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Exposure to Organophosphate Pesticides in a General Population Living in a Rice Growing Area: An Exploratory Study

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A number of studies have focused on characterising pesticide exposure in occupational groups, mostly agricultural workers and pesticide applicators (van Hemmen 1994). By contrast, studies about the features of pesticide exposure in the general population are scarce. Yet, the general population is exposed to pesticides through several routes. Residues of different pesticides have been found in foods and water, usually at low levels (MacIntosh et al. 1996). Domestic use of many of these chemicals may be an important route of exposure as well (Whitmore et al. 1994). Pesticides applied on the crops can diffuse to the air and be detected in large areas, although pesticide air levels are also usually low (Seiber et al. 1989; Beard et al. 1995).

This work was carried out as an exploratory study to assess environmental exposure to organophosphate pesticides in a general population living in a rice growing area near the city of Valencia, Spain, mainly in relation to aerial spraying of the rice crops with pyridaphenthion [*O*-(1,6-dihydro-6-oxo-1-phenyl-3-pyridazinyl) *O*,*O*-diehtyl phosphorothioate], an ethyl organophosphorus insecticide applied from light aircrafts. In the treated area there are several small towns. In order to characterise pesticide exposure of residents living in this area, three approaches were developed: i) measurement of air levels of pyridaphenthion in three selected towns during the treatments, ii) a self-answered questionnaire collecting information about general health status and selected acute symptoms in a sample of volunteer women living in these towns, and iii) analysis of dialkyl phosphate residues in urine of a subsample of these women.

MATERIALS AND METHODS

Between June and August 1998 there were two aerial treatments with pyridaphenthion in the study area, a rice growing area near the city of Valencia, at the East of Spain (first treatment: 19th-24th of June; second treatment: 28th July-3rd August). Three residential areas, wholly surrounded by the sprayed rice crops, were selected for the study. These towns,

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Sollana, El Palmar and El Perelló, have around 4.500, 1.000 and 1.500 inhabitants, respectively.

The equipment for ambient air monitoring was installed inside the urban area of each selected town during and around the days of aerial treatments. Each air monitoring site was equipped with one air-sampler pump (sampling flow 1 liter/sec) with solid phase extraction (SPE) cartridges of activated carbon. Air samples were collected for a limited number of days coincidental with aerial spraying periods with pyridaphenthion. Cartridges were renewed each 24 hours during sampling days. Pyridaphenthion levels were analysed in air samples by gas chromatography with nitrogen phosphorus detector (NPD).

It was decided to carry out the study on women, as it was more likely that women spent most of the day inside the selected towns than men (many of the men in the study area worked outside their living towns). A sample of volunteer women living and working in the selected towns was reached through local pharmacies and housewives associations during three periods: first aerial treatment, second aerial treatment and a control period (February 1999) when no agricultural treatments with pesticides were carried out in the study area. The women were informed about the main objective of the study. They answered a questionnaire gathering information about personal characteristics (age, time period living in the selected town, occupational situation, present occupation, engagement on agricultural activities during the previous week and agricultural activities of other people living in the same house), health indicators (subjective general health status - very bad / bad / regular / good / very good -, diseases, respiratory, digestive or neurologic acute symptoms and other symptoms - yes / no -) and open air activities carried out the day before the questionnaire was filled.

During the second treatment and the control periods, participating women were asked to give a sample of urine and a voluntary subgroup acceded to. The urine (first urine in the morning) was analysed for dialkyl phosphate metabolites: O,O-dimethyl phosphate (DMP), O,O-dimethyl phosphorothionate (DMTP), O,O-dimethyl phosphorodithioate (DMDTP), O,O-diethyl phosphorothionate (DETP) and O,O-diethyl phosphorodithioate (DEDTP). Urine residues of these five metabolites were analysed by gas chrormatography with flame photometric detector (FPD) as previously described (Reid and Watts 1981). Recoveries of all five metabolites ranged between 78% and 95%. Limit of detection was 0.01 µg/mL.

Analysed dialkyl phosphates are used as highly specific biological markers for organophosphate exposure, although these metabolites do not allow to evaluate the exposure to specific chemicals (He 1993). Dimethyl organophosphates give dimethyl phosphate metabolites (DMP, DMTP and

DMDTP, among others), while diethyl organophosphates are metabolised to diethyl phosphate metabolites (DETP and DEDTP, among others). As pyridaphenthion is a diethyl phosphate, exposure to this compound is expected to be related to diethyl phosphate metabolites in urine.

RESULTS AND DISCUSSION

Results from the analysis of air samples are presented in Table 1. Mean levels of pyridaphenthion measured in all the samples during both treatments (excluding samples with no detectable levels) were 1.11 ng/m^3 (SD = 1.71 ng/m^3). Data in Table 1 show that aerial levels of pyridaphenthion were not always detectable in the air of the selected towns during spraying days. Pyridaphenthion air levels during the second treatment increased and decreased across the sampling days in the three air monitoring sites.

A total of 88 questionnaires (44, 37 and 7 questionnaires, respectively during first and second spraying period and control period) and 34 urine samples (28 and 6 urine samples, respectively during second spraying period and control period) were collected. Mean age of the women completing the questionnaire during the first treatment was 44.1 years (standard deviation, SD=16.7 years), ranging between 18 and 80 years. During the second treatment, mean age was 44.3 years (SD=16.8 years, range 15-75 years). Finally, during the control period mean age was 49.0 years (SD=17.8 years, range 21-63 years). Mean time residing in the selected towns for these women was over 35 years, with a range of 15 to 80 years. Mean time spent by the women in the open air (the day before the questionnaire was filled and the urine sample collected) was around 2.5 hours during treatment and control periods, without major differences among them.

Regarding general health status, assessed through a five-steps categorical item in the questionnaire, 73%, 76% and 71% of the women considered that their health was "very good" or "good", respectively during the first and second spraying periods and during the control period. Table 2 presents the main results on acute symptoms. In general, variations among the three periods were small and data do not seem to suggest a higher frequency of these symptoms during periods of aerial treatment with pyridaphenthion. In fact, the mean number of symptoms per woman was very similar during the first and second spraying periods (respectively, mean $\pm$ SD: 1.9 $\pm$ 1.4, 1.8 $\pm$1.7) and it was higher during the control period (2.6 $\pm$1.6). However, because of the low number of observations during the control period, this difference was not statistically significative ($p > 0.2$)

Results of the analysis of dialkyl phosphate residues in urine samples are presented in Table 3. During both the treatment and the control periods, dialkyl phosphate residues were detectable in the urine of some women. In general, neither the frequency of positive samples nor the levels of dialkyl phosphate metabolites show large differences among treatment and control periods. This observation is particularly relevant regarding measured diethyl phosphate levels (DETP, DEDTP), as these metabolites are directly related to pyridaphenthion exposure. In fact, for diethyl phosphate metabolites, the frequency of women with detectable levels was higher during the control period (respectively, 17% and 50% of positive urine samples), although the mean levels were slightly higher during the spraying period (respectively, 0.09 and 0.11 μg/mL).

The relationship between acute symptoms and detectable levels of dialkyl phosphate residues and diethyl phosphate residues in urine was analysed as well. Mean number of total symptoms per woman were not significantly different for women with and without urine detectable levels of all dialkyl phosphates or diethyl phosphates at the 0.05 level (respectively, p= 0.76 and p= 0.82).

Results from this work show that during aerial spraying of rice crops in the study area there were detectable levels of pyridaphenthion in nearby residential areas, and therefore it was possible for the population living there to have aerial exposure to this chemical around and during treatment periods. However, in the urine of participating women there were detectable levels of dialkyl phosphates (diethyl and dimethyl phosphates) during spraying and non spraying periods, and this finding suggests that women in the study could have other sources of exposure to organophosphate pesticides. In fact, aerial spraying with pyridaphenthion in the rice crop area of the study do not increase noticeably the level of exposure to organophosphates in the study sample of women living nearby treated areas. Perhaps aerial levels of pyridaphenthion in the selected towns for this study are not high enough to significantly modify pesticides baseline exposure levels of organophosphates in the population. On the other hand, selected acute symptoms are not more frequent during treatment periods, neither among women with detectable levels of all dialkyl phosphate or diethyl phosphates metabolites in urine.

To our knowledge, there are very few studies measuring dialkyl phosphates residues in the urine of general population not occupationally exposed to pesticides. Aprea et al. (1996) analysed urine samples from 124 residents in an agricultural region of Italy and found an association between the proportion of samples with detectable levels of dialkyl phosphate metabolites and the consumption of foodstuffs not produced in their own crops. These authors did not find a relationship between the presence of the metabolites and the place of residence of their study subjects. On the other hand, data from the Second National Health and Nutrition Examination Survey (NHANES II) on 21,000 persons examined between 1976 and 1980 enabled the analysis of dialkyl phosphate levels in urine and other pesticide metabolites (Murphy et al. 1983).

In Table 4 data from the studies by Aprea et al. and Murphy et al. regarding detectable levels of dialkyl phosphate metabolites in urine are presented together with data from this study. As it can be seen, the frequency of urine samples with detectable levels of the metabolites is higher in our study. Our research was performed in a highly agricultural area. Murphy et al. studied a representative sample of the U.S. general population, while the Italian study included people living in coastal and mountainous zones of Toscana, excluding persons with potential occupational exposure to organophosphates. In our case, the number of women working in agriculture was very low (n=5, 7%), although the number of women having performed an agricultural activity during the week prior to completing the questionnaire (agricultural activities did not involve direct handling of pesticides in any case) was slightly higher (n=10, 14%). Besides, the number of women living with someone working in agriculture (n=19, 26%) or the number living with someone involved in agricultural activities during the previous week (n=40, 55%) were still higher. It is possible for these women to have pesticide exposure when they are working on the crops, even if they do not directly manipulate the chemicals (Popendorf 1992), or through "home contamination" related to other exposed people living in the same house (McDiarmid and Weaver 1993).

The present work has some limitations. In particular, the number of women included during the period without pesticides treatments is small and, hence, the corresponding results have low precision. Besides, self-reporting of acute symptoms could be influenced by the year season (summer during the treatment periods and winter during the control period). Participation was attained on a fully voluntary basis. Thus, generalization to the whole population living in the study area was limited.

Nevertheless, our results suggest that aerial spraying with pyridaphenthion on rice crops in the study area did not affect noticeably levels of dialkyl phosphate residues in urine of the residents. Therefore, this population is likely exposed to other sources of pesticides, such as consumed foodstuffs and domestic pesticides (MacIntosh et al. 1996; Whitmore et al. 1994; Aprea et al. 1996). These alternative sources of exposure deserve a full scale field investigation.

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Table 1. Pyridaphenthion levels (ng/m^3) in the air samples collected during periods of aerial spraying in three towns surrounded by a rice growing area

	Monitoring dates (first aerial spray period: 19th - 24th June, 1998)					
	19/06/98	*20/06/98*	*23/06/98*	*24/06/98*	*25/06/98*	*26/06/98*
Sollana	n.d.	n.d.	0.16	n.d.	0.59	n.d.
El Perelló	n.d.	n.d.	n.d.	0.48	n.d.	n.d.
El Palmar	n.d.	n.d.	n.d.	n.d.	0.17	n.d.

	Monitoring dates (second aerial spray period: 28th July - 3rd August, 1998)			
	28/07/98	*29/07/98*	*30/07/98*	*31/07/98*
Sollana	n.d.	1.23	5.97	0.08
El Perelló	n.d.	0.05	0.82	0.23
El Palmar	n.d.	n.d.	2.78	0.81

n.d.: Non-detectable levels

Table 2. Frequency of women with selected acute symptoms during spraying and control periods

	First spray period		Second spray period		Control period	
Symptom	**n**	**%**	**n**	**%**	**n**	**%**
Cough	4	9.1	3	8.1	2	28.6
Eyes irritation	10	22.7	9	24.3	1	14.3
Throat irritation	3	6.8	7	18.9	2	28.6
Nose irritation	7	15.9	3	8.11	1	14.3
Skin irritation, itching	8	18.2	4	10.8	1	14.3
Wheezing	3	6.8	4	10.8	2	28.6
Shortness of breath	4	9.1	2	5.4	1	14.3
Headache	14	31.8	13	35.1	4	57.1
Stomach ache, diarrhoea	4	9.1	4	10.8	1	14.3
Nausea	2	4.6	1	2.7	0	---
Dizziness	6	13.6	4	10.8	1	14.3
Hand/head trembling	5	11.4	0	---	0	---
Fatigue / general discomfort	13	29.6	12	32.4	2	28.6
Total of symptoms per woman						
Mean ± SD	1.9 ± 1.4		1.8 ± 1.7		2.6 ± 1.6	
Range	0-6		0-6		1-6	

SD: Standard deviation

Table 3. Analysis of dialkyl phosphate residues in urine samples during second treatment and control periods

	Second spray period (number of samples=28)	Control period (number of samples=6)
Dimethyl phosphate (DMP)		
Detectable level: n (%)	9 (32%)	2 (33%)
Mean ± SD (μg/mL)	0.25 ± 0.17	0.25 ± 0.07
Range (μg/mL)	0.07-0.60	0.2-0.3
Dimethyl phosphorothionathe (DMTP)		
Detectable levels: n, %	11 (39%)	1 (17%)
Mean ± SD (μg/mL)	0.43 ± 0.39	0.5
Range (μg/mL)	0.15-1.49	0.5
Dimethyl phosphorodithioathe (DMDTP)		
Detectable levels: n (%)	5 (18%)	0
Mean ± SD (μg/mL)	0.06 ± 0.02	---
Range (μg/mL)	0.04-0.08	---
Diethyl phosphorothionathe (DETP)		
Detectable levels: n, %	2 (7%)	1 (17%)
Mean ± SD (μg/mL)	0.09 ± 0.04	0.03
Range (μg/mL)	0.06-0.12	0.03
Diethyl phosphorodithioathe (DEDTP)		
Detectable levels: n, %	5 (18%)	3 (50%)
Mean ± SD (μg/mL)	0.11 ± 0.09	0.05 ± 0.02
Range (μg/mL)	0.04-0.26	0.03-0.07

SD: Standard deviation

Table 4. Frequency of detectable levels of dialkyl phosphate metabolites in urine on the general population. Results from studies carried out in the United States, Italy and Spain.

	Frequency of detectable levels[a]		
	Murphy et al. United States	*Aprea et al. Toscana, Italy*	*García et al. Valencia, Spain*[b]
DMP	12%	15%	32%
DMTP	6%	32%	35%
DMDTP	< 1%	3%	15%
DEP	7%	6%	--
DETP	6%	4%	9%
DEDTP	< 1%	0%	24%

[a] These data have been calculated for a common limit of detection for the three studies equal to 0.02 μg/mL.

[b] Present study. Data are referred to the total of urine samples analysed during spraying and control periods.

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