



## Organophosphate pesticides exposure bring out neurological symptoms in the children of agriculture workers in rural India: A community based cross-sectional study from Maharashtra.

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### Abstract

**Background:** In rural areas of developing countries, children of agricultural workers have a high potential for exposure to organophosphorus (OP) pesticides. This puts them at an increased risk of damage to neurobehavioural performance, cognitive and psychomotor dysfunction. The main objective of this study was to assess the neurological risk of exposure to organophosphate pesticides in the children of agricultural workers in rural India.

**Methods:** A community based cross-sectional study was carried out in 200 children aged 8-15 years in adjoining villages of Taluka Chalisgaon, district Jalgaon, Maharashtra. Various neurological symptoms like muscarinic (diarrhea, urinary incontinence, lacrimation, excessive salivation), nicotinic (tremors, muscle weakness, tachycardia) and general symptoms (headache, insomnia, numbness in legs, fatigue, anorexia, nausea, vomiting, dizziness, lethargy) were assessed by a specially designed Q16 questionnaire. Exposure index (EI) was calculated by multiplying the number of hours exposed to OP pesticides and the number of years of exposure.

**Results:** The predominant clinical symptoms found in children in study group were muscarinic [diarrhea (12%), lacrimation (26%) and urinary incontinence (12%)]; nicotinic [muscle weakness (42%)] and general symptoms [pallor (58%), fatigue (34%), headache (30%), numbness in legs (24%), lethargy (20%)]. Also, the cognition and psychological function was more impaired in children exposed to OP pesticides than the non-exposed children.

**Conclusion:** Our study findings suggest that neurologic symptoms involved both central and peripheral nervous systems, resulting from occupational and environmental exposure to OP pesticides. The high prevalence of neurologic symptoms could be attributed to chronic effects of OP pesticides on the central nervous system.

**Keywords:** Children of agriculture workers, Neurological symptoms, Organophosphate (OP) pesticides, Rural

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### Background

Organophosphate (OP) compounds although being primarily neurotoxic are used as pesticides and industrial chemicals on a large commercial basis. They may involve both central and peripheral nervous systems causing well-defined muscarinic, nicotinic and cholinergic neurosymptoms [1]. OP pesticide self poisoning has also been found to be an important clinical as well as public health problem particularly in rural areas of the developing countries. Surprisingly, it kills an estimated 200,000 people worldwide. One of the best documented health effects of increased exposure to pesticide is Neurologic dysfunction. The early evidence of neurologic dysfunctions would be increased prevalence of neurologic symptom even before the appearance of clinical signs which are measurable [2, 3]. It has

also been observed that exposure to OP pesticides also constitutes occupational hazard for people working in farm and it may affect their children through the take-home pathway. If the pesticides are sprayed during the dry season, it may affect adversely the children of the farmers due to increase tendency of organophosphate exposures [4].

In rural India, it has been evident that children of agricultural families have a great possibility of OP pesticide exposure. This exposure tend to increase during the activities of pesticide mixing, application, and intensive hand labor performed by the parents in treated fields. Chemicals may be brought into the home on work boots, tools, work clothing, or on the skin by the parents of children themselves or other family members while spraying pesticides. Moreover children's exposure to pesticides may be greatly increased by household proximity to farms treated with pesticides which may enter into houses or other neighborhood playing areas of children. In other cases children may use fields as play areas, with or without the knowledge of their parents [5]. Prolonged exposure to pesticides being toxic to the brain may involve both central and peripheral nervous systems causing well-defined muscarinic, nicotinic and cholinergic neuro symptoms [6].

There have been only few scientific studies which have estimated the extent of children's exposure to OP pesticide [7]. Since pesticide exposure in children is very rampant in rural areas of developing countries, the present study was carried out to assess the neurological risk of exposure to OP pesticides in children of agricultural workers in rural India.

## **Material and methods**

**Study period** - The present study was conducted in the year 2012.

### **Study design and the participants**

A community based cross-sectional study was carried out in 200 children in the adjoining villages (Khedgaon, Mehunbare, Khedikhurd) of Taluka Chalisgaon, district Jalgaon, Maharashtra. Study (OP exposed) and control group both comprised of 100 rural children in each group, age 8-15 years belonging to similar socio-economic strata.

### **Data collection**

Investigators of this study (Sukshohale ND, Sawant PR, Thakre SS, Khamgaonkar MB, Agrawal SB) collected the data by interviews with the villagers. They used extensive probing and questions (Q16) to obtain data about the OP exposure and clinical manifestations. Investigators allowed respondents to

express their understanding in their own terms. During interviewing we also instructed to the subjects "Feel free to take a couple minutes to think about it".

### **Inclusion criteria**

Children exposed to OP (study group) directly or indirectly were included. Children never involved in pesticide handling (neither outdoor nor indoor) and having no family history of pesticide exposure were included in the control group.

### **Exclusion criteria**

Parents who were not willing to allow their children to participate in this study, children who are suffering from any disease which mimics the affects of OP poisoning and those who are under medication were excluded because of negative influence on this study.

### **Ethical committee approval**

The study was approved by Institutional Ethics Committee. After obtaining written informed consent from the parents of children, various neurological symptoms like muscarinic (diarrhea, urinary incontinence, lacrimation, excessive salivation), nicotinic (tremors, muscle weakness, tachycardia) and general symptoms like headache, insomnia, numbness in legs, fatigue, anorexia, nausea, vomiting, dizziness and lethargy were assessed.

### **Questionnaire and its validity**

The questions on the symptoms were based on an established specially designed questionnaire Q16 that was used to evaluate the effects of occupational exposure to neurotoxicants by Lundberg et al [8].

Many Questionnaires have been developed to monitor early effects of neurotoxic exposures in occupational workers. However, a validation procedure has been described only for the questionnaire 16 (Q16). The questionnaire 16 (Q16) has been used most commonly to study the prevalence of neurotoxic symptoms among workers exposed to organic solvents. It contains 16 short questions with yes or no response alternatives on symptoms commonly described by workers exposed to solvents. The understanding of the questions was investigated by physicians, psychologists, and workers. The reliability was studied by test-retest procedures. The validity was evaluated by investigating the power of the questions to discriminate between exposed and non-exposed groups and by comparisons of groups with and without a "psycho-organic syndrome" [9]. The current recommendation is that exposed

workers reporting more than six symptoms should be referred for further examination of possible chronic toxic encephalopathy.

Exposure index (EI) was calculated by multiplying the number of hours exposed to organophosphate (OP) pesticides and the number of years of exposure. Children were categorized according to exposure index as < 1, 1- 4 and >4 hours years of exposure.

Also, detailed history regarding demographic variables, socioeconomic status, personal history and presenting complaints if any were inquired. In addition complete clinical examination including general and systemic examination was done.

**Data management and statistical analysis**

Data analyses (differences between categorical variables tested with chi-square test) were conducted by means of statistical software Open Epi Info version 2.3 year 2009. Statistical significance was assessed at a type I error rate of 0.05.

**Results**

**Table 1** shows the demographic and other characteristics of study subjects. It was observed that majority of the children in study group were in the age group of 12 – 13 years and 8 – 11 years in the control group. The predominant children were males 52 (52%) in study group and females 54 (54%) in control group. In both the groups’ majority of children were Hindu by religion and educated up to secondary school. Considering the physical activity, it was seen that maximum children i.e. 25 (50%) from study group were involved in heavy physical activity and 98 (98%) children in control group were involved in moderate physical activity. Dietary habits of children showed that subjects in both groups were preferably subsisting on mixed diet. Majority of children (56%) were found to be exposed to OP pesticides daily for less than 1 hour. Least number of children 11 (22%) were exposed for 1 – 4 hours and 13 (26%) were exposed for more than 4 hours. Exposure index >4 was found in maximum number of children i.e. 27 (54%). Moreover, hand to mouth activity was found in 54 (54%) of children in study group.

The prevalence of pesticide-related neurologic symptoms observed in exposed and non-exposed rural children are illustrated in **Table 2**. The predominant symptoms found in children in study group were muscarinic [diarrhea (12%), lacrimation (26%), salivation (4%) and urinary incontinence (12%)] and nicotinic [muscle weakness (42%) and tremors (8%)].

**Table 1: Sociodemographic characteristics of subjects**

Characteristics	Study group (N = 100)	Control group (N = 100)
<b>Age group (years)</b>		
8 – 9	26 (26)	30 (30)
10 – 11	24 (24)	30 (30)
12 – 13	36 (36)	20 (20)
14 – 15	14 (14)	20 (20)
<b>Gender</b>		
Male	52 (52)	46 (46)
Female	48 (48)	54 (54)
<b>Religion</b>		
Hindu	94 (94)	76 (76)
Muslim	6 (6)	24 (24)
<b>Education</b>		
Primary (up to 4th standard)	28 (28)	38 (38)
Secondary (5th to 10th standard)	72 (72)	62 (62)
<b>Type of family</b>		
Nuclear	32 (32)	90 (90)
Joint	14 (14)	0
Three Generation	54 (54)	10 (10)
<b>Physical activity</b>		
Heavy	50 (50)	2 (2)
Moderate	44 (44)	98 (98)
Sedentary	6 (6)	0
<b>Type of diet</b>		
Vegetarian	22 (22)	20 (20)
Mixed	78 (78)	80 (80)
<b>Range of exposure</b>		NA
<1 hr	52 (52)	-
1 to 4 hrs	22 (22)	-
>4 hrs	26 (26)	-
<b>Exposure index</b>		NA
<1	2 (2)	
1 to 4	44 (44)	
>4	54 (54)	
<b>Hand to mouth activity</b>		NA
Present	54 (54)	
Absent	56 (56)	

Figures in parentheses indicate percentage.

Moreover, the differences in muscarinic symptoms like diarrhea and urinary continence were found to be statistically significant between study and control group (P<0.05). Whereas no statistically significant difference was found for nicotinic symptoms between the two groups (P>0.05).

As far as general symptoms are concerned, majority of children in study group had pallor (58%), fatigue (34%), headache (30%), numbness in legs (24%), insomnia (10%), lethargy (20%), nausea and vomiting (12%). On applying chi square test, the difference in general symptoms between study and control group were

found to be statistically significant for pallor, fatigue, headache, numbness in legs, insomnia, nausea and vomiting (P<0.05).

**Table 2: Prevalence of pesticide-related neurologic symptoms**

Neurologic Symptoms	Study group (N= 100)	Control group (N = 100)	χ <sup>2</sup>	df	1 P value
<b>Muscarinic</b>					
Diarrhoea	12 (12)	4 (4)	4.34	0.03 <sup>†</sup>	
Lacrimation	26 (26)	16 (16)	3.01	0.08 <sup>x</sup>	
Salivation	4 (4)	2 (2)	0.68	0.40 <sup>x</sup>	
Urinary incontinence	12 (12)	2 (2)	7.68	0.005 <sup>†</sup>	
<b>Nicotinic</b>					
Muscle weakness	42 (42)	30 (30)	3.12	0.07 <sup>x</sup>	
Tremors	8 (8)	4 (4)	1.41	0.23 <sup>x</sup>	
<b>General</b>					
Pallor	58 (58)	38 (38)	8.01	0.004 <sup>†</sup>	
Anorexia	18 (18)	26 (26)	1.86	0.17 <sup>x</sup>	
Fatigue	34 (34)	18 (18)	6.65	0.009 <sup>†</sup>	
Dizziness	10 (10)	16 (16)	1.59	0.20 <sup>x</sup>	
Headache	30 (30)	10 (10)	12.5	0.00 <sup>†</sup>	
Numbness in legs	24 (24)	12 (12)	4.87	0.02 <sup>†</sup>	
Insomnia	10 (10)	2 (2)	5.67	0.01 <sup>†</sup>	
Lethargy	20 (20)	12 (12)	2.38	0.12 <sup>x</sup>	
Nausea and vomiting	12 (12)	2 (2)	7.68	0.005 <sup>†</sup>	

Figures in parentheses indicate percentage.

<sup>†</sup>P<0.01, statistically significant

<sup>x</sup>P<0.05, statistically significant

<sup>\*</sup>P>0.05, statistically not significant

**Table 3** shows the Neurological assessment (Questionnaire -16) of study subjects. When children were administered questionnaire, majority of children from study group responded YES for question no. 1, 3, 4, 5, 7, 8, 10, 12 and 15. Whereas maximum number of children from control group responded NO for the above questions. The responses were same for question no. 6 in both the groups. On applying chi square test, the difference between study and control groups was found to be statistically significant for question no. 4, 5, 7, 8, 10 and question no. 14 (P<0.05).

**Discussion**

The present report evaluated the role of exposure to organophosphate (OP) pesticide exposure in the children aged 8 to 15 years of rural areas in India. Our findings revealed that general symptoms (Pallor, headache, anorexia, fatigue, dizziness, numbness in legs, insomnia, lethargy, nausea and vomiting) and among all the neurologic symptoms, muscarinic (diarrhea, lacrimation, salivation and urinary incontinence) and

nicotinic (tremors and muscle weakness) were the most important clinical manifestations attributed to OP pesticide exposure.

**Table 3: Neurological assessment (Questionnaire-16) of study subjects**

Questions	Study group (n= 100)		Control group (n = 100)		χ <sup>2</sup>	df	1 P value
	Yes	No	Yes	No			
1) Are you abnormally tired?	48 (48)	52 (52)	44 (44)	76 (76)	2.88	0.08 <sup>x</sup>	
2) Do you have palpitations even when you don't exert yourself?	12 (12)	88 (88)	14 (14)	86 (86)	0.17	0.67 <sup>x</sup>	
3) Do you often have a painful tingling in some part of your body?	26 (26)	74 (74)	18 (18)	82 (82)	1.86	0.17 <sup>x</sup>	
4) Do you often feel irritated without any particular reason?	48 (48)	52 (52)	32 (32)	68 (68)	5.33	0.02 <sup>†</sup>	
5) Do you often feel depressed without any particular reason?	36 (36)	64 (64)	18 (18)	82 (82)	8.21	0.000 <sup>†</sup>	
6) Do you have problems with concentrating?	68 (68)	32 (32)	68 (68)	32 (32)		NA	
7) Do you have a short memory?	78 (78)	22 (22)	64 (64)	36 (36)	4.76	0.02 <sup>†</sup>	
8) Do you perspire without any particular reason?	28 (28)	72 (72)	8 (8)	92 (92)	13.55	0.000 <sup>†</sup>	
9) Do you have any problems with buttoning and unbuttoning?	4 (4)	96 (96)	6 (6)	94 (94)	0.42	0.51 <sup>x</sup>	
10) Do you generally find it hard to get the meaning from reading books?	32 (32)	68 (68)	48 (48)	52 (52)	5.33	0.02 <sup>†</sup>	
11) Have your relatives told you that you have a short memory?	14 (14)	86 (86)	22 (22)	78 (78)	2.16	0.14 <sup>x</sup>	
12) Do you sometimes feel an oppression of your chest?	10 (10)	90 (90)	8 (8)	92 (92)	0.24	0.62 <sup>x</sup>	
13) Do you often have to make notes about what you must remember?	6 (6)	94 (94)	8 (8)	92 (92)	0.30	0.57 <sup>x</sup>	
14) Do you often have to go back and check things you have done such as turned off the stove, locked the door, etc.?	0	100 (100)	4 (4)	96 (96)	4.08	0.04 <sup>†</sup>	
15) Do you have a headache at least once a week?	36 (36)	64 (64)	34 (34)	66 (66)	0.08	0.76 <sup>x</sup>	
16) Are you less interested in sex than what you think is normal?	Not applicable						

Figures in parentheses indicate percentage.

<sup>†</sup>P<0.01, statistically significant

<sup>x</sup>P<0.05, statistically significant

<sup>\*</sup>P>0.05, statistically not significant

### Neurological symptoms with OP exposure

Similar clinical findings have been reported by other researchers. Farhat et al who studied the association between pesticide exposure and neurologic endpoints concluded that environmental and occupational exposure to OP pesticides leads to neurodegenerative functions in agricultural workers [10]. Lotti, also observed that the association between OP pesticide exposure and neurotoxic effects are well known. Moreover, the cognition and psychological function was more impaired in children exposed to OP pesticides than the non-exposed children [11].

Our study findings also conform to the findings of studies done by other investigators, which reported increase prevalence of cholinergic symptoms like insomnia, anorexia, headache and numbness in legs in both male and female children as a result of chronic exposure to organophosphate pesticides [2-3, 12-14]. Rastogi SK et al [2] reported the maximum prevalence of muscarinic symptoms like salivation (18.22%), whereas lacrimation was noted in 17.33% cases, followed by diarrhea in 9.33% cases. The nicotinic clinical manifestations of acute OP poisoning revealed excessive sweating in 13.78% cases and tremors in 9.3% cases followed by mydriasis in 8.4% exposed children. Studies on neurologic symptoms among Sri Lankan farmers showed that 24% of the neurologic symptoms resulted from occupational exposure to AChE-inhibiting OP insecticides [15].

Previously, it was believed that OP pesticides mainly influenced brain development. The mechanism was thought to be due to initial inhibition of cholinesterase followed by cholinergic hyper stimulation induced by OP pesticides. Chronic exposure to most of the OP pesticides mainly affects the peripheral and central nervous systems which might cause important clinical neurologic manifestations, such as headache, tremors, dizziness, and numbness [16].

Another important cause for pesticide-related neurologic symptoms, both muscarinic and nicotinic observed in the present study could be because of the inhibition of red blood cells, acetyl cholinesterase, as well as plasma butyryl cholinesterase recorded in the study group. Similar mechanism has been reported in earlier previous studies [17, 18]. The long term exposure of OP pesticides causing neurologic symptoms also leads to delayed toxicity since the breakdown of these chemicals in the body is slow resulting in the storage of pesticides in the body fat. Few OP pesticides such as diazinon and methyl parathion are associated with significant lipid solubility. This permits the storage of fat leading to delayed toxicity because of late release of fat. Other OP pesticides

especially dichlorvos, fenthion and demetonmethyl may also cause delayed toxicity [19].

### Limitations and future scope of the study

In the present study, the duration and dose of exposure of OP was not documented. Information about exposure and symptoms among study and control group were collected by same investigator hence some biases might have introduced in this study. Also, we could not measure the blood levels of OP due to time constraints.

In spite of availability of many strong evidences to support that use of pesticide is scientifically linked to increased prevalence of neurosymptoms and deficits in neurobehavioral performance, many lacunae in the form of differences in methodology, lack of data regarding the detail exposure estimation exist in some previous research studies as well as present study. Therefore more scientific research is required in future to resolve these unsolved issues. The existing current literature is also insufficient to elicit whether certain functional domains could be more sensitive to pesticides than others. The above mentioned limitations need to be addressed by further scientific research in the form of cross sectional studies. A prospective study can also provide exact exposure, duration of exposure and probable outcome.

The qualitative and quantitative aspects of the exposure to OP pesticides as well as the ability to assess exposure differ in various studies. This is a serious concern and should be taken into account by taking up further innovative qualitative research. Nevertheless, the positive results obtained from present study with more comprehensive exposure assessment, together with strong positive support from other studies, reinforces the hypothesis of an association between OP pesticide exposure and neurological abnormalities.

### Conclusion

Our study findings suggest that the major morbidity in the children of agricultural workers comprises of a higher prevalence of neurologic disorders. Moreover, neurologic symptoms involved both central and peripheral nervous systems, resulting from occupational and environmental exposure to OP pesticides. The high prevalence of neurologic symptoms (muscarinic, nicotinic and general) reported in exposed children could be attributed to chronic effects of OP pesticides on the central nervous system.

**Abbreviations**

Organophosphate (OP), Exposure index (EI)

**Competing interests**

We do not have any financial or other relationships which could be construed as a conflict of interest and that all sources of financial support for this study have been disclosed and are indicated in the acknowledgement.

**Authors' contribution**

Sukhsohale ND, Sawant PR designed the study concepts, performed the experiment, search literature, collected and interpreted the data, drafted the manuscript, and reviewed it. Sukhsohale ND, Sawant PR, Thakre SS, Khamgaonkar MB, Agrawal SB analysed the data, prepared, edited and revised the manuscript. Final manuscript was approved by all authors.

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