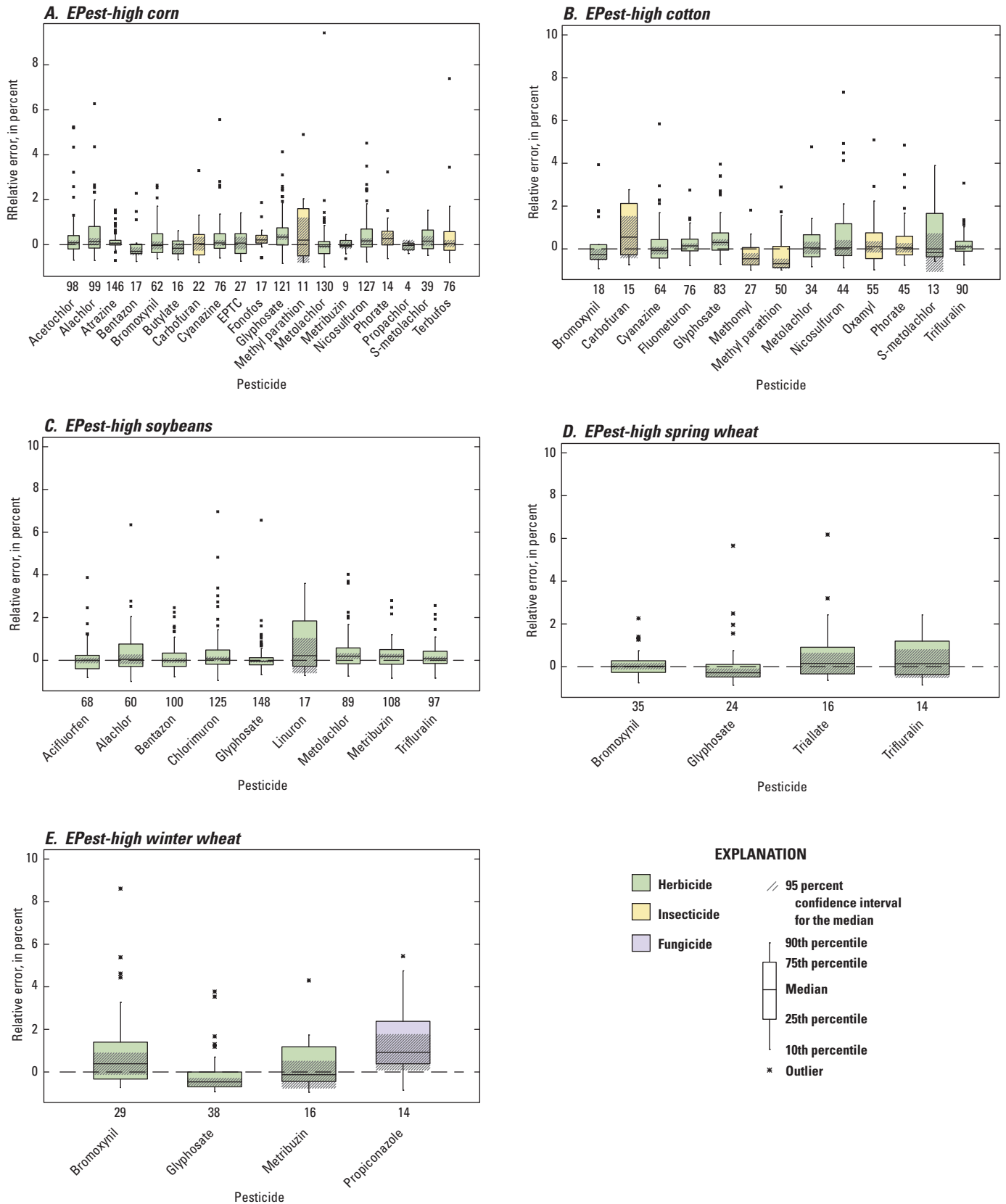


Figure 10. Distribution of relative error between EPest-high and National Agricultural Statistics Service (NASS) use estimates for (A) corn, (B) cotton, (C) soybeans, (D) spring wheat, and (E) winter wheat. Relative error expressed as (EPEST - NASS)/NASS.

Table 3. Summary of statistics from comparison of EPest-low and National Agricultural Statistics Service (NASS) pesticide-by-crop estimates.

[Abbreviations: N, number of estimates compared; P, probability of significance; >, greater than; <, less than; -, no data]

Pesticide	Type	Corn				Cotton							
		N	Wilcoxon signed rank, P (two-tailed)	Wilcoxon signed rank, P (NASS >EPest)	Wilcoxon signed rank, P (NASS <EPest)	Median relative error	Spearman correlation coefficient	N	Wilcoxon signed rank, P (two-tailed)	Wilcoxon signed rank, P (NASS >EPest)	Wilcoxon signed rank, P (NASS <EPest)	Median relative error	Spearman correlation coefficient
Acetochlor	Herbicide	98	0.19	0.90	0.10	0.07	0.93	-	-	-	-	-	-
Aciflurofen	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-
Alachlor	Herbicide	99	0.13	0.94	0.06	0.08	0.83	-	-	-	-	-	-
Atrazine	Herbicide	146	0.00	1.00	0.00	0.07	0.97	-	-	-	-	-	-
Bentazon	Herbicide	17	0.01	0.01	0.99	(0.39)	0.42	-	-	-	-	-	-
Bromoxynil	Herbicide	62	0.29	0.15	0.86	(0.09)	0.85	18	0.12	0.06	0.95	(0.36)	0.71
Butylate	Herbicide	16	0.00	0.00	1.00	(0.47)	0.91	-	-	-	-	-	-
Carbofuran	Insecticide	22	0.18	0.09	0.92	(0.22)	0.68	15	0.56	0.74	0.28	(0.07)	0.73
Chlorimuron	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-
Cyanazine	Herbicide	76	0.20	0.90	0.10	0.07	0.92	64	0.65	0.32	0.68	(0.11)	0.82
EPTC	Herbicide	27	0.24	0.12	0.88	(0.13)	0.60	-	-	-	-	-	-
Fluometuron	Herbicide	-	-	-	-	-	-	76	0.00	1.00	0.00	0.12	0.93
Fonofos	Insecticide	17	0.85	0.59	0.43	(0.03)	0.72	-	-	-	-	-	-
Glyphosate	Herbicide	121	0.00	1.00	0.00	0.34	0.78	83	0.00	1.00	0.00	0.30	0.93
Linuron	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-
Methomyl	Insecticide	-	-	-	-	-	-	27	0.01	0.00	1.00	(0.61)	0.76
Methyl parathion	Insecticide	11	0.15	0.07	0.94	(0.24)	0.32	50	0.00	0.00	1.00	(0.78)	0.47
Metolachlor	Herbicide	130	0.00	0.00	1.00	(0.10)	0.87	34	0.30	0.15	0.85	(0.04)	0.58
Metribuzin	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-
Nicosulfuron	Herbicide	127	0.00	1.00	0.00	0.14	0.84	-	-	-	-	-	-
Norflurazon	Herbicide	-	-	-	-	-	-	44	0.48	0.24	0.76	(0.06)	0.78
Oxamyl	Insecticide	-	-	-	-	-	-	55	0.47	0.23	0.77	0.02	0.68
Phorate	Insecticide	14	0.86	0.60	0.43	0.03	0.50	45	0.62	0.31	0.69	(0.12)	0.80
Propanil	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-
Propiconazole	Fungicide	-	-	-	-	-	-	-	-	-	-	-	-
S-Metolachlor	Herbicide	39	0.06	0.97	0.03	0.08	0.90	13	0.41	0.21	0.81	(0.21)	0.37
Terbufos	Insecticide	76	0.29	0.15	0.86	(0.05)	0.83	-	-	-	-	-	-
Triallate	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-
Trifluralin	Herbicide	-	-	-	-	-	-	90	0.08	0.96	0.04	0.06	0.96

Table 4. Summary of statistics from comparison of EPest-high and National Agricultural Statistics Service (NASS) pesticide-by-crop estimates.

[Abbreviations: N, number of estimates compared; P, probability of significance; <, less than; >, greater than; -, no data]

Pesticide	Type	Corn						Cotton						
		N	Wilcoxon signed rank, P (two-tailed)	Wilcoxon signed rank, P (NASS > EPest)	Wilcoxon signed rank, P (NASS < EPest)	Median relative error	Spearman correlation coefficient	N	Wilcoxon signed rank, P (two-tailed)	Wilcoxon signed rank, P (NASS > EPest)	Wilcoxon signed rank, P (NASS < EPest)	Median relative error	Spearman correlation coefficient	
Acetochlor	Herbicide	98	0.07	0.96	0.36	0.08	0.93	-	-	-	-	-	-	-
Aciflurofen	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-	-
Alachlor	Herbicide	99	0.01	1.00	0.00	0.13	0.82	-	-	-	-	-	-	-
Atrazine	Herbicide	146	0.00	1.00	0.00	0.07	0.97	-	-	-	-	-	-	-
Bentazon	Herbicide	17	0.04	0.22	0.98	(0.30)	0.34	-	-	-	-	-	-	-
Bromoxynil	Herbicide	62	0.96	0.53	0.48	(0.03)	0.85	18	0.15	0.08	0.93	(0.26)	0.74	0.74
Butylate	Herbicide	16	0.19	0.10	0.91	(0.16)	0.81	-	-	-	-	-	-	-
Carbofuran	Insecticide	22	0.82	0.41	0.60	0.05	0.74	15	0.30	0.86	0.15	0.55	0.63	0.63
Chlorimuron	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-	-
Cyanazine	Herbicide	76	0.09	0.95	0.05	0.08	0.92	64	0.92	0.54	0.46	(0.08)	0.83	0.83
EPTC	Herbicide	27	0.71	0.35	0.65	0.07	0.56	-	-	-	-	-	-	-
Fluometuron	Herbicide	-	-	-	-	-	-	76	0.00	1.00	0.00	0.14	0.93	0.93
Fonofos	Insecticide	17	0.05	0.98	0.03	0.21	0.73	-	-	-	-	-	-	-
Glyphosate	Herbicide	121	0.00	1.00	0.00	0.34	0.79	83	0.00	1.00	0.00	0.30	0.92	0.92
Linuron	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-	-
Methomyl	Insecticide	-	-	-	-	-	-	27	0.05	0.27	0.97	(0.46)	0.74	0.74
Methyl parathion	Insecticide	11	1.00	0.52	0.52	0.20	0.16	50	0.00	0.00	1.00	(0.69)	0.52	0.52
Metolachlor	Herbicide	130	0.01	0.00	1.00	(0.07)	0.88	34	0.99	0.49	0.51	0.05	0.65	0.65
Metribuzin	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-	-
Nicosulfuron	Herbicide	127	0.00	1.00	0.00	0.17	0.84	-	-	-	-	-	-	-
Norflurazon	Herbicide	-	-	-	-	-	-	44	0.38	0.81	0.19	0.05	0.74	0.74
Oxamyl	Insecticide	-	-	-	-	-	-	55	0.65	0.68	0.32	0.10	0.64	0.64
Phorate	Insecticide	14	0.05	0.98	0.02	0.28	0.59	45	0.22	0.89	0.11	0.05	0.71	0.71
Propanil	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-	-
Propiconazole	Fungicide	-	-	-	-	-	-	-	-	-	-	-	-	-
S-Metolachlor	Herbicide	39	0.04	0.98	0.02	0.16	0.91	13	1.00	0.53	0.50	(0.17)	0.37	0.37
Terbufos	Insecticide	76	0.51	0.75	0.25	0.04	0.83	-	-	-	-	-	-	-
Triallate	Herbicide	-	-	-	-	-	-	-	-	-	-	-	-	-
Trifluralin	Herbicide	-	-	-	-	-	-	90	0.03	0.99	0.01	0.10	0.95	0.95

Comparisons of EPest-low tended to show stronger correlation to NASS use estimates than EPest-high and also had a greater number of RE values less than 0.15, which, along with fewer significant differences between medians, indicated that EPest-low totals better approximated NASS use estimates than EPest-high overall. In general, however, the majority of the comparisons of estimates showed agreement, although low sample size limited the power of the tests for some pesticide-by-crop combinations.

Comparisons of EPest-low and EPest-high crop-pesticide combinations with NASS use estimates were further examined to evaluate differences between the estimates. These comparisons provide an understanding of the types and degrees of differences between EPest and NASS estimates and how the statistical tests summarize them.

Herbicide Estimate Comparisons between EPest and NASS

Statistically significant differences in median estimates between the methods are important to understand because they can provide information about similarities and differences in the estimates. One or both EPest medians for 11 of the 21 herbicides were significantly different than NASS median use estimates (*tables 3 and 4*). For six of these herbicides—atrazine, bentazon, fluometuron, glyphosate, metolachlor, and nicosulfuron—both EPest-low and EPest-high medians differed significantly from NASS median use estimates. In addition, EPest-high (but not EPest-low) medians for alachlor,

metribuzin, *S*-metolachlor, and trifluralin were significantly different from NASS median use estimates, and EPest-low (but not EPest-high) medians for butylate were significantly different from NASS median use estimates. Use estimates for more than one crop were compared for some pesticides, such as metolachlor and bentazon, and both EPest medians (low and high) were significantly different from NASS median use estimates for some but not all of the crops that were compared. For example, EPest-low and EPest-high bentazon medians were significantly different than NASS median use estimates for corn but not soybeans.

Examining the data and statistical results of the pesticide-by-crop comparisons can help to better assess and understand how well the EPest method approximated current NASS pesticide-use estimates. The following sections present the data graphically and discuss the results of the statistical tests for a selection of the pesticide-by-crop combinations that showed significant differences for one or both methods. For all pesticide-by-crop combinations presented, two plots are shown: (1) a scatterplot of EPest-low and NASS state pesticide-use totals for the years compared (only plots of EPest-low estimates were used because they are similar to the EPest-high versions of the scatterplots) and (2) a plot of differences between EPest estimates and NASS state pesticide-use estimates on a common scale, organized by USDA Farm Production Regions. Because their boundaries conform to state boundaries, Farm Production Regions (*fig. 11*) were selected rather than the USDA Farm Resource Regions that were used to calculate EPest regional rates.

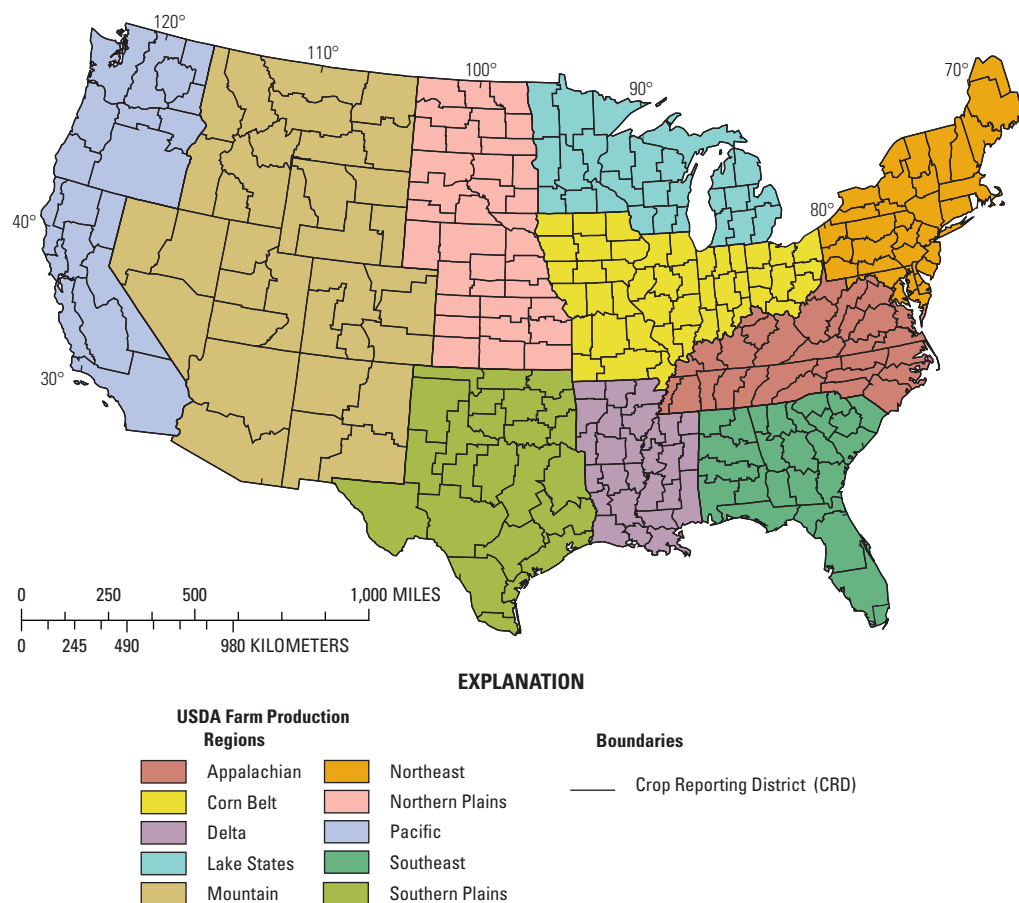


Figure 11. U.S. Department of Agriculture Farm Production Regions.

Alachlor

For 19 states and most of the years from 1992 through 2003, 99 EPest-low and EPest-high estimates of alachlor use on corn were compared with NASS estimates. Only EPest-high estimates significantly differed ($p < 0.05$) from NASS use estimates, but both EPest totals tended to be greater than NASS totals. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 8 and 13 percent greater, respectively, indicating a general tendency for EPest estimates to be greater than NASS estimates. Correlation coefficients for EPest-low and NASS comparisons

were 0.83 and were 0.82 for EPest-high. The relation between EPest-low and NASS estimates for alachlor is shown in *figure 12A*, and the differences between NASS estimates and both EPest-low and EPest-high are shown by region and state in *figure 12B*.

The majority of EPest-low and EPest-high estimates differed from NASS use estimates by less than a factor of two (*fig. 12B*), and most EPest and NASS use estimates followed similar trends use for the years compared. Of the approximately 20 percent (20 of 99) of EPest-high estimates that were more than double the NASS estimate, most were in the Corn Belt and Lake States regions.

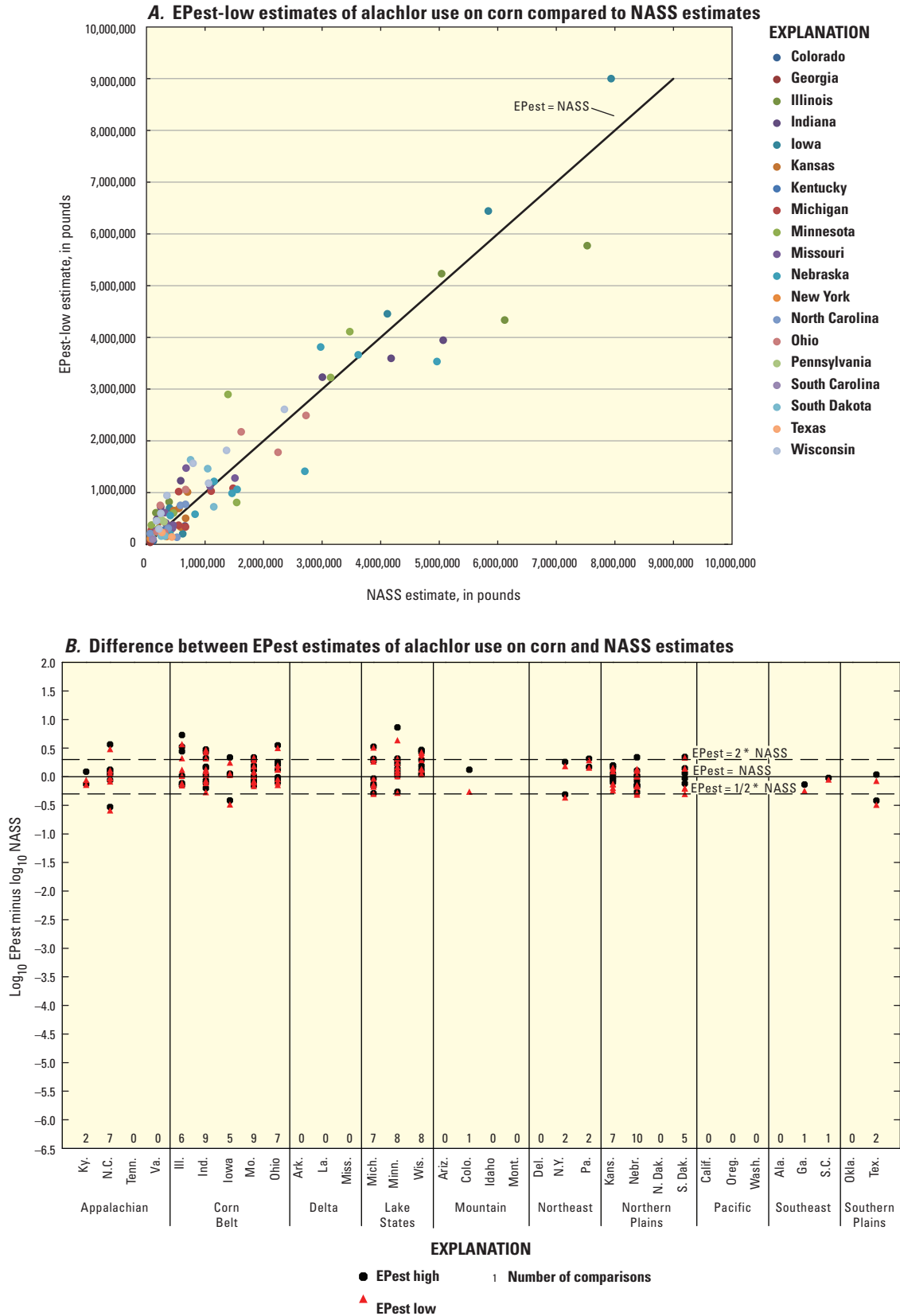


Figure 12. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of alachlor use on corn: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

Atrazine

For various years from 1992 to 2003, 146 EPest-low and EPest-high estimates of atrazine use on corn were compared with NASS use estimates for 20 states located in the Appalachian, Corn Belt, Lake States, Mountain, Northeast, Northern Plains, Southeast, and Southern Plains regions. Both EPest-low and EPest-high estimates were significantly different than NASS use estimates ($p < 0.05$). The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were both 7 percent greater, indicating a general tendency for EPest estimates to be slightly greater than NASS estimates. Both EPest-low and EPest-high had correlation coefficients of 0.97 with NASS use estimates, which were among the strongest correlations between pesticide use estimates in this study. The relation between EPest and NASS estimates of atrazine estimates is shown in *figure 13A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 13B*.

Almost all of the EPest and NASS estimates (142 of 146) differed by less than a factor of two (*fig. 13B*), but a majority of EPest estimates were slightly greater than NASS estimates. EPest and NASS use estimates were about the same for the Appalachian, Corn Belt, Northeast, and Southeast regions, but greater differences were found for one or more estimates from the Lake States, Mountain, and Northern Plains regions.

Bentazon

For various years from 1992 through 2001, 17 EPest-low and EPest-high estimates of bentazon use on corn estimates were compared with NASS estimates for four states from the Corn Belt and Lake States regions. Both EPest-low and EPest-high estimates significantly differed from NASS use estimates ($p < 0.05$). The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 39 and 30 percent less, respectively, indicating a general tendency for EPest estimates to be less than NASS estimates. The correlation coefficients for the relation between the EPest and NASS estimates were 0.42 for EPest-low and 0.34 for EPest-high. The relation between EPest-low and NASS estimates of bentazon use on corn is shown in *figure 14A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 14B*.

About one-half (9 of 17) of the EPest-low estimates and 65 percent (11 of 17) of the EPest-high estimates differed by less than a factor of two from NASS estimates. There were large differences between the EPest estimates and NASS use estimates for some states and years, which, in conjunction with a relatively small sample size, likely contributes to the poor correlation between the estimates.

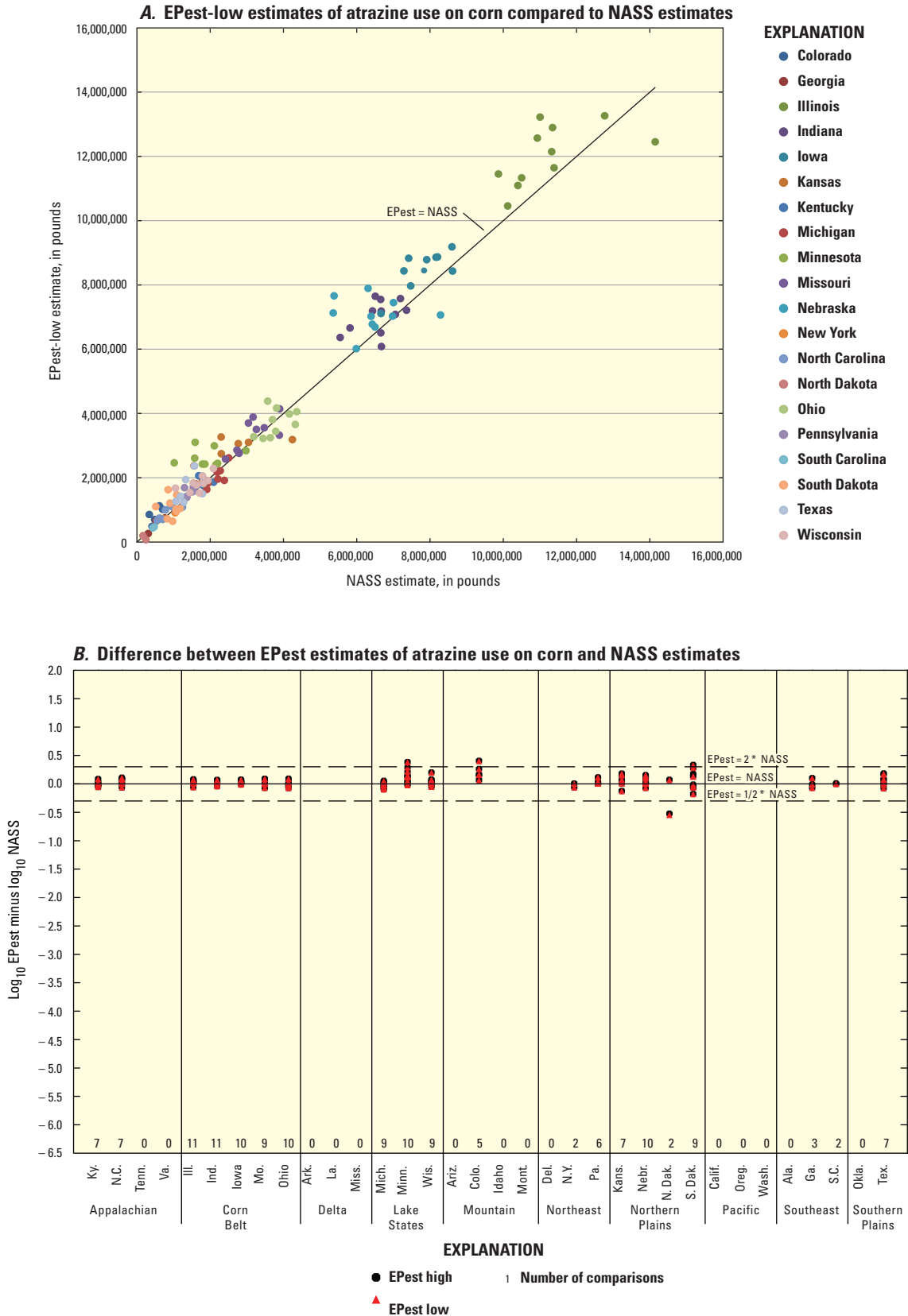


Figure 13. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of atrazine use on corn: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

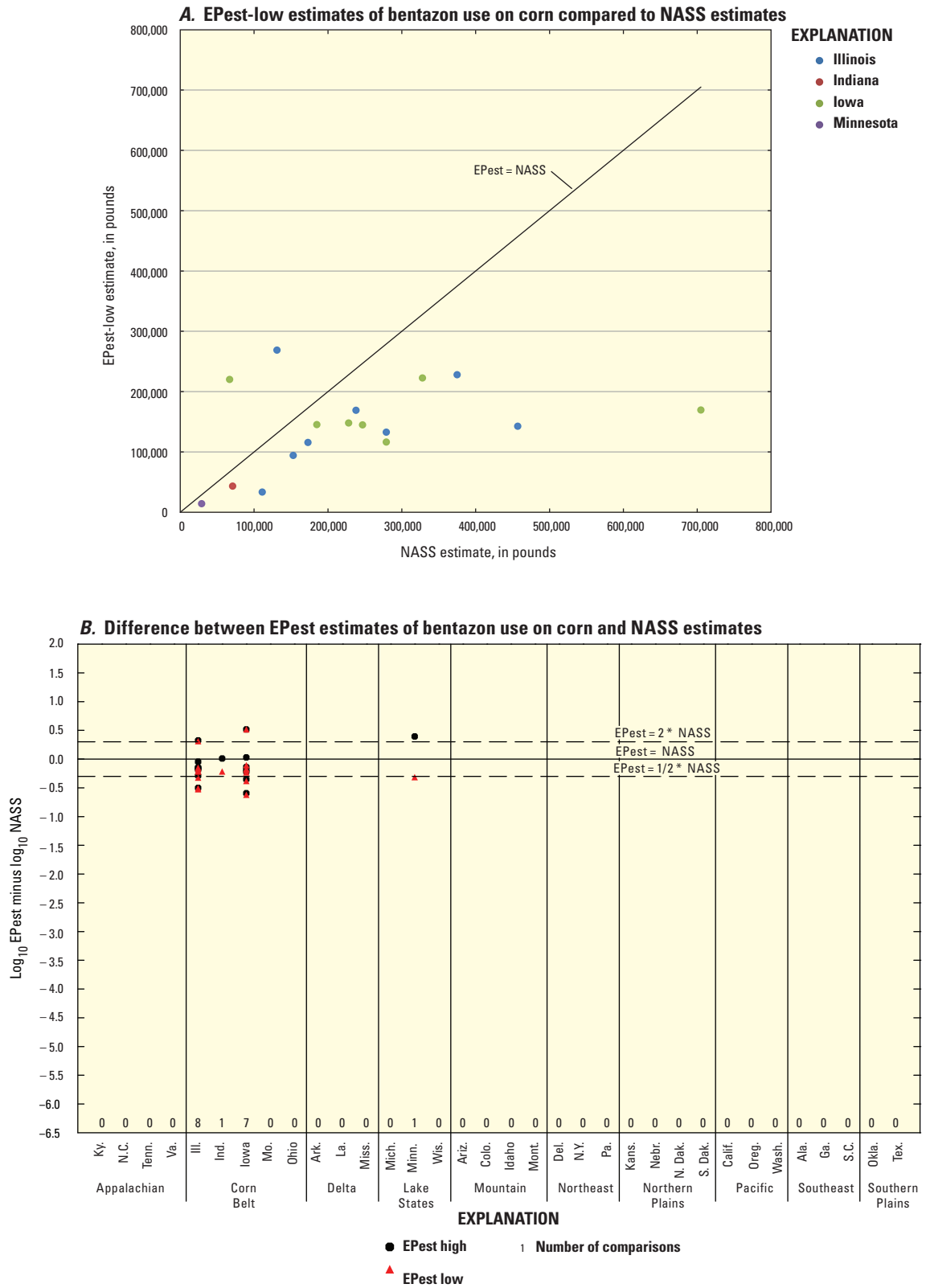


Figure 14. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of bentazon use on corn: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates (\log_{10} EPest – \log_{10} NASS).

Butylate

Sixteen EPest-low and EPest-high estimates of butylate use on corn estimates were compared with NASS estimates for eight states from the Appalachian, Corn Belt, Northern Plains, and Southeast regions from 1992 through 1994. Only EPest-low estimates significantly differed from NASS use estimates ($p < 0.05$). The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 47 and 16 percent less, respectively, indicating a general tendency for EPest estimates to be less than NASS estimates. The correlation coefficient for comparison to NASS estimates to EPest-low was 0.91 and was 0.81 for EPest-high. The relation between EPest-low and NASS estimates for butylate use is shown in *figure 15A*, and the differences between NASS estimates and both EPest-low and EPest-high are shown by region and state in *figure 15B*.

The majority of EPest estimates (14 of 16 EPest-low and 10 of 16 EPest-high) were less than NASS estimates, but there was a fairly strong correlation between the estimates. Most EPest-low butylate estimates were 15 to 80 percent less than NASS estimates.

Fluometuron

For various years from 1992 through 2005, 76 EPest and NASS estimates of fluometuron use on cotton were compared for 11 states from the Appalachian, Corn Belt, Delta, Mountain, Southeast, and Southern Plains regions. Both EPest-low and EPest-high estimates significantly differed ($p < 0.05$) from NASS estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 12 and 14 percent greater, respectively, indicating a general tendency for EPest estimates to be slightly greater than NASS estimates. Both EPest-low and EPest-high had correlation coefficients of 0.93 with NASS use estimates. The relation between EPest-low and NASS estimates for fluometuron is shown in *figure 16A*, and the differences between NASS estimates and both EPest-low and EPest-high rate estimates are shown by region and state in *figure 16B*.

The majority of the EPest-low (68 of 76) and EPest-high (67 of 76) estimates differed from NASS use estimates by less than a factor of two. EPest estimates tended to be greater than NASS estimates for most of the regions compared, including one or more estimates for states from the Mountain, Southeast and Southern Plains regions, which were at least twice NASS estimates. EPest totals tended to be less than NASS use estimates for some of the states in the Appalachian, Delta, and Southern Plains, however.

Glyphosate

EPest and NASS estimates of glyphosate use were compared for corn, cotton, soybeans, spring wheat, and winter wheat crops. EPest estimates significantly differed from NASS estimates for the crops evaluated, except for soybeans, which also had the highest correlation coefficient between EPest and NASS estimates and the lowest median RE. Comparisons of EPest and NASS estimates for glyphosate use on spring and winter wheat crops showed low correlation coefficients and small sample sizes, which limits the power of the statistical tests. EPest and NASS estimates of glyphosate use on corn and cotton are discussed in the following sections.

Corn

For glyphosate use on corn, 121 EPest and NASS estimates were compared from 19 states from the Appalachian, Corn Belt, Lake States, Mountain, Northeast, Northern Plains, Southeast, and Southern Plains regions. Both EPest-low and EPest-high estimates significantly differed ($p < 0.05$) from NASS estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were both 34 percent greater, indicating a general tendency for EPest estimates to be greater than NASS estimates. Correlation coefficients for EPest-low and NASS comparisons were 0.78 and were 0.79 for EPest-high. The relation between EPest-low and NASS estimates for glyphosate use on corn is shown in *figure 17A*, and the differences between NASS estimates and both EPest-low and EPest-high are shown by region and state in *figure 17B*.

Most of the EPest and NASS estimates (90 or more of 121) differed by less than a factor of two. EPest-low and EPest-high estimates for the Corn Belt, Lake States, Northeast, Southeast, and Southern Plains regions tended to be greater than NASS estimates, and estimates for one or more states in each of these regions had EPest estimates that were more than twice the NASS estimate (*fig. 17B*).

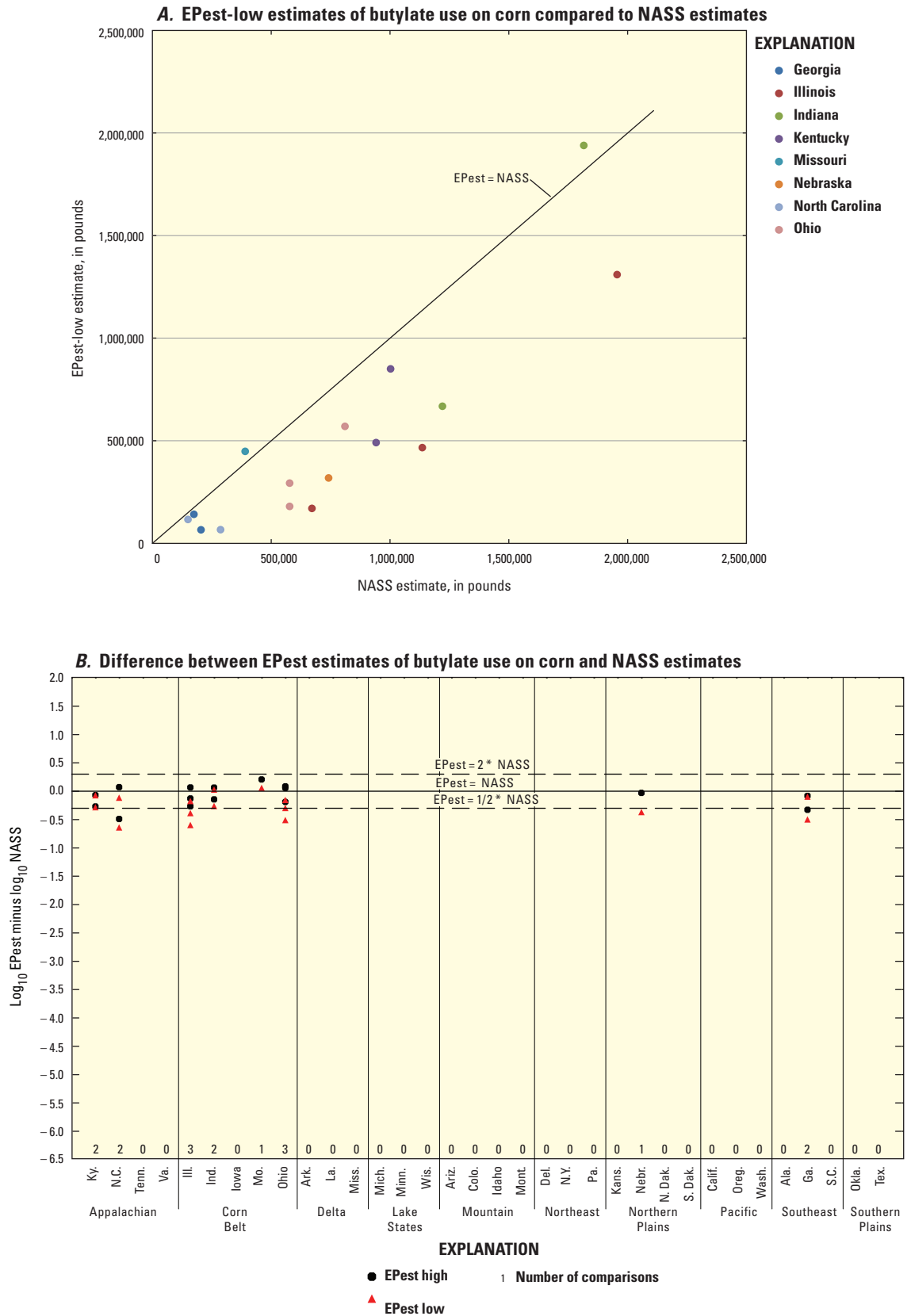


Figure 15. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of butylate use on corn: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

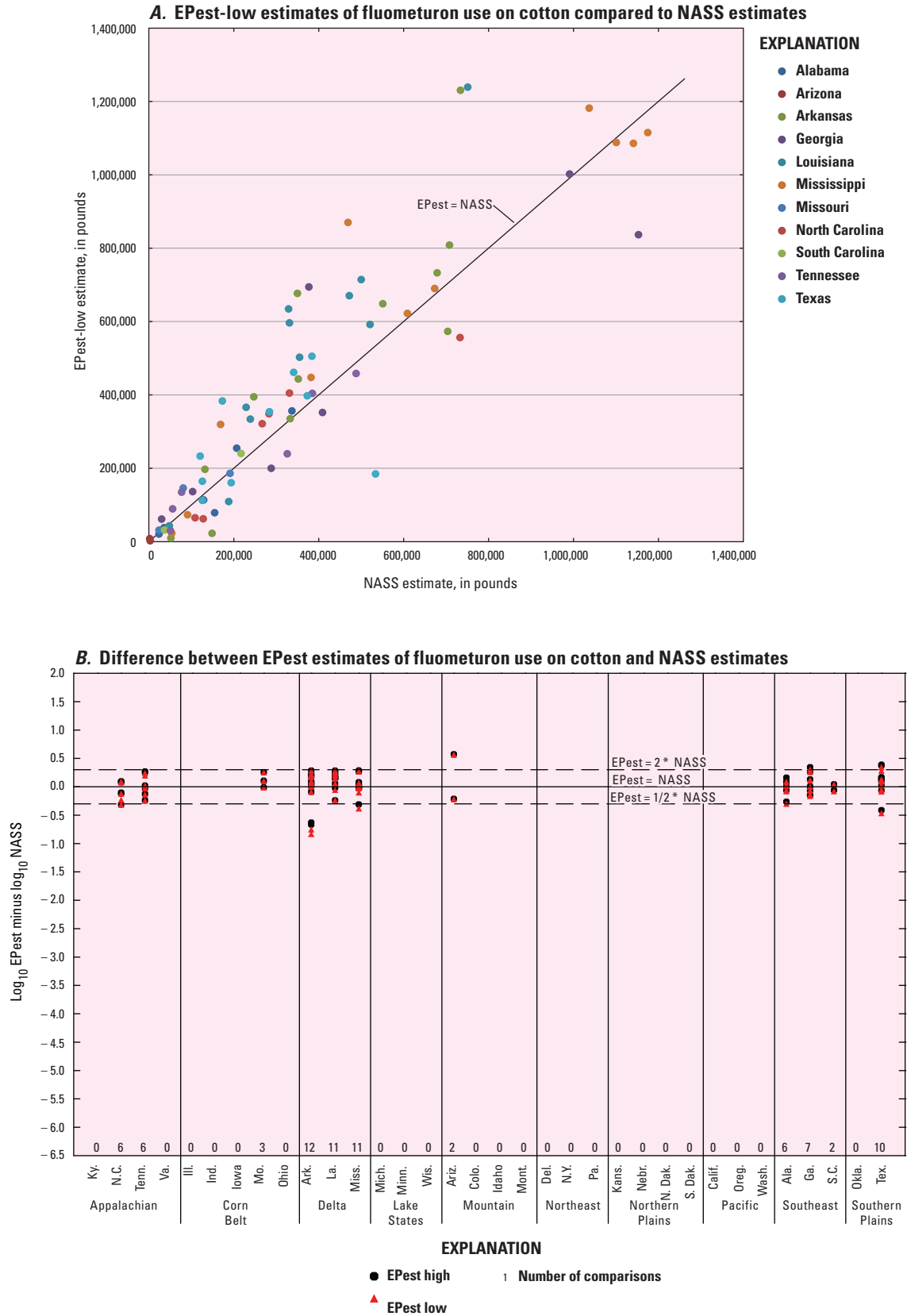


Figure 16. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of fluometuron use on cotton: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

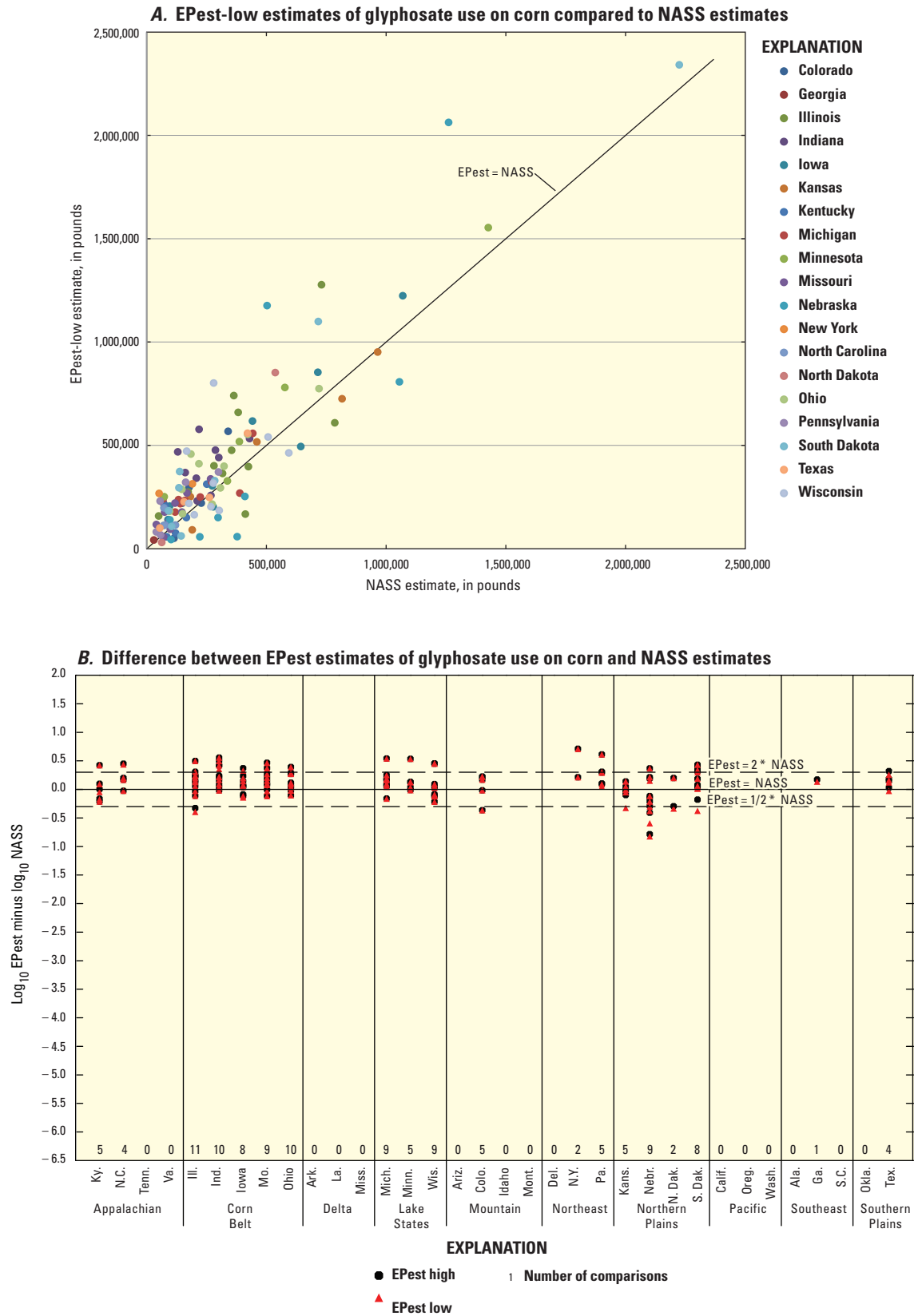


Figure 17. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of glyphosate use on corn: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

Cotton

For various years from 1992 through 2005, 83 EPest-low and EPest-high estimates of glyphosate use on cotton were compared with NASS estimates for 12 states from Appalachian, Corn Belt, Delta, Mountain, Pacific, Southeast, and Southern Plains regions. Both EPest-low and EPest-high estimates significantly differed ($p < 0.05$) from NASS use estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were both 30 percent greater, indicating a general tendency for EPest estimates to be greater than NASS estimates. Correlation coefficients for EPest-low and NASS comparisons were 0.93 and were 0.92 for EPest-high. The relation between EPest and NASS estimates of glyphosate use on cotton is shown in *figure 18A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 18B*.

Most EPest and NASS estimates (63 of 83) differed by less than a factor of two. EPest estimates for the Appalachian, Delta and Corn Belt regions bracketed NASS use estimates, whereas in most other regions, EPest estimates were greater than NASS use estimates. One reason for this difference could be that EPest pesticide totals include pesticide use on both upland and Pima cotton, whereas NASS reports pesticide use for upland cotton only.

Metolachlor

Corn

For various years from 1992 through 2003, 130 EPest-low and EPest-high estimates of metolachlor use on corn were compared with NASS estimates for 18 states from the Appalachian, Corn Belt, Lake States, Mountain, Northeast, Southeast, and Northern and Southern Plains regions. Both EPest-low and EPest-high estimates significantly differed ($p < 0.05$) from NASS use estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 10 and 7 percent lower, respectively, indicating a general tendency for EPest estimates to be less than NASS estimates. Correlation coefficients for EPest-low and NASS comparisons were 0.87 and were 0.88 for EPest-high. The relation between EPest-low and NASS estimates of metolachlor use on corn is shown in *figure 19A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 19B*.

Most EPest estimates differed from NASS estimates by less than a factor of two, and estimates for most states bracketed NASS estimates. From 1998 through 2003, however, there were 30 EPest-low and EPest-high estimates that were more than 50 percent lower than NASS estimates, representing some of the greatest underestimates of EPest compared to NASS. Beginning in the late 1990s and early 2000s, metolachlor use was being replaced by use of *S*-metolachlor. It is possible that this difference in estimates could be related to how metolachlor and *S*-metolachlor were surveyed and reported. NASS estimates for metolachlor may have also included information for the related compound *S*-metolachlor. For example, beginning in 2002, EPest-low estimates of metolachlor use were zero for several states, such as Illinois and Iowa, while NASS reported several hundred pounds to over one million pounds of metolachlor use in these same states.

Soybeans

For various years from 1992 through 2000, 89 EPest-low and EPest-high estimates of metolachlor use on soybeans were compared with NASS estimates for 18 states from the Appalachian, Corn Belt, Delta, Lake States, Northeast, and Northern Plains regions. Only EPest-high estimates significantly differed ($p < 0.05$) from NASS estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 8 and 19 percent greater, respectively, indicating a general tendency for EPest estimates to be greater than NASS estimates. The correlation coefficients for EPest-low and NASS comparisons were 0.76 and were 0.75 for EPest-high. The relation between EPest-low and NASS estimates of metolachlor use on soybeans are shown in *figure 20A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 20B*.

The majority (71 of 89) of EPest and NASS estimates differed by less than a factor of two (*fig. 20B*). EPest estimates for most regions tended to be greater than NASS estimates, but in the Appalachian region, they tended to be less than NASS estimates.

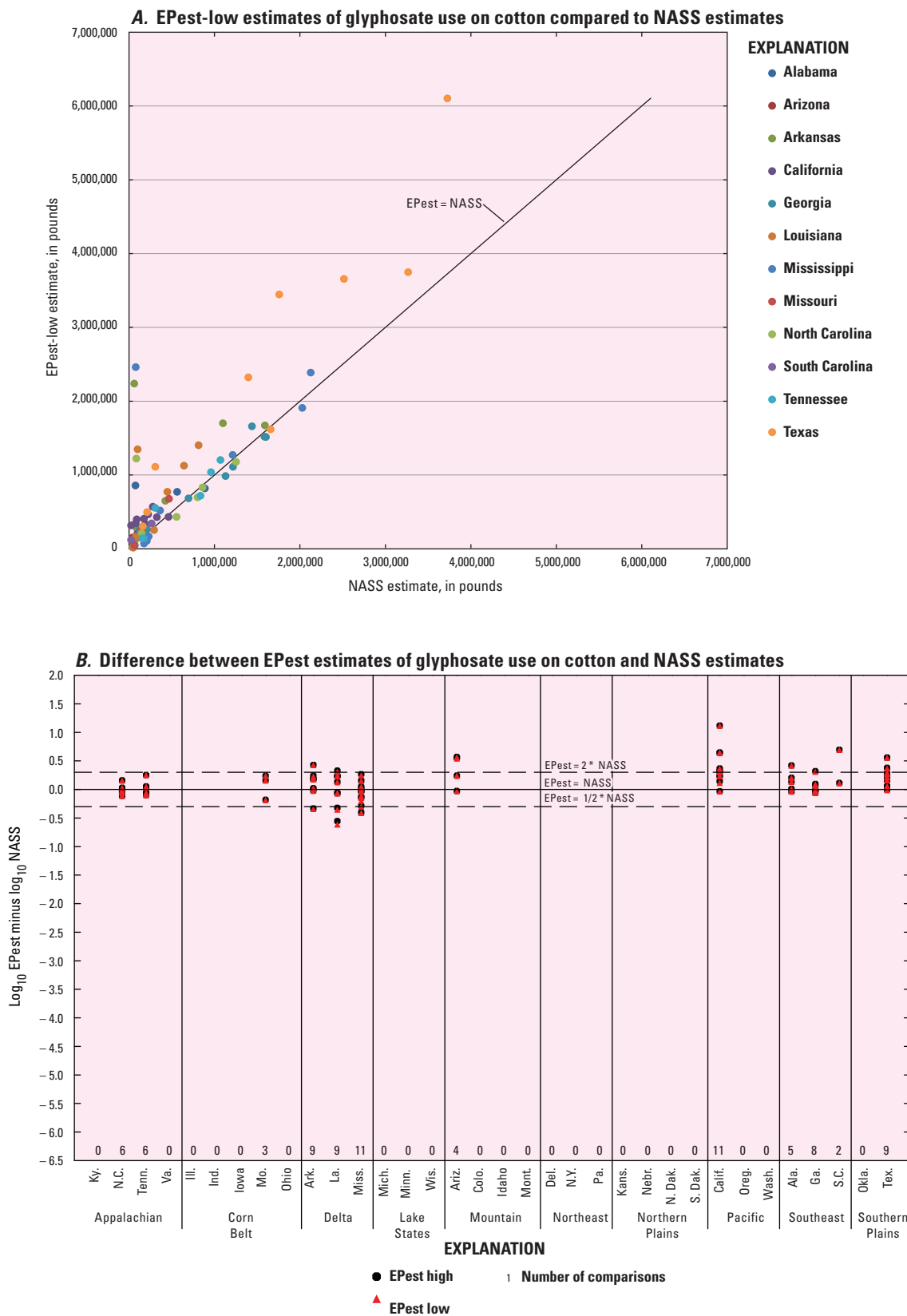


Figure 18. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of glyphosate use on cotton: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

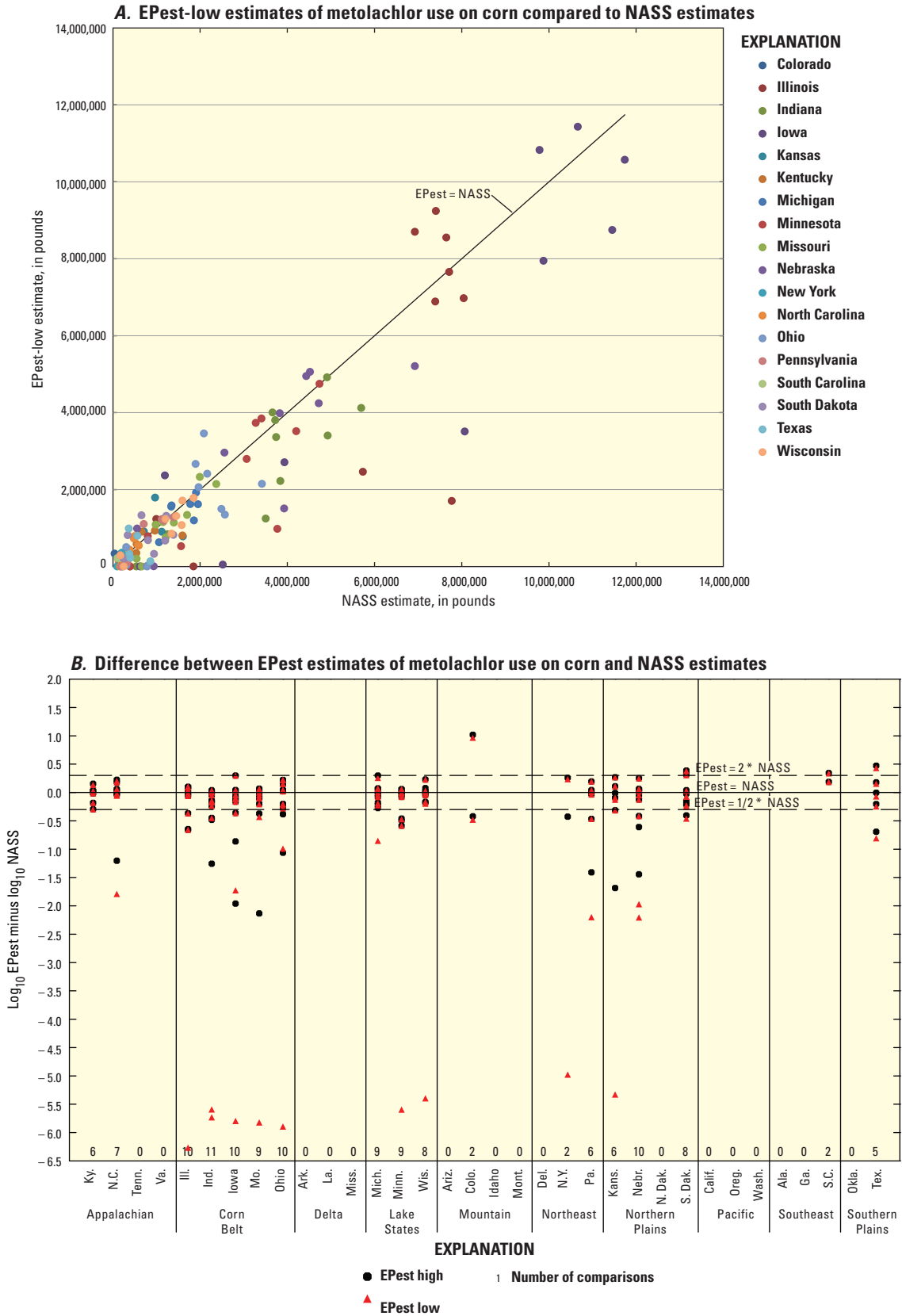


Figure 19. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of metolachlor use on corn: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

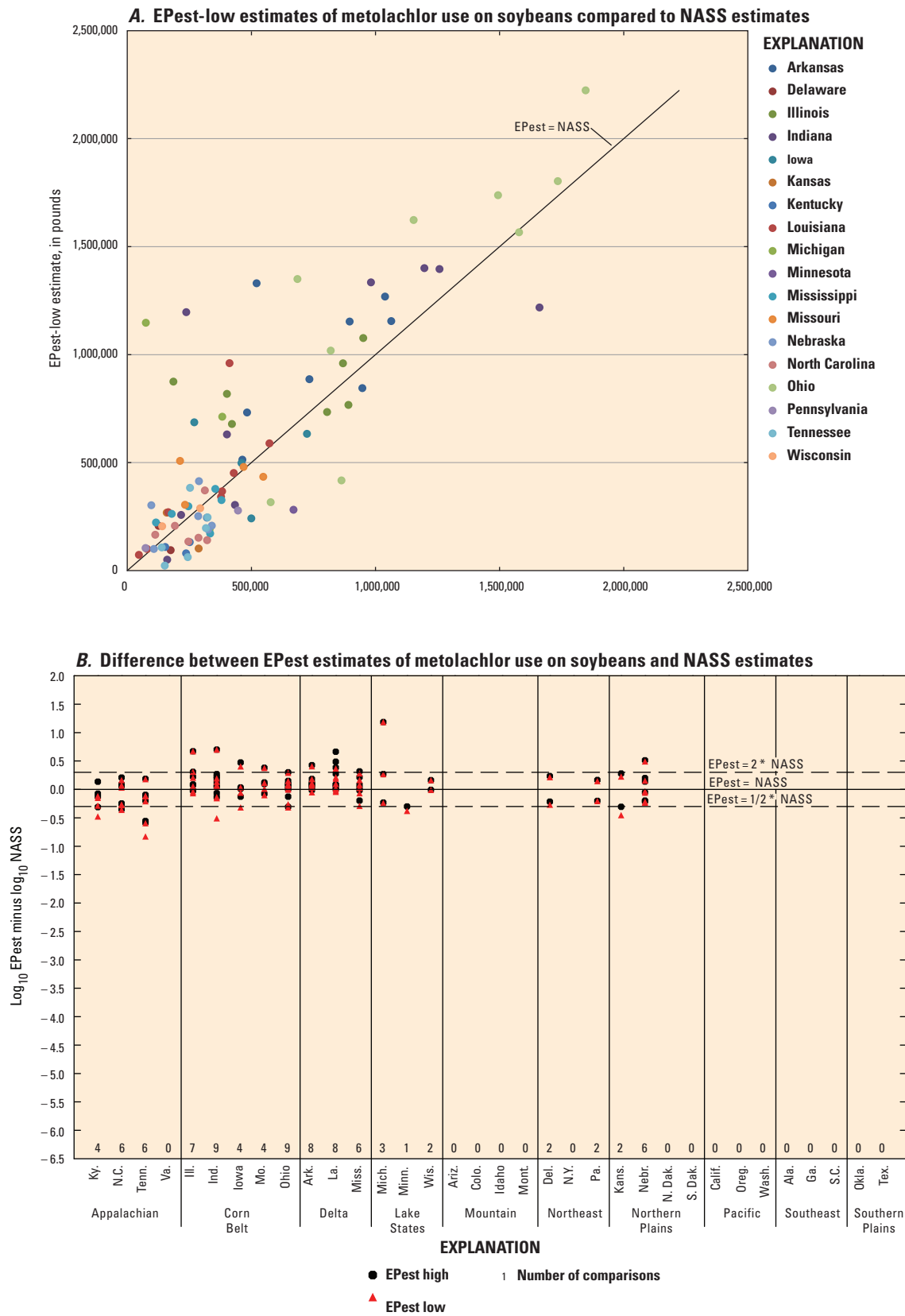


Figure 20. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of metolachlor use on soybeans: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

Metribuzin

For various years from 1992 through 2006, 108 EPest-low and EPest-high estimates of metribuzin use on soybeans were compared with NASS estimates in 19 states located in the Appalachian, Corn Belt, Delta, Lake States, Northeast, and Southeast regions. Only EPest-high estimates were significantly different ($p < 0.05$) from NASS estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 12 and 18 percent greater, respectively, indicating a general tendency for EPest estimates to be slightly greater than NASS estimates. Correlation coefficients for EPest-low and NASS comparisons were 0.81 and were 0.80 for EPest-high. The relation between EPest-low and NASS estimates of metribuzin use is shown in *figure 21A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 21B*.

The majority of EPest estimates were within a factor of two of NASS estimates (*fig. 21B*). EPest estimates for all of the regions bracketed NASS estimates, but estimates from Arkansas and Nebraska showed some large differences.

Nicosulfuron

For various years from 1992 through 2003, 127 EPest-low and EPest-high estimates of nicosulfuron use on corn were compared with NASS estimates for 20 states located in Appalachian, Corn Belt, Lake States, Mountain, Northeast, Northern Plains, Southeast, and Southern Plains regions. EPest-low and EPest-high estimates significantly differed ($p < 0.05$) from NASS estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 14 and 17 percent greater, respectively, indicating a general tendency for EPest estimates to be greater than NASS estimates. Correlation coefficients for EPest and NASS comparisons were 0.84 for both EPest-low and EPest-high. The relation between EPest-low and NASS estimates of nicosulfuron use on corn is shown in *figure 22A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 22B*.

Most of the EPest estimates were greater than NASS estimates, and the majority (98 of 127) of comparisons differed by less than a factor of two, although one or more EPest estimates from the Appalachian, Corn Belt, Lake States, Northeast, Northern Plains, and Southern Plains regions were at least twice NASS estimates. For some of the same states in these regions, however, EPest totals were half or less of NASS estimates.

S-Metolachlor

For 17 states from the Appalachian, Corn Belt, Lake States, Mountain, Northeast, Northern, and Southern Plains regions from 2001 through 2003, 39 EPest-low and EPest-high estimates of *S*-metolachlor use on corn were compared with NASS estimates. Only EPest-high estimates significantly differed ($p < 0.05$) from NASS estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 8 and 16 percent greater, respectively, indicating a general tendency for EPest estimates to be slightly greater than NASS estimates. Correlation coefficients for EPest-low and NASS comparisons were 0.90 and were 0.91 for EPest-high. The relation between EPest and NASS estimates of *S*-metolachlor use is shown in *figure 23A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 23B*.

EPest and NASS estimates for the majority (36 of 39) of states and years were within a factor of two (*fig. 23B*). EPest estimates for the Corn Belt, Mountain, Northern Plains, and Southern Plains regions tended to be greater than NASS estimates, whereas EPest estimates for the Lake States and Northeast tended to be less than NASS estimates.

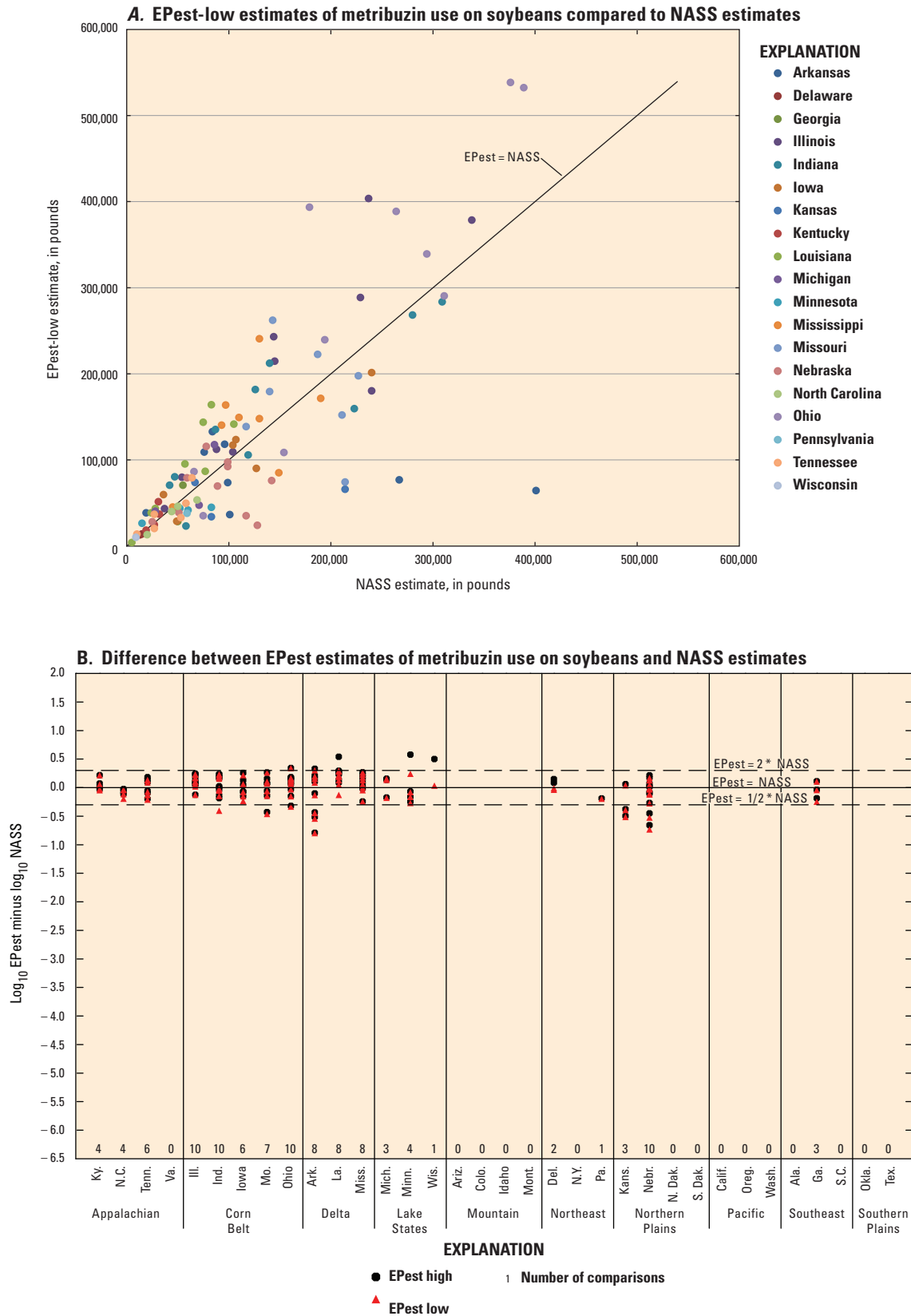


Figure 21. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of metribuzin use on soybeans: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

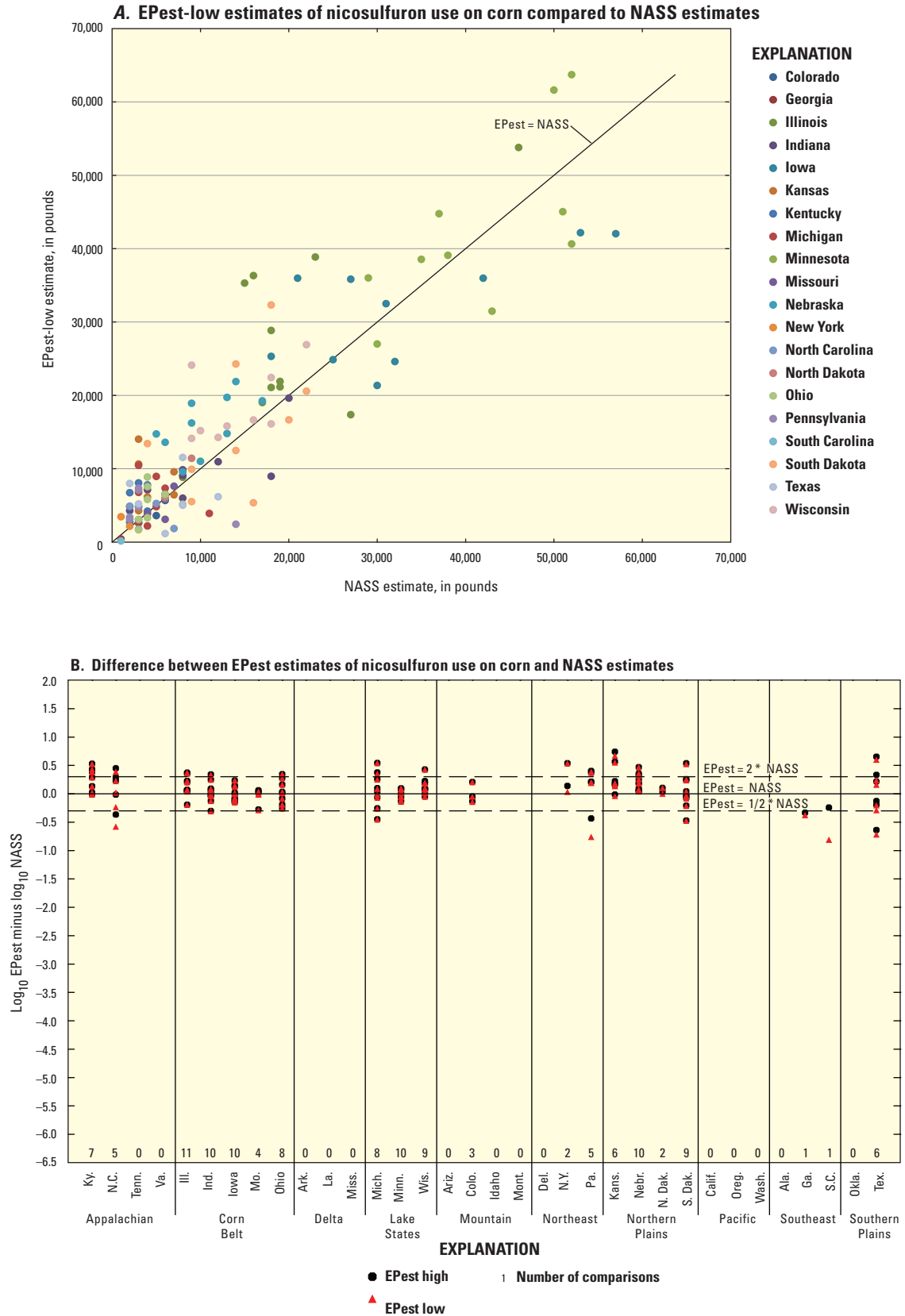


Figure 22. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of nicosulfuron use on corn: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

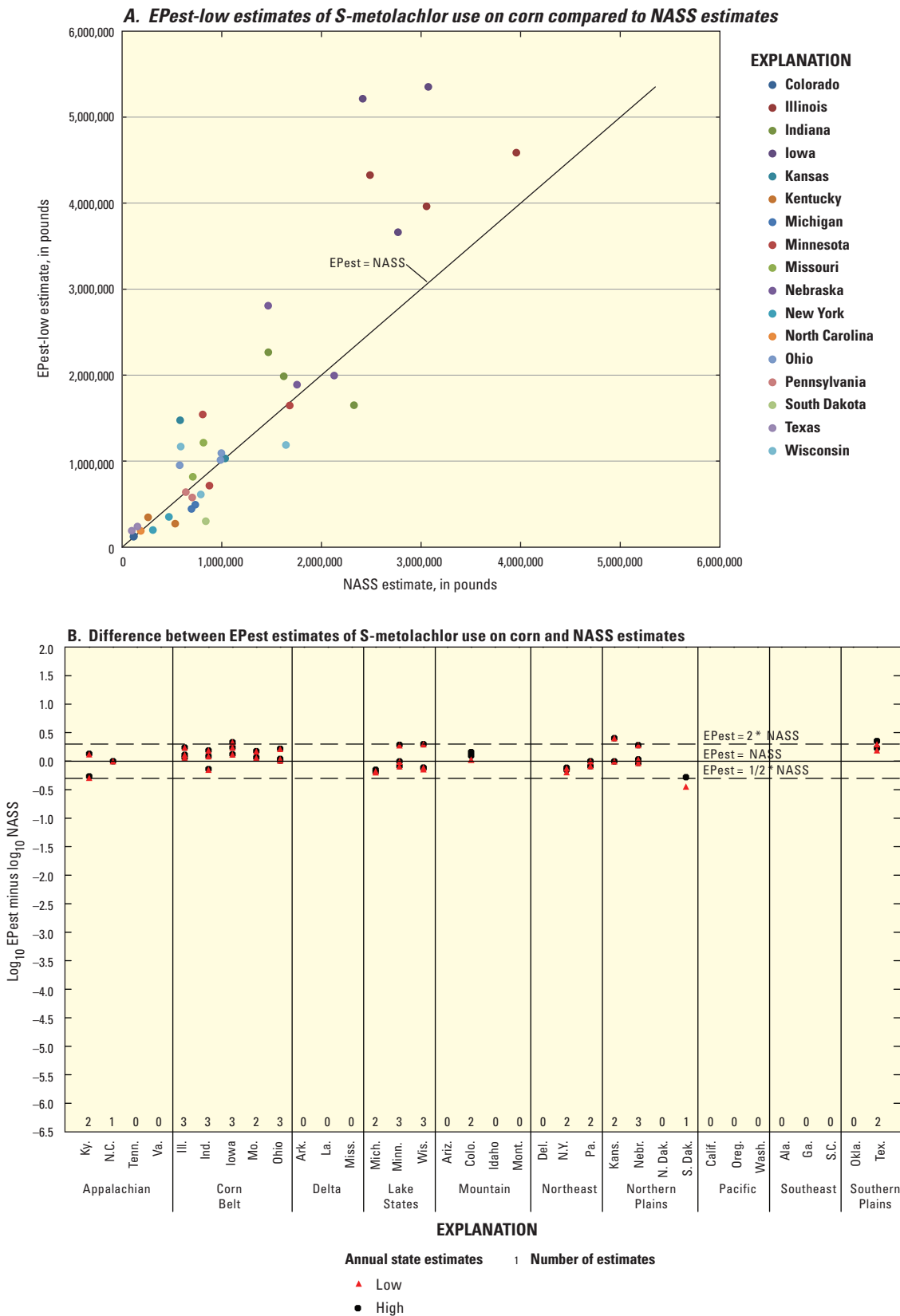


Figure 23. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of S-metolachlor use on corn: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

Trifluralin

Cotton

For various years from 1992 through 2005, 90 EPest-low and EPest-high estimates of trifluralin use on cotton were compared with NASS estimates for 12 states from the Appalachian, Corn Belt, Mountain, Pacific, Southeast, and Southern Plains regions. Only EPest-high estimates significantly differed ($p < 0.05$) from NASS estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 6 and 10 percent greater, respectively, indicating a general tendency for EPest estimates to be slightly greater than NASS estimates. Correlation coefficients for EPest and NASS comparisons were 0.95 for both EPest-low and EPest-high. The relation between EPest-low and NASS estimates of trifluralin use on cotton is shown in *figure 24A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 24B*.

The majority of EPest estimates differed from NASS estimates by less than a factor of two. The EPest estimates for most of the states in a particular region were evenly distributed around NASS use estimates. The strong correlation between estimates was driven by use estimates in Texas, which showed the least differences between EPest and NASS estimates of all the states.

Soybeans

For various years from 1992 through 2006, 97 EPest-low and EPest-high estimates of trifluralin use on soybeans were compared for 18 states from the Appalachian, Corn Belt, Delta, Lake States, Northeast, Southeast, and Northern Plains regions. Only EPest-high estimates significantly differed ($p < 0.05$) from NASS estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 3 and 7 percent greater, respectively, indicating a general tendency for EPest estimates to be slightly greater than NASS estimates. Correlation coefficients for EPest and NASS comparisons were 0.91 for both EPest-low and EPest-high. The relation between EPest-low and NASS estimates of trifluralin use on soybeans is shown in *figure 25A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 25B*.

The majority of EPest and NASS estimates were within a factor of two. One or more EPest and NASS estimates from every region except the Northern Plains differed by more than a factor of two. Iowa had greater trifluralin use on soybeans than other states.

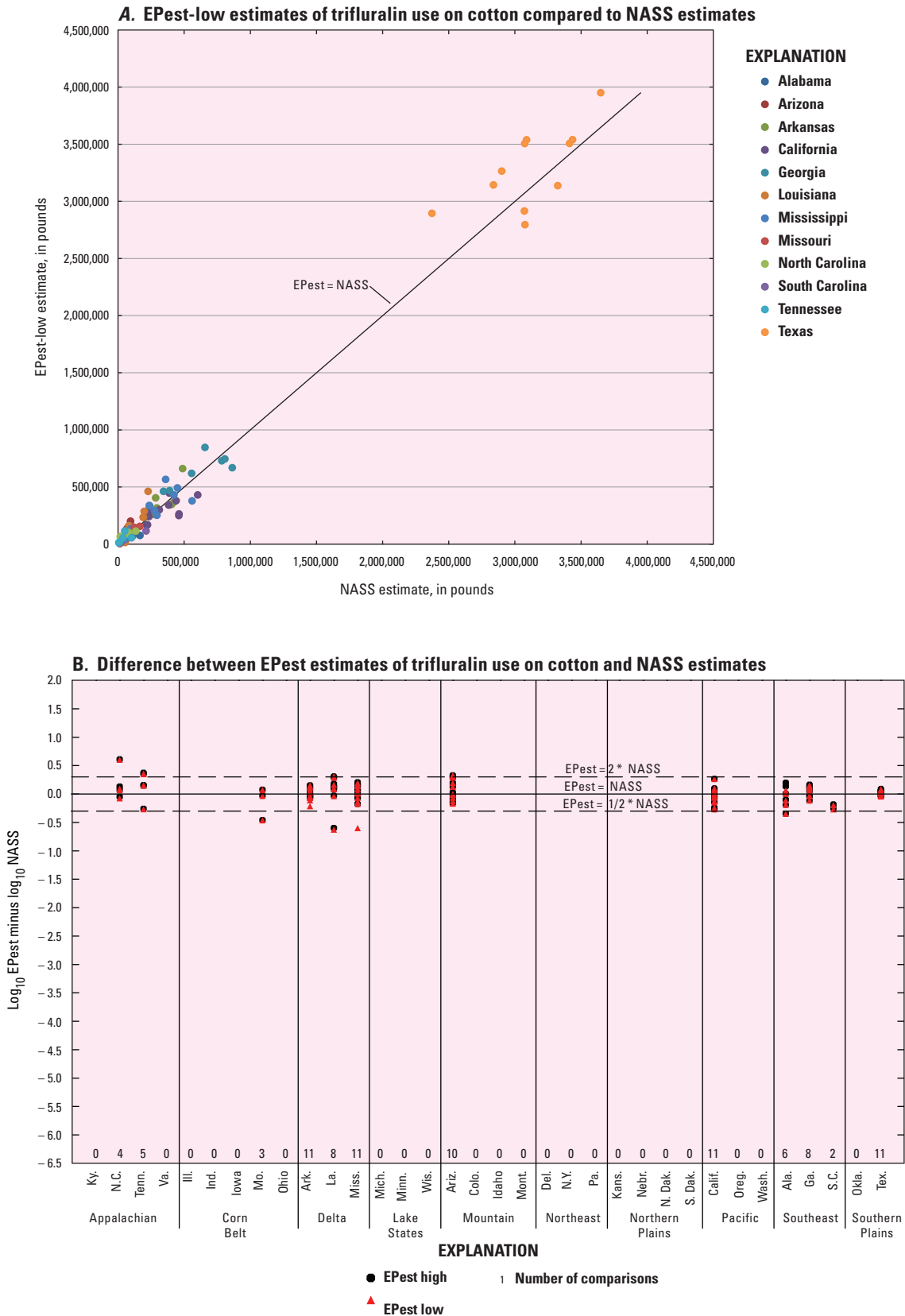


Figure 24. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of trifluralin use on cotton: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

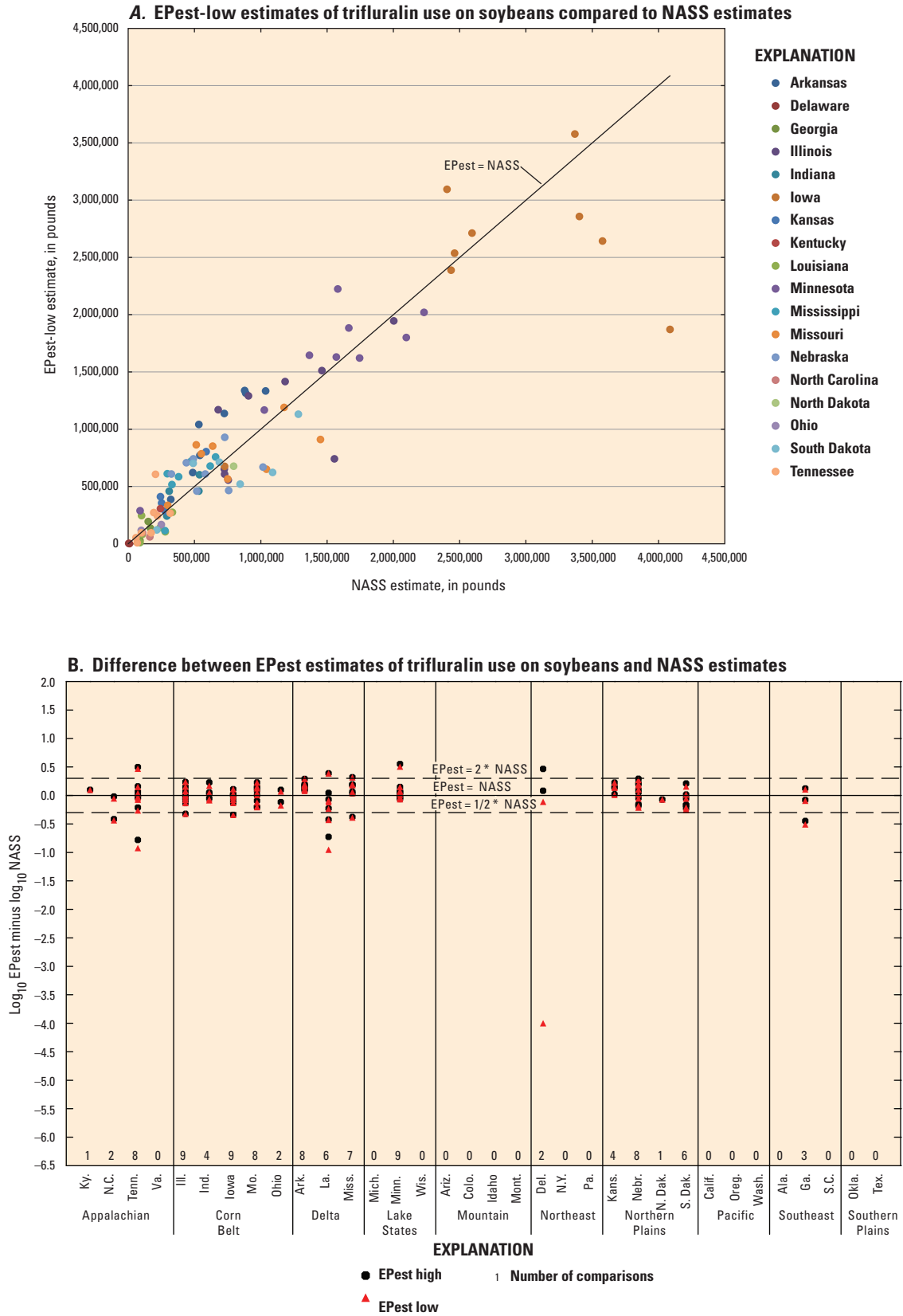


Figure 25. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of trifluralin use on soybeans: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

Insecticide Estimate Comparisons between EPest and NASS

EPest and NASS estimates were compared for seven insecticides used on corn, cotton, or both, as summarized in *tables 3 and 4*. Only 2 of the 10 insecticide comparisons had sample numbers greater than 50; both of these were not significant and had RE values of 0.1 or less, indicating agreement between the estimates. Most of the other comparisons were not significant and had RE values of 0.15 or less, but methomyl and methyl parathion estimates for cotton significantly differed and had RE values greater than 0.6, which are discussed in the following sections.

Methomyl

For various years from 1992 through 2003, 27 EPest-low and EPest-high estimates of methomyl use on cotton were compared with NASS estimates for 9 states from the Appalachian, Delta, Mountain, Pacific, Southeast, and Southern Plains regions. Only EPest-low estimates significantly differed ($p < 0.05$) from NASS estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 61 and 46 percent less, respectively, indicating a general tendency for EPest estimates to be less than NASS estimates. Correlation coefficients for EPest-low and NASS comparisons were 0.76 and were 0.74 for EPest-high. The relation between EPest-low and NASS estimates of methomyl use on cotton is shown in *figure 26A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 26B*.

More than half of the EPest estimates were less than 50 percent of NASS estimates, although one EPest estimate from Arkansas was more than double the NASS estimate. The few EPest and NASS estimates for California, Georgia, and Texas were in closer agreement than the estimates for other states.

Methyl Parathion

For various years from 1992 through 2005, 50 EPest-low and EPest-high estimates of methyl parathion use on cotton were compared with NASS estimates for 8 states from the Appalachian, Corn Belt, Delta, Mountain, Southeast, and Southern Plains regions. Both EPest-low and EPest-high estimates significantly differed ($p < 0.05$) from NASS estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 78 and 69 percent less, respectively, indicating a general tendency for EPest estimates to be less than NASS estimates. Correlation coefficients for EPest-low and NASS comparisons were 0.47 and were 0.52 for EPest-high. The relation between EPest-low and NASS estimates of methyl parathion use on cotton is shown in *figure 27A*, and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 27B*.

Most EPest and NASS estimates (EPest-low 37 of 50 and EPest-high 34 of 50) differed by more than a factor of two. The majority of EPest-low and EPest-high estimates were less than half NASS estimates, but, conversely, some EPest totals were at least twice NASS estimates. Generally, agreement between the estimates for methyl parathion was poor, and the RE was among the largest of all of the pesticides compared.

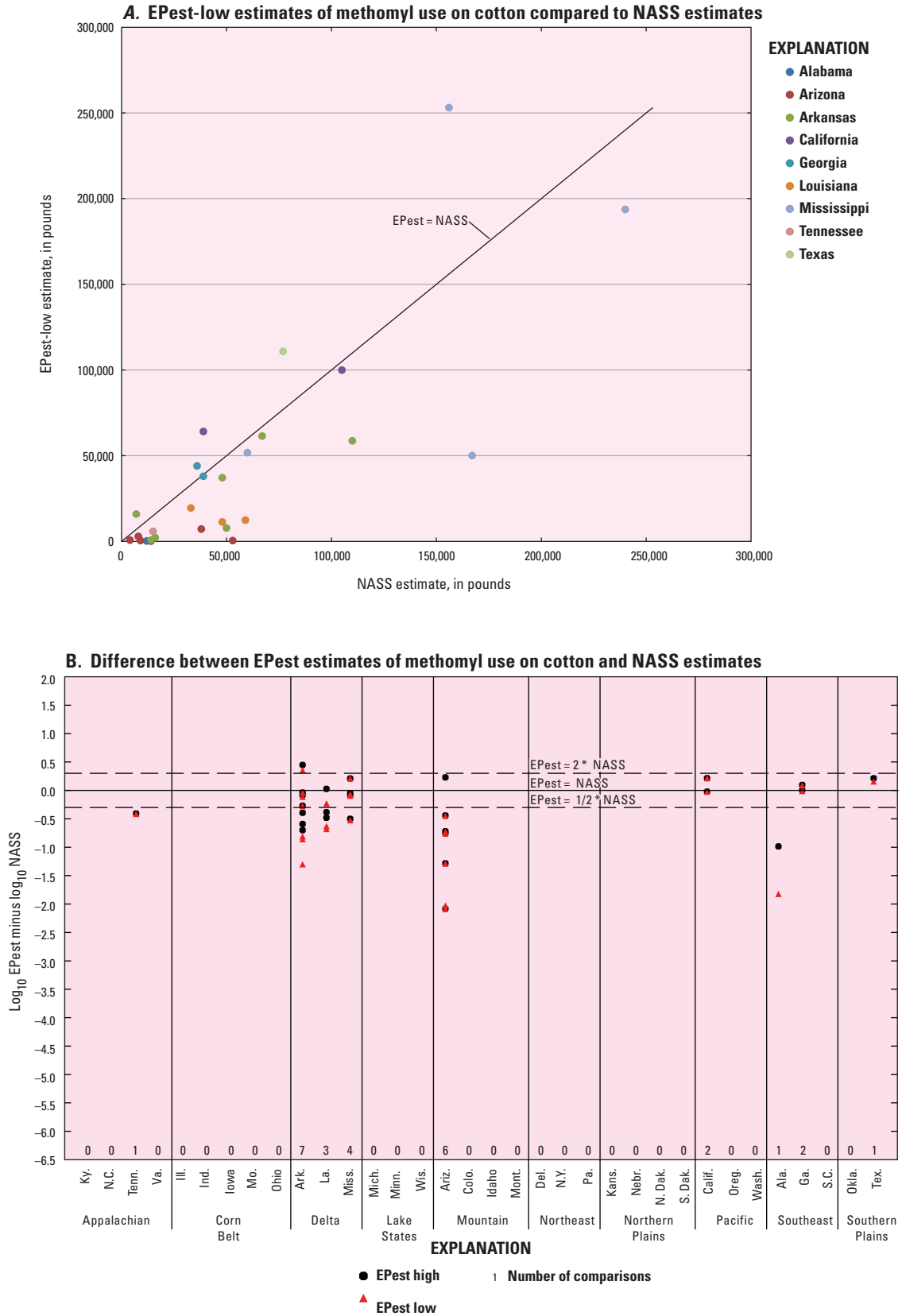


Figure 26. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of methomyl use on cotton: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

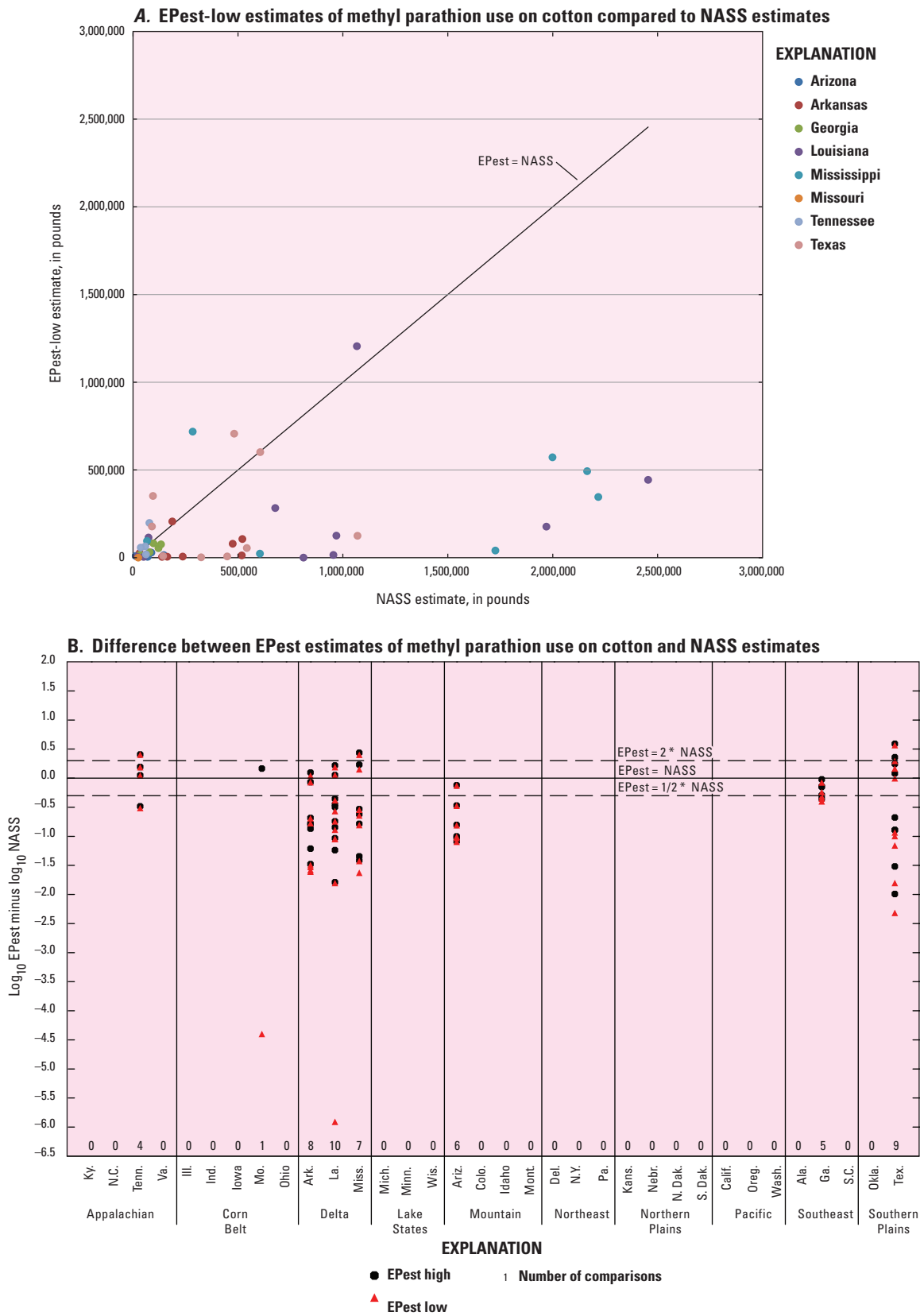


Figure 27. Comparison of EPest and National Agricultural Statistics Service (NASS) state estimates of methyl parathion use on cotton: (A) EPest-low estimates compared to NASS estimates, and (B) Difference between EPest estimates and NASS estimates ($\log_{10} \text{EPest} - \log_{10} \text{NASS}$).

Fungicide Estimate Comparisons between EPest and NASS—Propiconazole

For various years from 1993 to 2006, 14 EPest-low and EPest-high estimates of propiconazole use on winter wheat were compared with NASS estimates for 5 states from the Corn Belt, Lake States, Northern Plains, and Pacific regions. Only EPest-high estimates significantly differed ($p < 0.05$) from NASS estimates. The medians of the RE distributions comparing EPest-low and EPest-high to NASS estimates were 27 and 92 percent greater, respectively, indicating a general tendency for EPest estimates to be greater than NASS

estimates. Correlation coefficients for EPest-low and NASS comparisons were 0.78 and were 0.65 for EPest-high. The relation between EPest and NASS estimates of propiconazole use is shown in *figure 28A* (low) and *28B* (high), and the differences between NASS estimates and both EPest-low and EPest-high estimates are shown by region and state in *figure 28C*.

About half of the EPest-low and EPest-high estimates differed from NASS estimates by less than a factor of two. Almost all EPest-high estimates were greater than NASS estimates, whereas more than half of the EPest-low estimates were lower than NASS estimates.

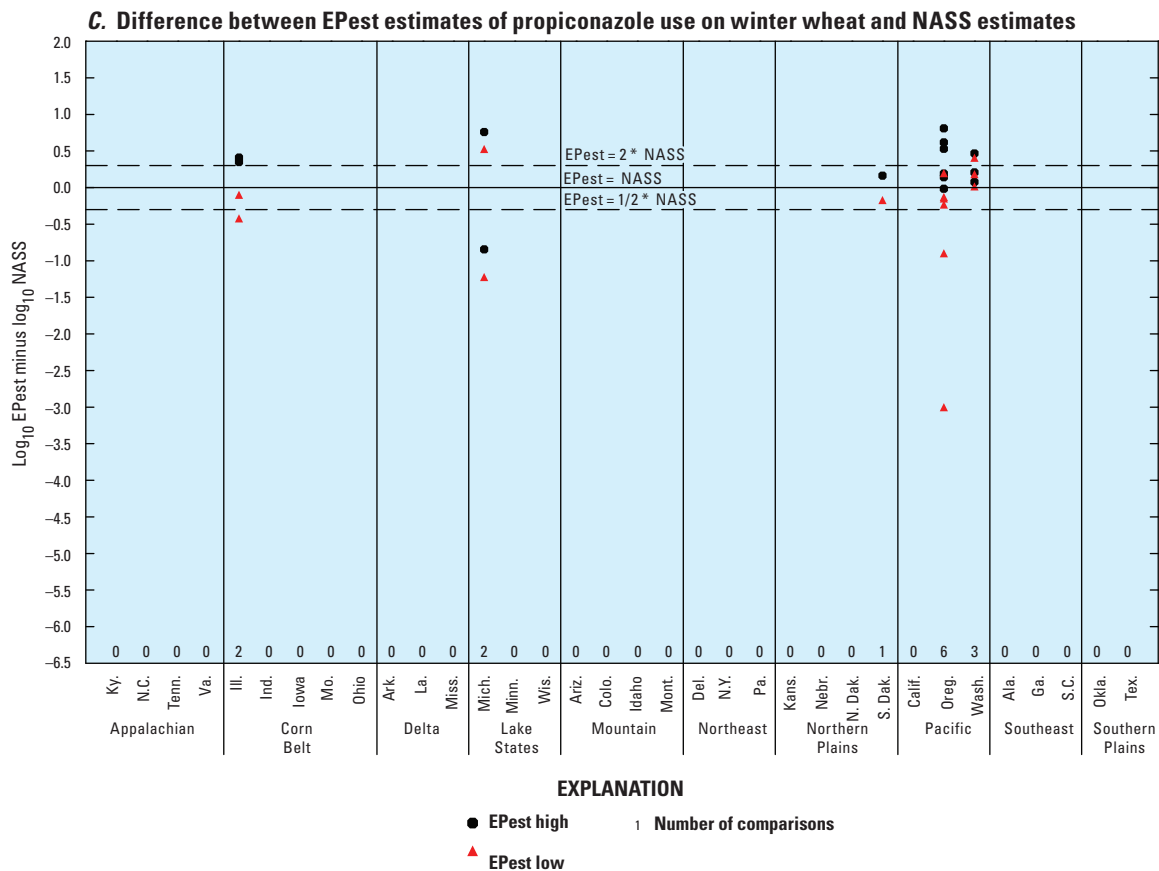
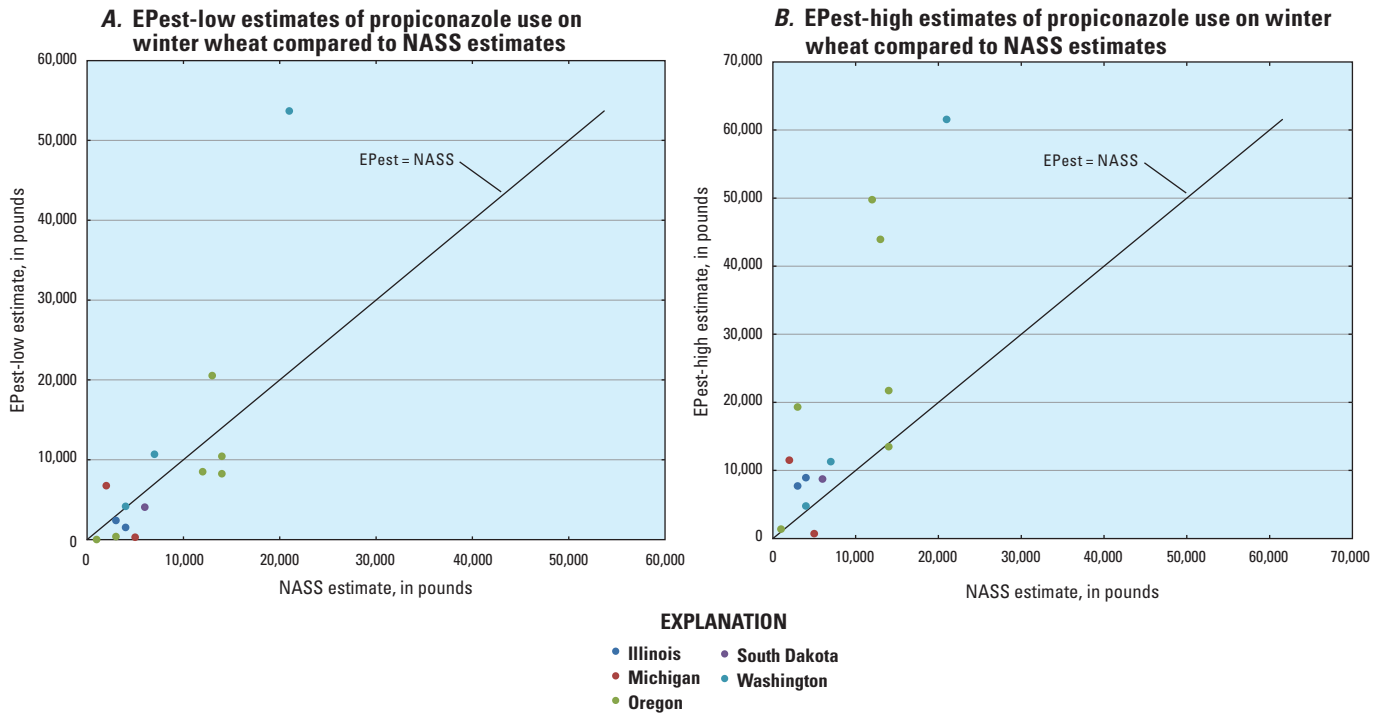


Figure 28. Comparison of Epest and National Agricultural Statistics Service (NASS) state estimates of propiconazole use on winter wheat: (A) Epest-low estimates compared to NASS estimates, (B) Epest-high estimates compared to NASS estimates, and (C) Difference between Epest estimates and NASS estimates ($\log_{10} \text{Epest} - \log_{10} \text{NASS}$).

Summary of Comparisons

EPest and NASS state estimates for as many as 34 states from 10 USDA Farm Production Regions were compared for 48 pesticide-by-crop combinations for various years from 1992 through 2006. These comparisons included 21 herbicides used on corn, cotton, rice, soybeans, spring wheat, or winter wheat; 7 insecticides used on corn or cotton; and 1 fungicide used on winter wheat.

Overall, 73 percent of the EPest-low to NASS comparisons for herbicide-by-crop (27 of 37) and 60 percent of the comparisons for insecticide-by-crop (6 of 10) had medians of the RE distributions within 0.15. About 22 percent of the herbicide-by-crop (8 of 37) and 40 percent of the insecticide-by-crop (4 of 10) EPest-low to NASS comparisons had medians of the RE distributions that indicated EPest-low estimates tended to be lower than NASS estimates. Only two herbicide-by-crop EPest-low to NASS comparisons, but none of the insecticide-by-crop comparisons, had medians of the RE distributions that indicated EPest-low estimates tended to be greater than NASS estimates.

There was somewhat less agreement between EPest-high and NASS estimates. About 60 percent of the EPest-high to NASS comparisons for herbicide-by-crop and 30 percent of the comparisons for insecticide-by-crop had median of the RE distributions within 0.15. About 16 percent of the herbicide-by-crop and 10 percent of the insecticide-by-crop EPest-high to NASS comparisons had medians of the RE distributions that indicated EPest-high estimates tended to be less than NASS estimates. About 22 percent of the herbicide-by-crop and 60 percent of the insecticide-by-crop EPest-high to NASS comparisons had medians of the RE distributions that indicated EPest-high tended to be greater than NASS estimates.

Overall, the comparisons between EPest and NASS estimates generally support the representativeness and use of the EPest method to estimate pesticide use. Most EPest and NASS estimates for the same pesticides, crops, years, and states were not significantly different from each other. EPest and NASS estimates were produced from different surveys of individual farm operations, and the methods used to expand the surveyed data to estimate state use also differed; therefore, some disagreement in the estimates is expected.

Applications of EPest Use Data

Estimates of pesticide use developed by this study provide information on the amounts, distribution, and trends in agricultural use of 39 pesticides for 1992 through 2009. Maps showing the geographic distribution of estimated average annual pesticide use intensity in each county of the conterminous United States and a graph showing each

pesticide's national use-trend from 1992 through 2009 are provided at <http://water.usgs.gov/nawqa/pnsp/usage/maps/>.

The pesticide-use intensity estimates shown on the maps were calculated by dividing the pounds of pesticide applied annually to each county by the area of agricultural land (in square miles) in the county. These annual-use rates were applied to the satellite-based 2009 Cropland Data Layer (CDL) produced by the USDA (Johnson and Mueller, 2010). The CDL is a crop-specific land-cover dataset mapped at 56-meter resolution. Each 56-meter cell is assigned to one of over 100 agricultural or nonagricultural land-use classes. For the purpose of mapping pesticide-use intensity, the CDL was generalized into 1-kilometer cells. First, the CDL was divided equally into 1-meter cells and then it was converted into a binary raster with each cell labeled as either agriculture or non-agriculture and assigned a value of 1 or 0, respectively. The 1-meter cells were next aggregated to 1-kilometer cells, and the percentage of agricultural or non-agricultural land use in the 1-kilometer cell was calculated. County pesticide-use estimates were then multiplied by the percentage of agricultural land in each cell.

The county-level estimates are suitable for making national, regional, statewide, and watershed assessments of annual pesticide use during 1992–2009. Although estimates are provided by county to facilitate estimation of watershed-use rates for a wide variety of watersheds, there is a high degree of uncertainty in individual county-level estimates because (1) pesticide-by-crop use rates were developed on the basis of pesticide use on harvested acres in multi-county areas (CRDs) and then allocated to county harvested cropland; (2) pesticide-by-crop use rates were not available for all CRDs in the conterminous United States, and extrapolation methods were used to estimate pesticide use for some counties; and (3) it is possible that surveyed pesticide-by-crop use rates do not reflect all agricultural uses or crops grown.

For water-quality studies, estimates of pesticide use within watersheds and groundwater recharge areas can be used to assist with study design and to help explain and model pesticide occurrence in water resources. Information on pesticide use and other watershed characteristics serve as explanatory variables in regression models developed to predict concentrations of pesticides in streams and groundwater (Barbash and others, 2001; Stackelberg and others, 2006; Stone and Gilliom, 2009). Pesticide-use information has also been used to explain the atmospheric transport of agricultural chemicals from the area the pesticides were applied to other sites where they are detected in air and rain samples (Majewski and others, 1998). The availability of pesticide-use information for the 18-year study period enables assessments of the temporal and spatial variations in pesticide use that can relate these patterns to changes in water quality (Sullivan and others, 2009). The methods developed in this study are applicable to other agricultural pesticides and years.

Summary and Conclusions

A method was developed to estimate pesticide use (EPest) for 39 pesticides used on a variety of row crops, fruit, nut, and specialty crops grown throughout the conterminous United States for 1992 through 2009. EPest pesticide-by-crop rates were developed for individual crops on the basis of (1) surveyed pesticide-use reports from farm operations within CRDs and (2) harvested crop acreage reported by USDA Census of Agriculture and NASS annual crop surveys. EPest rates were developed for all crops that were surveyed in a particular year by dividing the pounds of a pesticide applied to each crop grown in the CRD by the harvested acreage for that crop. Not all crops were surveyed in each year and CRD; therefore, extrapolated rates for non-surveyed CRDs, referred to as tier 1, tier 2, and regional EPest rates, were developed by using information from adjacent CRDs.

The EPest rates were applied to county harvested-crop acreage differently for surveyed CRDs with unreported pesticide-by-crop estimates to produce EPest-low and EPest-high estimates of pesticide use for every year from 1992 through 2009. If a CRD was surveyed, but there was no reported pesticide use, then the EPest-low method did not estimate pesticide use for the CRD; EPest-high treated these non-reported estimates as unsurveyed, and pesticide use was estimated on the basis of an EPest extrapolated rate. For both methods, if a CRD was not surveyed, then pesticide use was estimated by using EPest extrapolated rates, if possible.

About 45 percent of the national EPest-low and EPest-high annual pesticide-by-year estimates differed from one another by less than 25 percent, including the estimates for several of the most widely used pesticides, such as acetochlor, atrazine, glyphosate, and metolachlor. EPest-high estimates, however, were more than double EPest-low totals for six or more years for the pesticides alachlor, butylate, carbofuran, cyanazine, ethoprophos, linuron, methyl parathion, metolachlor, pebulate, propachlor, and terbacil. EPest extrapolated rates used to calculate EPest-high estimates contributed a significant amount to the national total for some pesticides and years for some specialty crops and major crops, such as corn and alfalfa, and land uses, such as summer fallow, pasture, and rangeland. In general, non-surveyed use represented a greater percentage of the national estimate for some pesticides and crops because some pesticides were reported less frequently and some crops were not surveyed as extensively during the latter part of the study. EPest tier 1, tier 2, and regional rates have inherently greater uncertainty than rates for surveyed CRDs because a pesticide could

have been applied to a localized area in response to a pest infestation, while the same crop grown in another part of the same region would not be managed in the same way, which can result in misrepresentative estimates of pesticide use.

National and state annual estimates for a subset of the 39 pesticides were compared with data published by other sources. EPest-low and EPest-high national estimates for seven herbicides were compared with published data from the USEPA, NASS, and NPUD for three periods (1997, 2001–02, and 2006–07). Overall, there was agreement between EPest estimates and the estimates from USEPA and NPUD; however, EPest estimates tended to be greater than NASS estimates, which are not complete national estimates.

A second set of evaluations compared EPest state and state-by-crop estimates for selected pesticides with NASS estimates. State estimates for 33 pesticides that had 5 or more estimates for a combination of states, crops, or years were evaluated, in addition to the estimates for 29 pesticides that had 10 or more state and year estimates for corn, cotton, soybeans, spring wheat, or winter wheat. Of the 33 pesticides evaluated, less than one-third—10 EPest-low and 8 EPest-high—had median RE values significantly different from zero based on the 95-percent confidence interval on the median. EPest-high estimates were mostly greater than NASS estimates when they differed significantly, whereas EPest-low estimates were more evenly distributed around NASS estimates when they differed significantly.

EPest and NASS estimates for individual states and crops were compared for selected years from 1992 to 2006. This comparison was made for 48 pesticide-by-crop combinations, including 21 herbicides, 7 insecticides, and 1 fungicide used on corn, cotton, soybeans, rice, spring wheat, or winter wheat. Most EPest and NASS pesticide-by-crop estimates were not significantly different, had low median relative errors ($RE < 0.15$), and had relatively strong correlation coefficients ($r > 0.75$). EPest-low and EPest-high state estimates for some pesticide-by-crop combinations, however, were significantly different ($p < 0.5$) from NASS estimates. Among the pesticide-by-crop estimate comparisons, those that did show a significant difference between EPest and NASS estimates did not show clear or consistent patterns by pesticide type, crop, year, or state. EPest and NASS estimates were produced from different surveys of individual farm operations, and the methods used to expand the surveyed data to estimate state use also differed; therefore, some disagreement in the estimates is expected. The comparisons between EPest and NASS estimates generally support the representativeness and use of the EPest method to estimate pesticide use.

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Appendix 1. Summary of Epest-Low and Epest-High Annual National Totals by Pesticide and Crop Type.

Appendix 1 is available in Microsoft Excel® format at <http://pubs.usgs.gov/sir/2013/5009/appendix1.xlsx>

Appendix 2. Epest-Low and Epest-High Annual National Totals Derived from Epest Surveyed, Tier 1, Tier 2, and Regional Rate Estimates.

Appendix 2 is available in Microsoft Excel® format at <http://pubs.usgs.gov/sir/2013/5009/appendix2.xlsx>

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