Influence of Organophosphorus Pesticides on Visuo-Motor Function

有機磷殺虫剤の視覚・平衡機能系に対する影響

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Synopsis

Environmental exposure to organophosphorus pesticide (OP) produced pupil abnormality and latent disturbance of the body balance. The impairments may be produced by inhibition of richly innervated cholinergic sites such as pupil sphincter muscle, Edinger-Westphal nucleus and the vestibular nucleus in the midbrain since reduced activity of red cell-acetylcholinesterase (true Ch-E) was found together with the residue of OP in the peripheral blood of the patients suspected the intoxication by OP. These were not seen in the controls. It was therefore concluded that the measurement of the pupil size as well as its analyses of the dynamics of pupil-light reflex and the measurement of the balance with or without the loading of the eye movement was considered to be an important methods to establish the diagnosis of OP intoxication. The measurement of pseud Ch-E had no significant value for the diagnosis.

Introduction

Organophosphorus pesticide (OP), which is a potent anticholinesterase action, has been extensively used throughout the world, and many cases of acute intoxication have been reported. The OP produces an intense cholinomimetic actions of nicotinic as well as muscarinic reaction in cholinergically innervated nervous system. Therefore, the diagnosis of acute intoxication is very easy. However, chronic contamination with OP for example environmental exposure, the establishment of diagnosis is not simple. Usually, the disease starts with signs and symptoms by flu-like symptom or neuro-myasthenia etc. The disease is recently known as “chemical supersensitivity” of the entire body. On the other hand, most patients sometimes complain of relatively severe dizziness, oscillopsia, and difficulty of standing. The dizziness gets better when the patient is following the smoothly moving target and reverse is true for the eye closure. Later on, chronic exposure occasionally produces “delayed neurotoxicity” in humans. This is summarized as “optic-autonomic neuropathy.”

In the present study, firstly, a typical patient with chronic OP intoxication will be introduced with special emphasis on his standing ability as well as the ability with the loading of the eye movement i.e. smooth pursuit eye movement. Secondly, standing ability of the subjects

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who were diagnosed as chronic OP intoxication will be shown and the results obtained from the control will be compared. Thirdly, pupil finding of 20 selected patients will be added.

Methods

We studied a total of 109 patients with chronic OP intoxication who visited our department during past 7 years. Over 90% of the patients were farmer or professional OP sprayer with the exception a few children of farmers. The mean age was 35.8 years ranging 6 years to 58 years. The diagnostic criteria was established in 1976 of chronic OP intoxication (3). They are shown in the followings:

All patients must have a prolonged or a heavy contact with OP at least over 2 years. The patients underwent 70 written questionnaire with Yes or No answers prior to the systemic examination of intoxication (3). When "Yes" was over 20 out of 70 questions, a special examination were carried.

DIAGNOSTIC CRITERIA OF CHRONIC OF INTOXICATION

(1) ocular impairments:
   1. optic neuropathy with myopia & astigmatism
   2. dark adaptation or impaired Electroretinography
   3. smooth pursuit movement
   4. pupil and accommodation
   5. visual discomfort including photophobia and difficulty of focusing either far or near

(2) peripheral neuropathy

(3) autonomic nervous system:
   1. gastro-intestinal symptom including diarrhoe and cramp,
   2. nausea and vomiting,
   3. fatigability,
   4. numbness of the leg and hand,
   5. general malaise,
   6. muscle twitch, muscle pain or burning sensation of the leg or arm,
   7. joint pain,
   8. headache
   9. dizziness,
   10. neuro-circulatory asthenia

(4) reduced erythrocyte cholinesterase activity

(5) remarkable improvement of clinical manifestations by therapy

(6) elevation of acetyl-cholinesterase after the administration of antidotes prifinium bromide, atropine methylnitrate or 2-PAM (pralidoxime methiodide). After the examination, the patients must have at least four major manifestations (10- (4) described above to establish the diagnosis.

Four major OP were used by farmers at the time of examinations during past two years; they were 1. fenitrothion (Sumithion: 0,0-dimethyl-0\(\rightarrow\)3-methyl-4 nitrophenyl/ phosphrothionate) 2,0,0 diethyl 0\(\rightarrow\)(2-isopropyl 6-methyl-4-pyrimidinyl) diazinon, 3.dipterex Trichlorfon: 0,0-dimethyl (2,2,2-trichloro-1-hydroxyethl) phosphonate and 4. 0,0 dimethyl-0-2,2-dichlorovinyl phosphate (dichlorvos). These were detected from their blood by FID gaschromatography analysis.

As for the controls, especially in the standing ability study, 1000 healthy subjects varying
the age who had not had contact with OP were selected. In more sophisticated balance examination with the loading of smooth pursuit eye movement, 32 healthy controls with the mean age of 32.0 years who had no contact with OP were randomly selected approximately matching their age from 1000 subjects. Standing ability was examined by the following procedures. The test subject stood on the plate of an electrogravitography (EEG - Anima) fixing his eyes on the projected target 1.5 meter away from him. Secondly, the subject closed the eyes. Thirdly, smooth pursuit eye movement as well as saccadic eye movement was elicited. The target was sinusoidally moved in the horizontal plane at frequencies ranging from 0.3 to 0.7 Hz and an amplitude from 0 degree (no eye movement with central fixation) to 40 degrees and the subject was asked to follow the target. The center of gravity at the sole was recorded by EEG in both right left (X) and front back (Y). Twenty seconds were used in each examination. The effect of loading on smooth pursuit tracking on EEG was also studied.

Pupil

Pupil is a sensitive indicator of OP intoxication. Pupils were examined in the dark room immediately after the dark adaptation of 15 minutes by infrared video-pupillography (Hamamatsu photonics C-2515). Twenty OP patients hospitalized for treatment were selected out of the 109 patients. The clinical manifestation of age, cholinesterase activity, OP, and pupil of the patients are shown in Table I. The pupil area in the dark was compared with those from the 100 healthy controls who were selected from our University employees. They are healthy people except a few mild degree of myopia.

<table>
<thead>
<tr>
<th>No.</th>
<th>Age</th>
<th>Cholinesterase</th>
<th>Organophosphorus Pesticides Name</th>
<th>Pupil in Darkness</th>
<th>Pupil before and after PS (μl)</th>
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<tr>
<td></td>
<td></td>
<td>true pseud</td>
<td>pesticide</td>
<td>right left</td>
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<td></td>
<td></td>
<td>umol/ul/min</td>
<td>(ppb)</td>
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<td>after PS</td>
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<tr>
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<td>46</td>
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<td>5.0</td>
<td>D</td>
<td>33</td>
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<tr>
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<td>62</td>
<td>1.3</td>
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Mean: 53.3  S.D.: 14.9

D: Diazinon, S: Sumithion, DDVP: Dichlorphos, PS: photostress
Photostress study of the pupil: The same predark adaptation of 15 minutes was given prior to the study. After the measurement of pupil, photostress with the light intensity of 10,000 cd/m² for 20 seconds was given to the fovea by indirect binocular ophthalmoscopy with 20 diopters lens focusing on the macula. Immediately after the photostress, the pupil was measured one and three minutes afterward. The pupil size was compared with above 20 healthy controls out of 32 subjects with the mean age of 35.7 years.

Laboratory examinations

Detailed descriptions have been described elsewhere. Initially, erythrocyte cholinesterase (ChE) and serum ChE were measured by Ellman method. Most of the patients revealed that the former was reduced and the latter rather elevates. This was the good contrast that both ChE activities reduced in acute intoxicated patients. Routine blood chemistry examinations of liver function such as GOT or GPT meant nothing to establish the diagnosis of chronic OP intoxication although there were plenty of publications were available describing the OP was the safe pesticide.

Studying the following items in the blood may assist in the diagnosis of chronic OP intoxication. They are: a reduction of vitamin C, folic acid, selenium, zinc, and magnesium levels. Abnormal level of glutathion peroxidase and superoxide dismutases may also help the diagnosis.

The residue of OP in the urine or in the blood, occasionally help to diagnose the patients, however, the reduction of the blood or urine level after the use of antidote is more reliable to establish the diagnosis. This is also applicable to erythrocyte ChE elevation. For brevity, the results of the blood chemistry will not be mentioned except ChE.

For example, one 48 year old male the highest loss of body balance sprayed DDVP about 11 times per month. His presumed daily intake was 2.05mg/Kg, calculated from sprayed DDVP considering the area sprayed. This is about 70 times the maximum allowable daily intake of DDVP, i.e. 0.03mg/Kg. Clinically, he had severe peripheral sensorial neuropathy at the lower legs. He had a difficulty looking at near distances for several years because of the cycloplegia of the eye. However, no hypermetropia existed. In order to settle the diagnosis, pupil and balance must be examined.

Results and discussions

Balance study with smooth pursuit loading

Typical results obtained from the patients are given in Figure 1. Inhibitory effect of smooth pursuit on EGG is shown. He had been a professional OP sprayer for past 18 years and had a severe "optic-autonomic-peripheral neuropathy". The effects of smooth pursuit stimulus at 0.5Hz and different amplitudes (0 - 40 degrees) on EGG (X & Y) and on EGG-trajectory are given. He could follow the target at 0 degree; otherwise, no eye movement was elicited. His standing ability was highly disturbed. This can be clearly seen in the EGG and also trajectory of the EGG. When the stimulus gradually increased to 40 degrees, his balance worsened. This inhibition of the body sway by loading of smooth pursuit eye movement was a unique phenomenon seen in most of the patients. The inhibition was not seen when he was asked to look at an optokinetic drum rotating at varying velocities while in his primary position. When the extraocular muscles were paralyzed by retrobulbar injection (1.5ml of 2% procain), the above inhibition was not seen. Therefore, this inhibition was partly due to an excitatory afferent effect or proprioceptive effect from the extraocular muscles to the structures affected by OP which control body balance. According to Mukuno's recent study, muscle spindle as well as palisade form at the tendinous junction is richly innervated by cholinergic nerve. This inhibition was not seen in the patients.
SMOOTH PURSUIT

Fig. 1: Smooth pursuit of the eyes: A marked improvement of standing ability with the loading of smooth horizontal eye movement. Stimulus: target movement (0.5 Hz) from subtending amplitude 0 degrees, i.e. no movement till 40 degrees amplitude. EGG: Electrogravitiography.

who had been treated by anticholinesterase drugs for at least 6 months, and it was never seen in other neurological diseases. Therefore, this was considered to be a specific phenomenon seen in the patients with chronic OP intoxication.

The above inhibition was also observed during saccadic movement in both normal and OP patients. Loading of ocular smooth pursuit movement on EGG was examined in all patients and controls. The results given in Figure 2, as an effect of loading by ocular smooth pursuit movement on EGG in both patients and normal controls. The results were: a. inhibited EGG (66 cases), b. excited (29 cases), c. combined findings (5 cases) and d. no change (8 cases) in the patients. Therefore, 61.1% of chronic OP intoxication patients could be diagnosed by this method. This inhibition definitely improved with systemic administration of antidotes such as pralidoxime methiodide or atropine methylnitrate for a prolonged period of time. Therefore, it was evident that the improvement of standing ability by loading with the smooth pursuits ocular stimulus was due to anticholinesterase action evoked by the OP mostly at the brainstem where abundant acetylcholinesterase exists.

Standing ability and age
Standing ability i.e. area of EGG cm² obtained from 20 patients with the OP and those from the control against age in years is shown in Figure 3. Closed circles with downward vertical bars denote the means and the standard deviations of the normal controls in each age group. The same is true with upward standard deviations (shade area). The EGG obtained from 20 patients are larger than that of the controls, except in three cases (No.8, 11, and 17) where true cholinesterase activities were 2.0, 2.0, and 1.8. This was slightly higher than the other patients. Increased EGG is more obvious in younger ages. This means that the standing ability of the patients is more involved and even at the age of 20, the patients' mean value was 6.68 + 1.76 cm², being larger than the mean value of a normal 70 year old (control group), namely, 5.53 + 1.98 cm². Even at a young age, the area is closer to that of a 60-70 year old indicating an involvement of vestibulocular control system in the brainstem richly innervated by cholinergic neurons. The possibility of acceleration in the aging process is suggested.
Fig. 2: Effect of loading by ocular smooth pursuit movement on EGG. Comparison between patients (white squares) and controls (solid squares). There are 4 types of responses on EGG by loading of the eye movement: inhibited, excited, combined and no change. Significant differences existed for inhibited and no change, p 0.01(∗∗) in the Figure.

Fig. 3: Area of EGG (both eyes open) against age in years in both controls (solid circles) and OP patients (solid triangles) expressed as patients p- intoxication. The means and the standard deviations of the controls are shown. The area of EGG is higher in most of hospitalized severe cases (triangles).
Pupil study

The patients with a larger area of EGG over the mean value were selected and underwent further pupil study. The pupil area in darkness showed scattered results among normal individuals at all ages (males: solid circles, females: open circles). Nevertheless, an age trend could be discerned, and it became clearer when the measurements of relatively large groups of subjects were averaged. Pupil area in darkness against age in years is plotted together with the controls and the OP patients, and the results are shown in Fig. 4. Twenty patients with OP (triangles), demonstrated smaller pupils than those of the controls in all age groups except for three cases (47, 47 and 58 years). Thus, the patients' pupil is constricted in darkness, however, constricted pupil is not a specific sign of the OP patients especially in chronic intoxication. Very often, the pupil is normal in size or rather dilated in the child (2). Therefore, the following test should be performed to establish the diagnosis.

Photostress test of the pupil

The pupil sizes elicited by intense light stimulus in the OP patients are unique. The summary of the photostress test in the OP patients and the controls are shown in Table II. All measurements were done in a dark room by infrared pupillography. In the control group, the pupil returned to regular size even 3 minutes after the photostress. When averaged, the results from the controls were: before photostress: 32.34 mm², 1 minute after photostress: 15.38 mm², and 3 minutes after the photostress, that was 30.36 mm², respectively. However, the pupils of the patients were: 17.80, 4.92 and 9.15 mm², respectively. A significant differences existed between the patients and the controls. The pupils of the patients were smaller in the darkness even before photostress but, with the variances. Pupillary constriction evoked by photostress produced an intense constriction.

Fig. 4: Pupil area in darkness in mm² against age in years of both controls (males: solid circles, females: open circles) and patients (both eyes: triangles). The pupils of the patients were generally smaller 1 S.D. than those of the controls in all age groups except in 8 cases.
Table 2. Photostress test of the patients and the controls (Mean ± S.D.)
(right eye: Pupil area in μm²)

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<tr>
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<th>Patients</th>
<th>Normal Controls</th>
<th>Difference</th>
</tr>
</thead>
<tbody>
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<td>Before photostress</td>
<td>17.80 ± 1.14</td>
<td>32.34 ± 0.13</td>
<td>p&lt;0.05</td>
</tr>
<tr>
<td>1 minute after photostress</td>
<td>4.92 ± 3.41</td>
<td>21.38 ± 0.69</td>
<td>p&lt;0.001</td>
</tr>
<tr>
<td>3 minutes after photostress</td>
<td>9.15 ± 5.53</td>
<td>30.36 ± 0.31</td>
<td>p&lt;0.01</td>
</tr>
</tbody>
</table>

of the pupil, especially in one minute after the photostress. Clinically if the patient’s pupil is still over 50% constricted even at 3 minutes after the stimulation, the OP intoxication is very possible. We routinely use this test as an aid establishing the diagnosis.

In conclusion, chronic environmental exposure to the OP may be an accelerator in aging. Since the eye is richly innervated by cholinergic system, the damage of the cholinergic nervous system can be best demonstrated by the examinations of the body balance with the loading of smooth pursuit, pupil and accommodation system. Environmental exposure to the OP is very dangerous because it produces effects especially on the eye. Detailed examinations of the sensorial systems are highly recommended.

Chemical measurement of the blood should follow.

We would like to emphasize that an early discovery and immediate treatment of the patient is absolutely necessary. If left alone, without diagnosis, some of the patients will lose their vision, and will experience severe neuropathy in their lower legs known as "delayed neurotoxicity" by OP.

References


