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Pesticide Residues in Urine of Adults Living in the United States: Reference Range Concentrations[1]

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We measured 12 analytes in urine of 1000 adults living in the United States to establish reference range concentrations for pesticide residues. We frequently found six of these analytes: 2,5-dichlorophenol (in 98% of adults); 2,4-dichlorophenol (in 64%); 1-naphthol (in 86%); 2-naphthol (in 81%); 3,5,6-trichloro-2-pyridinol (in 82%); and pentachlorophenol (in 64%). The 95th percentile concentration (95thPC) for 2,5-dichlorophenol (indicative of *p*-dichlorobenzene exposure) was 790 μg/liter; concentrations ranged up to 8700 μg/liter. 2,4-Dichlorophenol concentrations ranged up to 450 μg/liter, and the 95thPC was 64 μg/liter. 1-Naphthol and 2-naphthol (indicative of naphthalene exposure) had 95thPCs of 43 and 30 μg/liter, respectively; concentrations of 1-naphthol ranged up to 2500 μg/liter. Chlorpyrifos exposure was indicated by 3,5,6-tricholoro-2-pyridinol concentrations of 13 (95thPC) and 77 μg/liter (maximum observed). Pentachlorophenol had a 95thPC of 8.2 μg/liter. Other analytes measured included 4-nitrophenol (in 41%); 2,4,5-trichlorophenol (in 20%); 2,4,6-trichlorophenol (in 9.5%); 2,4-dichlorophenoxyacetic acid (in 12%); 2-isopropoxyphenol (in 6.8%); and 7-carbofuranphenol (in 1.5%). The 95thPCs of these analytes were <6 μg/liter. *p*-Dichlorobenzene exposure is ubiquitous; naphthalene and chlorpyrifos are also major sources of pesticide exposure. Exposure to chlorpyrifos appears to be increasing. Although pentachlorophenol exposure is frequent, exposure appears to be decreasing. These reference range concentrations provide information about pesticide exposure and serve as a basis against which to compare concentrations in subjects who may have been exposed to pesticides.

INTRODUCTION

Pesticides have brought us great benefits by improving our food supply, controlling harmful pests, and improving our quality of living; however, the widespread use of pesticides is not without risks for our environment and our health. In 1991 the United States used 2.7 billion pounds of pesticides—approximately 10 pounds of pesticide for every man, woman, and child living in the United States (EPA, 1991). It is virtually impossible for anyone to avoid exposure to low levels of several different pesticides in our environment (Morgan, 1992). Because pesticides have potential to cause harm in high doses, the public is concerned about pesticides' unknown chronic health effects at low levels. By using environmental measurements, we can estimate the extent and magnitude of this widespread exposure; however, estimates are often difficult or impossible on an individual basis because the sources and routes of exposure for individuals are frequently unknown. Assessments of exposure from multiple sources and routes are better made by measuring pesticides or their metabolites in human specimens because these measurements more nearly reflect total exposure from all routes of exposure (Needham, 1994). Such measurement of analytes in human specimens is known as biological monitoring, and the analytes are often called biomarkers of exposure.

We often develop and apply biological monitoring

[1] Samples were collected from human subjects who participated in the Third National Health and Nutrition Examination Survey (NHANES III) conducted by the National Center for Health Statistics. All protocols were reviewed and approved by appropriate human subjects review committees. These protocols complied with all national and institutional guidelines for the protection of human subjects.

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TABLE 1

Analyte or Pesticide Residue Measured and Possible Pesticides or Parent Compounds That May Result in These Residues

Analyte or pesticide residue	Possible parent compound(s)
Carbofuranphenol [2,3-dihydro-2,2-dimethyl-7-hydroxybenzofuran] (CFP)	Benfuracarb; carbofuran; carbosulfan; furathiocarb
2,4-Dichlorophenol (24DCP)	Bifenox[a]; chlomethoxyfen[a]; 2,4-D[a]; 2,4-DB[a]; dichlofenthion[a,b]; diclofop[a]; 1,3-dichlorobenzene; dichlorprop[a]; nitrofen[a,b]; phosdiphen; prothiofos
2,5-Dichlorophenol (25DCP)	1,4-Dichlorobenzene
2,4-Dichlorophenoxyacetic acid (24D)	2,4-D
2-Isopropoxyphenol (IPP)	Propoxur
1-Naphthol (1NAP)	Naphthalene; carbaryl; napropamide[a]
2-Naphthol (2NAP)	Naphthalene; naproanilide[a]; (2-naphthyloxy)acetic acid[a]
4-Nitrophenol (4NP)	Chlornitrofen[a]; EPN; fluorodifen[a,b]; methyl parathion; 4-nitroanisole; nitrobenzene; nitrofen[a,b]; parathion
Pentachlorophenol (PCP)	Pentachlorophenol; pentachloronitrobenzene
2,4,5-Trichlorophenol (245TCP)	Fenchlophos[b]; lindane; pentachloronitrobenzene; pentachlorophenol; 1,2,4-trichlorobenzene; trichloronat[b]
2,4,6-Trichlorophenol (246TCP)	Chlornitrofen[a]; hexachlorobenzene; lindane; pentachloronitrobenzene; pentachlorophenol; prochloraz[a]; 1,3,5-trichlorobenzene
3,5,6-Trichloro-2-pyridinol (TCPY)	Chlorpyrifos; chlorpyrifos-methyl

[a] These pesticides contain an ether function that could cleave to produce the corresponding analyte or pesticide residue in column 1.
[b] These pesticides may be no longer manufactured or marketed (Tomlin, 1994).

methods in investigations and studies of populations with known or suspected exposure to pesticides. To help in the interpretation of the results of these investigations, we conducted laboratory studies to determine reference range concentrations for selected toxicants (i.e., pesticide residues) in human specimens. Reference range concentrations are defined as those concentrations of a specific metabolite or residue that we expect to be present in the general population that did not have occupational exposure to the parent compound. As will be seen below, however, this general population has clearly been exposed to some measurable extent, although in most cases the level is very low compared to most occupational exposures. The purpose of our study was to determine the reference range concentrations for selected pesticides or their residues in urine from the general population.

As part of our efforts to provide information about pesticide exposure, we developed a biological monitoring method to measure the 12 urinary pesticide residues in Table 1. These 12 compounds are potential products of a metabolic process that reflects exposure to more than 30 pesticides (Hill *et al.*, 1995a). By using this new method, we measured and now report the concentrations of these 12 pesticide residues in the urine of approximately 1000 adults living in the United States. These results provide a measure of the extent of exposure to these pesticides in the United States and can serve as reference range concentrations for studies involving potentially exposed populations, for risk assessments, for the monitoring of trends, and for other public health efforts. We have previously reported our rationale for the selection of these analytes and have described our analytical methodology in detail (Hill *et al.*, 1995a).

MATERIALS AND METHODS

Urine samples were collected from approximately 1000 adults, a subset of those participating in the National Health and Nutrition Examination Survey III, also known as NHANES III (NCHS, 1994). These adults, ranging in age from 20 to 59 years, were selected from a relatively broad spectrum of the U.S. population reflecting both sexes and different age groups, races/ethnicities, urban/rural residences, and regions of the country (Needham *et al.*, 1995).

The urinary pesticide residues were measured by the method of Hill *et al.* (1995a), which involved (1) the addition of carbon-13-labeled internal standards to 10 ml of urine; (2) enzymatic hydrolysis and then extraction with 1-chlorobutane/ether; (3) back-extraction with base and then treatment with 1-chloro-3-iodopropane to form chloropropyl deriva-

TABLE 2

Frequency with Which Pesticide Residues or Metabolites Are Detected in Urine, Their Mean Concentration, and Their Concentrations at Selected Population Percentiles for Adults Living in the United States[a]

Analyte	*N*	Frequency of detection (%)	Mean	5%	25%	50%	75%	90%	95%	99%	100%
CFP	993	1.5	<1	ND	ND	ND	ND	ND	ND	2.1	14
24DCP	988	64	14	ND	ND	2.2	9.1	36	64	240	450
25DCP	982	98	200	2.2	11	30	120	460	790	3500	8700
24D	983	12	<1	ND	ND	ND	ND	1.2	1.8	6.3	37
IPP	995	6.8	<1	ND	ND	ND	ND	ND	1.7	4.7	10
1N	983	86	17	ND	1.7	4.4	12	26	43	290	2500
2N	977	81	7.8	ND	1.2	3.4	9.9	21	30	54	88
4NP	974	41	1.6	ND	ND	ND	1.7	3.3	5.2	16	63
PCP	951	64	2.5	ND	ND	1.5	3.0	5.3	8.2	19	55
245TCP	934	20	1.0	ND	ND	ND	ND	1.8	3.0	8.3	25
246TCP	945	9.5	<2	ND	ND	ND	ND	ND	3.3	25	63
TCPY	993	82	4.5	ND	1.3	3.0	5.9	9.5	13	27	77

[a] Micrograms per liter or parts per billion (ppb).

tives; (4) cleanup and concentration for analysis; and (5) analysis by using capillary GC/MS/MS.[2] This method was highly specific and sensitive. The detection limit for all analytes in a 10-ml urine sample, except 246TCP,[1] was 1 μg/liter or 1 ppb; the detection limit for 246TCP was 2 μg/liter.

RESULTS

Table 1 lists the 12 analytes or pesticide residues and their possible parents. The list of possible parent pesticides is based upon available information on humans or animals or upon the examination of the structure of these pesticides and predictions about their possible metabolism. There is little or no information about human metabolism of many of these pesticides. Some of the possible parent pesticides may not be metabolized in significant amounts to that particular pesticide residue or analyte in humans. Several of these pesticides contain an ether functional group and have the potential to produce an analyte to pesticide residue only if that ether function is cleaved (see Table 1). Although there are reports that some of these pesticides produce phenolic metabolites in plants, soil, or some animals (Aizawa, 1982, 1989), studies of 24D metabolism in humans indicate that the ether cleavage products of some of these pesticides are unlikely to be major human metabolites (Kohli *et al.*, 1974). The British Crop Protection Council included four pesticides—dichlofenthion, fenchlophos, nitrofen, trichloronat—in their list of pesticides that are no longer manufactured or marketed (Tomlin, 1994).

Tables 2 and 3 report the results of the analyses for these selected pesticide residues in urine samples from approximately 1000 adults living in the United States. In Table 2 we show the number of adults [*N*], the frequency of detection, the mean urinary concentrations, and the urinary concentrations of each analyte at the 5th, 25th, 50th (median), 75th, 90th, 95th, 99th, and 100th percentiles (maximum observed) in micrograms per liter or parts per billion (ppb). Figure 1 shows a comparison of the frequencies of detection for all analytes. Figure 2 shows the 5th, 50th (median), and 95th percentile concentrations of all analytes. In Table 3 we present the creatinine-corrected mean urinary concentrations and the urinary concentrations at each percentile in micrograms per gram creatinine. The data in Table 3 reflect only those urine samples that had creatinine values ≥30 mg/dl; urine samples with creatinine values below 30 mg/dl are generally regarded as being too dilute to provide valid results (Lauwerys and Hoet, 1993). We have chosen to report both uncorrected concentrations (μg/liter or ppb) and creatinine-corrected concentrations (μg/g creatinine) to provide a broader based reference range for comparison. Although we have observed better correla-

[2] Abbreviations used: GC/MS/MS, gas chromatography with tandem mass spectrometry; 246TCP, 2,4,6-trichlorophenol; ppb, parts per billion; ppm, parts per million; 24DCP, 2,4-dichlorophenol; 1NAP, 1-naphthol; 2NAP, 2-naphthol; PCP, pentachlorophenol; TCPY, 3,5,6-trichloro-2-pyridinol; DCB, 1,4-dichlorobenzene; PAH, polycyclic aromatic hydrocarbons; NHANES II, National Health and Nutrition Examination Survey; 4NP, 4-nitrophenol; 245TCP, 2,4,5-trichlorophenol; IPP isopropoxyphenol; CFP, carbofuranphenol; 25 DCP, 2,5-dichlorophenol; 24D, (2,4 dichlorophenoxy)acetic acid; EPN, *O*-ethyl *O*-*p*-nitrophenyl phenylphosphonothioate.

TABLE 3

Mean Creatinine-Corrected Concentration and Creatinine-Corrected Concentrations at Selected Population Percentiles of Pesticide Residues or Metabolites in Urine for Adults Living in the United States[a]

Analyte	*N*	Mean	5%	25%	50%	75%	90%	95%	99%	100%
CFP	902	<1	ND	ND	ND	ND	ND	ND	1.4	8.5
24DCP	900	9.3	ND	ND	1.8	6.6	22	45	120	270
25DCP	892	150	3.4	9.7	24	80	370	670	1800	12,000
24D	896	<1	ND	ND	ND	ND	1.2	1.5	5.1	15
IPP	902	<1	ND	ND	ND	ND	ND	1.6	4.3	9.6
1N	891	15	ND	1.4	3.4	9.6	21	36	190	1400
2N	893	5.4	ND	1.1	2.6	7.6	14	18	32	48
4NP	886	1.2	ND	ND	ND	1.3	2.2	3.8	9.5	44
PCP	886	1.8	ND	ND	1.2	2.0	3.7	5.4	9.6	29
245TCP	847	<1	ND	ND	ND	ND	1.4	2.0	6.1	19
246TCP	867	<2	ND	ND	ND	ND	ND	3.2	15	28
TCPY	900	3.1	ND	1.3	2.2	3.5	6.3	8.3	16	34

[a] Micrograms per gram creatinine.

tions between blood and urinary toxicant concentrations with creatinine-corrected urinary concentrations than with uncorrected urinary concentrations (Cline *et al.*, 1989; Hill *et al.*, 1995b), we used uncorrected concentrations in our discussion to avoid confusion when comparing to concentrations reported in the literature that frequently reported uncorrected concentrations in micrograms per liter, milligrams per liter, ppb, or ppm.

DISCUSSION

It is striking that six of the analytes—24DCP, 25DCP, 1NAP, 2NAP, PCP, TCPY—were detected in more than 50% of the population, likely indicating frequent exposure of the general public to the parent pesticides or their residues. The most prevalent analyte was 2,5-dichlorophenol, reflecting exposure of virtually all (98%) of our sample population to DCB. DCB is readily metabolized to 25DCP (Pagnotto and Walkley, 1965). We reported the strong correlation of 25DCP in urine with DCB in blood (Hill *et al.*, 1995b). Concentrations in our population ranged as high as 8700 μg/liter, with median and 95th percentile concentrations of 30 and 790 μg/liter, respectively. Figure 2 illustrates that 25DCP concentrations were approximately 10 to 500 times greater than any of the other analytes measured in our study. We previously reported 25DCP in the urine of 96% of 197 Arkansas children and that these children had median and maximum concentrations of 9 and 1200 μg/liter, respectively (Hill *et al.*, 1989). Although the 25DCP concentrations were somewhat lower than those in our adult population, they are in general agreement with the reference range. There have been other reports of 25DCP measurements. Angerer *et al.* (1992a) reported that 88% of 258 men and women from the general population in Germany had measurable levels of 24DCP and 25DCP com-

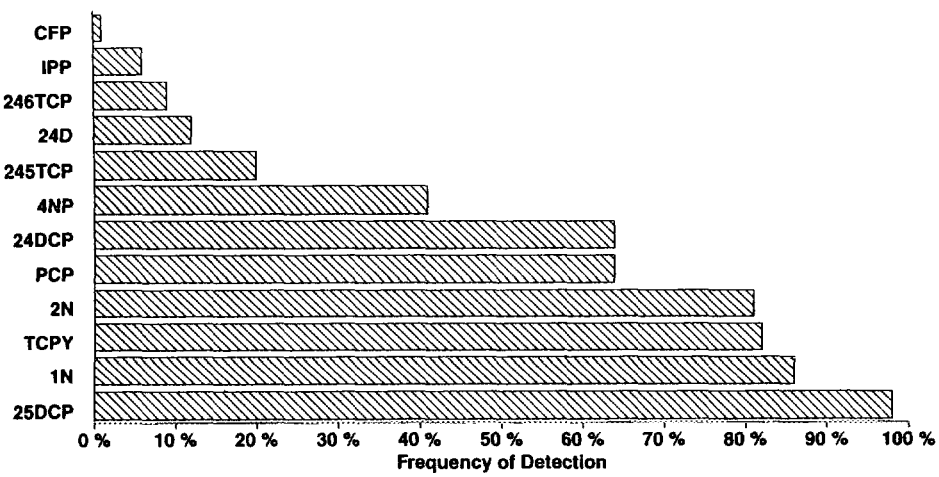

FIG. 1. Frequency of detection for all analytes: 25DCP (2,5-dichlorophenol); 24DCP (2,4-dichlorophenol); 1N (1-naphthol); 2N (2-naphthol); TCPY (3,5,6-trichloro-2-pyridinol); PCP (pentachlorophenol); 4NP (4-nitrophenol); 246TCP (2,4,6-trichlorophenol); 245TCP (2,4,5-trichlorophenol); 24D (2,4-dichlorophenoxyacetic acid); IPP (2-isopropoxyphenol); CFP (7-carbofuranphenol).

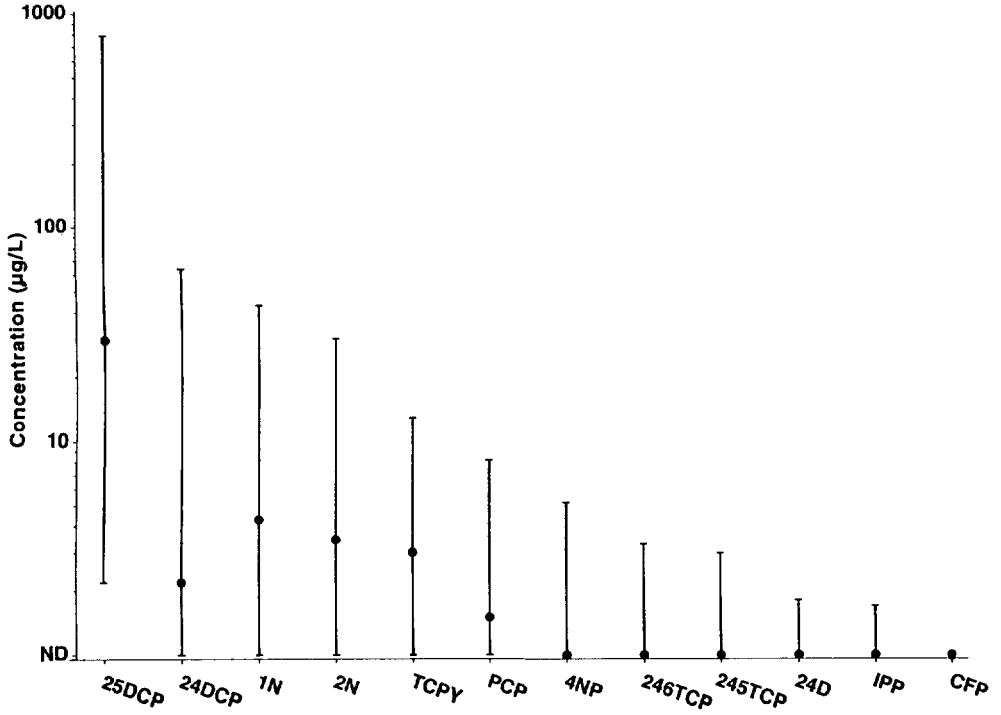

FIG. 2. Concentrations of each analyte at the 5th, 50th (median), and 95th percentiles. The top of each bar is the 95th percentile, the solid circle between these bars is the median, and the lower bar is the 5th percentile, which was not detected for any analytes except 24DCP. See Fig. 1 for identification of abbreviations.

bined and that their median and maximum urinary concentrations for 24DCP and 25DCP combined were 3.9 and 209 μg/liter, respectively. In another study, Angerer *et al.* (1992b) reported that 53 workers at a municipal waste incinerator had significantly higher urinary concentrations of dichlorophenols (median 10.5 μg/liter) than a reference population of 431 men and women (median 3.9 μg/liter). Occupational exposure to DCB resulted in urinary 25DCP concentrations of 10,000 to 233,000 μg/liter (Pagnotto and Walkley 1965); these concentrations were 13 to 300 times higher than our 95th percentile reference range concentrations.

DCB is readily found in our world, its principal uses being as a toilet deodorizer, a moth repellent, and a chemical intermediate for a polymer. It is widely available to the public in the United States in many grocery and retail stores and is frequently used in many public and private businesses, particularly in restrooms and toilets. The TEAM study reported 80% of homes tested had detectable DCB in the indoor air and hypothesized that moth crystals and room air deodorizers were important sources of DCB exposure in homes (Wallace *et al.*, 1987). Personal air concentrations taken in the TEAM study had DCB concentrations ranging from 0.02 to 2600 $\mu g/m^3$ (Pellizzari *et al.*, 1987). The prevalence of measurable 25DCP concentrations in the urine of our study population and the Arkansas children demonstrates ubiquitous exposure to DCB in the United States; these findings, when combined with observations from other countries, suggest that DCB is a worldwide contaminant (Hill *et al.*, 1989, 1995b).

We observed that 64% of our population had detectable concentrations of 24DCP. The median and 95th percentile concentrations of 24DCP were 2.2 and 64 μg/liter, respectively, and the maximum observed concentration was 450 μg/liter. We previously analyzed this analyte in urine samples from 197 children in Arkansas (Hill *et al.*, 1989) and found that 27% of the children had detectable 24DCP concentrations and that their median and 95th percentile 24DCP concentrations were <1 and 7 ppb, respectively. There are several possible sources for 24DCP (see Table 1). We observed a good correlation (Pearson correlation = 0.70, $P = 0.0001$) between 24DCP and 25DCP log creatinine-corrected concentrations, indicating that these compounds likely arise, at least in part, from a common source. Because there is a single source, DCB, for 25DCP, we suspect that a primary source of 24DCP concentrations in the general population may be from *m*-dichlorobenzene as an impurity in *p*-dichlorobenzene.

We found 1-naphthol in 86% of the samples tested

and 2-naphthol in 81%. The mean, 95th percentile, and maximum concentrations were 17, 43, and 2500 μg/liter, respectively, for 1NAP and 3.4, 30, and 88 μg/liter, respectively, for 2NAP. We found a good correlation (Pearson correlation = 0.64, $P = 0.0001$) between 1NAP and 2NAP log creatinine-corrected concentrations in these urine samples, providing support for the hypothesis that they were derived from a common source. The levels of 1NAP and 2NAP that we found most likely indicate exposure to naphthalene because naphthols are reported metabolites of naphthalene (Bieniek, 1994; Corner and Young, 1954). Naphthalene is found in petroleum products and cigarette smoke, and is used in moth balls. The approximately 25-fold difference between the maximum observed concentrations for 1NAP and 2NAP likely suggests that there was a source of 1NAP other than naphthalene. We speculate that the higher concentrations of 1NAP reflect exposure to carbaryl or occupational exposure to polycyclic aromatic hydrocarbons; however, we do not have information available to confirm this hypothesis. Although the pesticide carbaryl is readily metabolized to 1NAP (Knaak *et al.*, 1968), it seems unlikely that most of our population would be exposed to carbaryl (Whitmore *et al.*, 1994).

Keimig and Morgan (1983) found that urinary 1NAP could be used to monitor exposure to PAHs in occupationally exposed workers. However, Danish researchers found that concentrations of 1NAP did not differ between PAH-exposed workers and control subjects; of 236 male workers, 87% had detectable 1NAP concentrations and the mean 1NAP concentration of the group was 2.7 μmol/mol creatinine (3.45 μg/g creatinine) (Hansen *et al.*, 1994, 1995). Bieniek (1994) reported that workers employed in the distillation of napththalene oil had urinary 1NAP concentrations between 400 and 34,600 μg/liter; coke oven workers using two different technologies had mean urinary 1NAP concentrations of 890 and 4860 μg/liter. Shafik *et al.* (1971) reported that formulators in a carbaryl manufacturing plant had urinary 1NAP concentrations from 6200 to 78,800 μg/liter and that agricultural workers had 1NAP concentrations from 70 to 1700 μg/liter. All of these occupational exposures were approximately 2 to 2000 times higher than our 95th percentile reference range concentration for 1NAP.

We observed that 82% of our population had measurable concentrations of 3,5,6-trichloro-2-pyridinol, the metabolite of the pesticides chlorpyrifos and chlorpyrifos-methyl. This high frequency of exposure is consistent with the reported frequent use of chlorpyrifos as a common household insecticide, particularly as a replacement for the termiticide chlordane. These results differ significantly from the results of the second National Health and Nutrition Examination Survey (Kutz *et al.*, 1992) collected between 1976 and 1980. In NHANES II, 5.8% of the 6990 people studied had concentrations of TCPY greater than its detection limit of 5 μg/liter (5 ng/ml). Because the detection limit of the method we used was 1 μg/liter for TCPY, part of the increase in our frequency of detection is due to the sensitivity of the method. Nevertheless, 31% of our population and concentrations of 5 μg/liter or greater—a five-fold increase over concentrations observed in NHANES II. We believe our results are more likely due to an increase in the use of chlorpyrifos in the United States and a corresponding increase in the exposure of our population to this pesticide. Bartels and Kastl (1992) reported that TCPY was present in pooled urine of unexposed control subjects at a concentration of 5 μg/liter.

Pentachlorophenol was detected in 64% of our population, and the population's median, 95th percentile, and maximum concentrations were 1.5, 8.2, and 55 μg/liter, respectively. All 197 children in Arkansas had measurable urinary PCP concentrations, and the group had median, 95th percentile, and maximum concentrations of 14, 110, and 240 μg/liter, respectively (Hill *et al.*, 1989). Kutz *et al.* (1992) reported that 71.6% of the NHANES II population ($n = 6990$) had urinary PCP concentrations greater than 2 μg/liter (2 ng/mliter). However, our detection limit was 1 μg/liter compared with 2 μg/liter in the NHANES II; only 39% of the adults in our study had concentrations greater than 2 μg/liter. These data suggest that exposure to PCP has decreased between NHANES II and NHANES III during the late 1980s. The principal use for PCP was as a wood preservative until 1984 when EPA made PCP a restricted-use pesticide (EPA, 1984). The observed decrease in exposure likely reflects the decreased use of PCP as a wood preservative.

We previously observed that PCP was detected in all 143 people who were not overtly exposed to PCP; the median and maximum urinary PCP concentrations among these nonoccupationally exposed people were 3, and 17 μg/liter, respectively (Cline *et al.*, 1989). A study in Canada showed that 100% of 87 samples from a large pool of samples had detectable PCP urinary concentrations and that the median and maximum concentrations of the samples were 1.3 and 9.1 μg/liter, respectively (Thompson and Treble, 1994). Cline *et al.* (1989) reported that residents of homes made with PCP-preserved logs had an average urinary PCP concentration of 69 μg/liter

and that concentrations ranged up to 340 μg/liter; this average concentration is about 30 times greater than our reference range concentration. Four workers handling blocks of PCP had an average urinary PCP concentration of 10,000 μg/liter and individual concentrations that ranged from 2400 to 13,800 μg/liter; the worker with the highest PCP concentration died (Cline *et al.*, 1989). Pekari *et al.* (1991) studied sawmill workers exposed to chlorophenolate wood preservative and found that their average urinary PCP concentration was 90 μg/liter (0.34 μmol/liter).

4-Nitrophenol was detected in 41% of the urine samples from our population; the median, 95th percentile, and maximum concentrations were ≤1, 5.2, and 63 μg/liter, respectively. Results of the NHANES II study (n = 6990) shows that 2.4% of the U.S. population had detectable concentrations of NP; the limit of detection for 4NP was 10 μg/liter (Kutz *et al.*, 1992). Only 1.7% of our population had 4NP concentrations greater than or equal to 10 μg/liter. The detection of 4NP in 41% of the samples in our population is somewhat surprising, since we would not expect the general population to be frequently exposed to parathion, EPN, or nitrobenzene. We suspect that 4NP concentrations in the general population may be from acetaminophen use; this over-the-counter drug is commonly used in the United States and is manufactured from 4NP (ATSDR, 1992). Although Ascah and Hunter (1988) reported that 4NP is an expected impurity in acetaminophen preparations, we found no reported concentrations.

Occupational exposure to parathion results in high urinary 4NP concentrations. Durham *et al.* (1972) observed concentrations as high as 2400 μg/liter. Average urinary concentrations of 4NP were 40,300 μg/liter among people who died from acute parathion poisoning and 10,800 μg/liter for nonfatal cases of acute parathion poisoning (Davies *et al.*, 1966). Davies *et al.* also observed that formulators who usually handled parathion carefully had average urinary 4NP concentrations of 900 μg/liter, whereas formulators who handled it carelessly had average urinary 4NP concentrations of 4300 μg/liter, high absenteeism, and depressed cholinesterase activity. These occupational exposures were approximately 500 to 25,000 times greater than the reference range concentrations.

The herbicide 24D was observed in 12% of these samples and the 95th percentile and maximum concentrations of these samples were 1.8 and 37 μg/liter, respectively. Previously we reported that 20% of 197 Arkansas children had detectable concentrations of 24D and that these children's 95th percentile and maximum concentrations were 3 and 9 μg/liter (Hill *et al.*, 1989). A study of 6990 people from the general population living in the United States showed that 0.3% had 24D concentrations greater than or equal to 30 μg/liter (Kutz *et al.*, 1992). In our study population, we only had one sample that had 24D concentrations greater than or equal to 30 μg/liter; this is equivalent to 0.1% of our population.

There are several reports of occupational exposure to 24D. Researchers in Turkey reported pesticide formulators had urinary 24D concentrations that ranged from 60 to 9510 μg/liter, and averaged 1370 μg/liter (Vural and Burgaz, 1984). Investigators in Florida found that pesticide operators applying 24D to weeds using a handgun from an airboat had average urinary 24D concentrations ranging from 116 to 671 μg/liter (Nigg and Stamper, 1983). Forestry workers involved in 24D aerial applications had similar average urinary 24D concentrations ranging up to 837 μg/liter (Frank *et al.*, 1985). Individual urinary 24D concentrations in these forestry workers ranged as high as 2219 μg/liter. Sprayers in Germany using 24D had urinary concentrations after exposure ranging from 5.8 to 2392 μg/liter; 24D was not detected in their urine before exposure (Knopp and Glass, 1991). Rivers *et al.* (1970) measured urinary 24D in a person with acute poisoning and found that urine collected on the day the person was admitted to the hospital had a 24D concentration of 145,000 μg/liter; the concentration rose to a high of 1,900,000 μg/liter on the third day before falling to 250 μg/liter on Day 12.

2,4,5-Trichlorophenol and 2,4,6-trichlorophenol were found in 20 and 9.5%, respectively, of our population. The 95th percentile and maximum urinary concentrations were 3 and 25 μg/liter, respectively, for 245TCP, and 3.3 and 63 μg/liter, respectively, for 246TCP. Kutz *et al.* (1992) found that 3.4% of the NHANES II population had detectable concentrations of 245TCP; their detection limit for 245TCP was 5 μg/liter. In our study population, we found that 2.2% had 245TCP concentrations greater than or equal to 5 μg/liter. In the Arkansas study, we observed 245TCP in 54% of the children's samples, and 246TCP in 11% of the samples (Hill *et al.*, 1989), with concentrations ranging up to 32 μg/liter for 245TCP, and 41 μg/liter for 246TCP. A study of the general population composed of 258 men and women living in Germany included urinary 245TCP and 246TCP concentrations; 245TCP and 246TCP were found in 54 and 37% of this population, with maximum concentrations of 15 and 75 μg/liter, respectively (Angerer *et al.*, 1992a). The detection limits in this study were 0.8 μg/liter for 245TCP and 1.2 μg/

liter for 246TCP. The median, 95th percentile, and maximum concentrations of 245TCP were 1, 4.5 and 75 μg/liter, respectively; those concentrations for 246TCP were 2, 4.7, and 15 μg/liter, respectively (Angerer *et al.,* 1992a). The concentrations reported by these investigators were comparable with the results of our investigation.

There are several possible sources for 245TCP and 246TCP (see Table 1); however, we did observe a weak correlation of log creatinine-corrected concentrations for these two analytes (Pearson correlation = 0.42, $P = 0.0001$), perhaps indicating some common source. We did not, however, observe a correlation with pentachlorophenol. Lindane is the most likely source of 245TCP and 246TCP; these compounds are reported to be the main metabolites of lindane in man (Angerer *et al.,* 1983). Finnish investigators found average urinary 246TCP concentrations of 990 μg/liter (5.04 μmol/liter) among sawmill workers exposed to chlorophenolate used as a wood preservative (Pekari *et al.,* 1991).

Isopropoxyphenol and carbofuranphenol were observed in 6.8 and 1.5%, respectively, of our study population, and when detected, these residues were at relatively low concentrations, ranging up to a maximum of 10 and 14 μg/liter, respectively. Occupational exposure to propoxur in greenhouses resulted in excretion of IPP; however, the authors reported the IPP concentrations in micrograms excreted per 24 hours and not in micrograms per liter or micrograms per gram creatinine (Brouwer *et al.,* 1993). By assuming that approximately 1.2 liters of urine is excreted in 24 hr and then converting the daily excretion rate to micrograms per liter, these researchers found a mean urinary IPP concentration of about 190 μg/liter, with concentrations ranging from 12 to 1500 μg/liter.

CONCLUSIONS

Reference range concentrations for environmental contaminants serve an important role in environmental health investigations and studies. They provide information about the prevalence and magnitude of exposure, which can be used as a basis for comparing concentrations in subjects who have suspected or known exposure to a point source. To illustrate this, let us suppose that a community is located near a hazardous waste site or a plant where pesticides are made or adjacent to a place that uses pesticides. By comparing the urinary concentrations of the residues in the people in the community with the reference range concentrations, we can ascertain if that community has had elevated or unusual exposure. For example, in one study we measured these pesticide residues in a group of 31 people and found their urinary concentrations of 25DCP averaged 18 μg/liter and ranged from 1 to 140 μg/liter. Everyone in this group had detectable concentrations of 25DCP. Did this group of people have abnormal exposure to DCB? Comparison with the reference range concentrations in Table 2 shows that the observed concentrations of 25DCP among people in this group were all less than the concentration of the 95th percentile of our reference range population (i.e., ≤790 μg/liter) and agreed well with the expected frequency of detection. Thus, although this study group was exposed to DCB, the group's 25DCP level was not elevated or beyond the limits expected on the basis of the results from our reference range population. Without the reference range concentrations, the results would have been difficult to interpret.

The ability to obtain evidence of widespread pesticide exposure is of keen concern in the public health community (Morgan, 1992; British Medical Association, 1992; National Research Council, 1993). The National Research Council, in its report on pesticides in the diet of children, recognized the need to improve methods of estimating exposure to pesticides (National Research Council, 1993). A working group on exposure to pesticides recently recommended that "a concerted effort should be directed towards development of biologic methods to estimate internal dose in humans" (Nigg *et al.,* 1990). Biological monitoring is a very valuable tool in exposure assessment and environmental health investigations, and reference range concentrations for biomarkers provide a basis for evaluating exposure. The pesticide residues that we measured in this study are from only a selected few out of more than 200 frequently used pesticides. We need to develop additional biological monitoring methods for pesticides and to determine reference range concentrations for other pesticide residues. The findings of such research will assist in investigations of suspected pesticide exposures and provide a better picture of actual exposure in the United States. Knowledge of the extent and magnitude of exposure to pesticides can in turn help researchers identify areas of increased risks where pesticide usage should be reduced.

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