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The Effect of Injected Homatropine on Mortality and Signs of Organophosphate Poisoning in Lab Rats

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Organophosphate compounds are the third common cause of poisoning and the main cause of death by poisoning in iran. This study has been done to analyze the effect of Intraperitoneal injection of homatropine Eye drop on acute poisoning by organophosphate poisons. This experimental study has been done on three groups of ten rats. All the rats were injected with 25 mg/kg dichlorvos poison Subcutaneously and then they were divided into three groups by random. The first group was injected with normal saline, the second group with homatropine eye drop and the third group with Atropine. All the rats were observed for 120 minutes and their Cardiovascularandrespiratoryvital signs were measured with precision. The data were analyzed using Chi-squaretest and ANOVA. The significance level was considered to be 0.05. All the rats of control group were deceased. And all the rats who received homatropine and atropine survived. There was no significant difference regarding the time period to effect and effectiveness of atropine compared to homatropine for Respiratory rate (p=0.412) and heart rate (0.635). Using homatropine eye drop Intraperitonealy can prevent the mortality of lab rats the way atropine does in case of acute poisoning with organophosphate poisons.

Key words: Organophosphate poisons, homatropine , mortality.

Organophosphates are used from long time ego as pesticide in agriculture and also in chemical weapons. Exposure to organophosphates is one of the most common reason of being hospitalized for pesticide poisoning. about 3 million people in the world are poisoned with these poisons yearly and 250 thousand of them die[1].

There are more than 100 kinds of compounds including organophosphorus factors in large-scale trade in the world which are utilized as pesticide in stockbreeding, agriculture, and household consumptions (as raticide)[2, 3]. These compounds mostly consist of phosphate esters,thatthe atom of phosphate is in a center including 3 lateral chains[4]. Mortality resulted from organophosphorus poisoning is multifactorial but the most common cause is pulmonary complications[1] which are defined as syndrome of cholinergic acute respiratory insufficiency or intermediate syndrome(1); the respiratory insufficiency in first 24 hours is named paralysis type I and after 24 hours it is named paralysis type II[5]. In organophosphorus poisoning most of the time cardiac complications occur too which may be serious and often pernicious[6]. Organophosphates influence electrical function and conduction system of heart. The cardiac manifestations of organophosphorus poisoning

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are sinus bradycardia, sinus tachycardia, polymorphic ventricular tachycardia from the kind of Torsade de pointe, hypotension or hypertension, and pulmonary edema with cardiac origin[7]. Organophosphates cause Cholinesterase enzyme to be controlled and therefore Acetylcholine to accumulate in the surface of myoneural receptors, and their various effects appear in this way[8]. In organophosphorus poisoning 4 clinical phases are seen that 3 of them are acute and one is chronic as follows: 1. Acute cholinergic crisis 2.Intermediate syndrome 3. Organophosphate- induced delayed neuropathy 4. Organophosphate- induced neuropsychiatric chronic disorder[9]. The most common manifestations of organophosphorus poisoning are as follow: excessive salivation , Increase oflung secretions , Runny nose, Bronchorrhea, Rhinorrhea, Diarrhea, urinary incontinence, Bronchospasm, Nausea andmyosis , Excessive accumulation of secretionsin airways and pulmonary alveoli are the main reasons of death of these patients[1, 2]. In such cases in which subacute and chronic conditions are prominent and the rate of controlling Acetylcholine increases, induced oxidative stress is cited as the main cause of poisoning[4]. Organophosphorus poisoning treatment mainly includes Atropine, Oximes, Benzodiazepines, and respiratory protections[3]. Atropine has been the base of organophosphorus poisoning treatment for its high stability against temperature, speed of effectiveness, and reduction of central nervous systemdisorders[5, 6]. It takes a competitive antagonism effect for peripheral and central status of Acetylcholine[10]. Sometimes due to the need to high doses of anticholinergic drugs in organophosphorus poisoning the supply of Atropine in the hospital has finished[4, 5]. This lack of this drug is more obvious in cases of group poisoning like what happens while using chemical weapons[7]. Hence it is always necessary to have regard for providing antidotes alternative to Atropine in emergency conditions. Recently medication of ophthalmic antimuscarinic drugs such as Homatropine in parenteral way has been considered in treating organophosphorus poisoning. In 2006 Bryant and colleagues have represented the effect of medicating ophthalmic Homatropine in intramuscular way on preventing mortality as the result of poisoning with Dichlrovos poison in rats[8]. Since in clinical setting antidote is always medicated after exposing to poison, we decided to investigate the effect of medicating intraperitoneal ophthalmic Homatropine after taking poison on reducing mortality resulted from organophosphorus poisoning.

MATERIALS & METHODS

This study is an empirical one which was done on 70 days old male rats with estimated weight of 200-250 gram. The animals were randomly divided into three groups of ten. They were in the same conditions in respect of nutrition and life environment. First, the rats were numbered and lain on dorsal surface and their weights were precisely measured, all the rats were subcutaneously injected with 25 mg/kg Dichlrovos poison (equivalent to intraperitoneal 50LD for rat) diluted in Saline 0.9% (the reason of selecting such dose of poison was being sure about all the rats of control group dying and being such dose of poison sufficient to cause death. In similar studies with such a dose 100% of rats in control group die within 4 to 12 minutes. As it continued, in the first group one minute after injection of poison, 0.5ml normal saline was intraperitoneally medicated, in the second group one minute after injection of poison , 20 mg/kg ophthalmic Homatropine 2% was intraperitoneally medicated, and in the third group one minute after injection of poison , 10 mg/kg Atropine was intraperitoneally injected. As it resumed, a trained person without any information about certain group of rats and according to the number of rats , monitored all of them in respect of the time passed from injection of poison, and vital signs including number of heart beats and breaths; and diagnosis of rats' death was on the basis of monitoring of rats' heart beats so that having straight electrocardiogram (null) was considered as death and also the rats who did not show any sign of death by 120 minutes, were regarded as alive. In all the processes of performing study Helsinki Rules 2008 concerning animal studies were followed, and also before the study being started an authorization to do study was taken from Ethics Committee of Kerman University of Medical Sciences . All the data were analyzed by central and dispersion indices and the statistical significance level was taken into account equal to $p<0.05$.

Findings

The average weight of rats in the first group was 234.86 ± 24.34 grams, in the second 241.92 ± 44.13 grams, and in the third one 232.25 ± 26.13 grams. In our study there was no difference among three groups of rats in respect of weight (p= 0.327). The average life time of rats in the first or control group was 15.00 ± 0.86 minutes which the minimum was 12 minutes and the maximum 19 minutes. 100% of the rats in this group died. the second or Homatropine group and the third or Atropine group all rats survived.

A comparison has been made in Diagrams 1 and 2 with respect to the number of heart beats and breaths. Based on our findings using Atropine can improve the symptoms related to the number of heart beats and breaths faster than Homatropine does, but the rate and duration of the effect of Atropine as compared with Homatropine was in number of breaths per minute (p= 0.412) and in number of heart beats per minute (p= 0.635), statistically no difference was observed between two groups.

DISCUSSION

Dichlorvos poison is a kind of organophosphate which inhibits Cholinesterase enzyme in mammalian tissues[9]. Compared to other organophosphates,Dichlorvos causes early signs of poisoning and subsequently fast signs of recovery to appear[10] that is as a result of fast metabolism of Dichlorvos and its fast excretion from the body[11]. Patients with poor respiratory function, convulsive disorders, hepatic dysfunctions or with recent history of being exposed to organophosphates are more vulnerable to Dichlorvos poisoning[12]. Likewise taking alcohol, high ambient temperature or UV can increase the effect of Dichlorvos. Dichlorvos poison is absorbed percutaneously and through digestive system[13]. Due to volatility,Dichlorvosis absorbed less than other organophosphates percutaneously[13]. 28.28 mg/kg intraperitoneal injection of poison can rapidly cause the jactitation, respiratory depression, and convulsion that this is why this poison was selected for being used in this study[14]. After immediate use of oxygen 100% or intubation, the base of treating median to critical organophosphorus poisoning is using the drugs which may block the muscarinic receptors or compete with Acetylcholine on the muscarinic receptors[15]. The common drug in treating organophosphorus poisoning is Atropine which is injected intravenously, and its use must continue until the symptoms of respiratory tract secretions and bronchospasm stop[16]. Of course more than three times using therapeutic dose of Atropine involving 0.5 mg per kg in the children may cause the critical symptoms of neurologic poisoning to appear[17]. After formal approval by FDA of America in 2003, the use of Atropine expanded as first priority in organophosphorus poisoning treatment throughout emergencies around the world, and this drug is available in almost all the emergencies and therapy clinics[18]. But sometimes some conditions like lack of facilities in ambulances or long distance from the patient's place to a standard medical center lead to the necessity of recognizing new drugs which besides having properties close to Atropine, would easily be available in health centers and offices of general physicians [19].

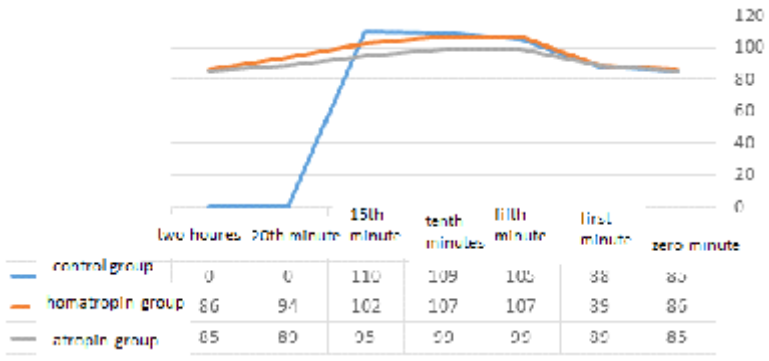
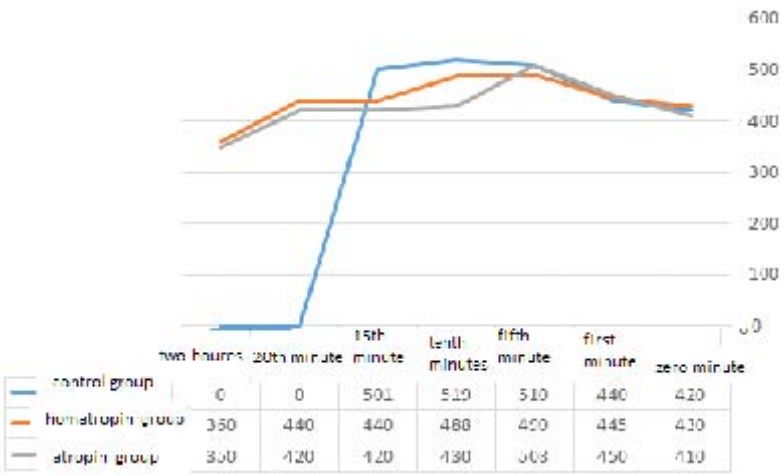


Fig. 1. The comparison of respiratoryrateamong the groups in various minutes



control group	0	0	501	519	510	440	420
homatropin group	350	440	440	488	450	445	430
atropin group	300	420	420	430	503	450	410

Fig. 2. The comparison of pulserateamong the groups in various minutes

So far the effect of various antimuscarininc drugs has been proved in organophosphorus poisoning treatment, as instance Nebulized Ipratropium has been proved as an effective drug in treating critical poisonings especially with respiratory symptoms[20]. Also different reports have been proved on Scopolamine Diphenhydramine, Glycopyrrolate Bromide, and Jimson extract[21]. But unavailability or difficulty in medicating or using these drugs have caused that they could not widely be used[22]. Homatropine is one other of these drugs, which is in the class of drugs anticholinergic, it is commonly used in the offices of general physicians. By blocking the muscarinic receptors in iris sphincter muscle and accommodator muscle of corpus ciliary, it prevents them being contracted by Acetylcholine and therefore causes mydriasisand paralysis of accommodation and it is used in routine visits by ophthalmologists and general physicians[23]. Also the fact that the eye drops in the market possess a high concentration of drug, has caused them to become the ideal drug which induce the possibility of more patients being treated by only one eye drop box, and due to Atropine's vials it is very ideal with respect to the cost and effectiveness. Furthermore, Homatropine eye drop includes some additional materials such as Sodium Chloride, Sodium Hydroxide, Hydrochloric acid, and water which are effective on regulating PH of the patient's blood and danger of poisoning with them in high concentration is little[24]. Moreover it has been demonstrated that the effects of systemic poisoning of this drug is less than Atropine[25]. Based on this particular study's findings, there is a significant difference in death rate between the rats used Homatropine and the rats used Atropine compared with rats in control group, and intraperitoneally ophthalmic Homatropine may be an adequate drug to treat the poisonings; it proved the findings of recent studies[8, 26]. In this study, it was also shown that there is no difference between ophthalmic Homatropine and Atropine with respect to the speed and power of effect on cardiorespiratory symptoms in patients. It is suggested that the effect of this drug be evaluated compared with Atropine as a clinical experience in emergencies of acute organophosphorus poisoning on human, and also a study be designed which may demonstrate that how much the short-term and long-term use of Homatropine would influence on the symptoms resulted from poisoning with organophosphates especially organophosphates with long half- life or dermal poisoning with organophosphates that the symptoms of poisoning appear with delay.

CONCLUSION

Based on our study's findings it was demonstrated that the early use of ophthalmic Homatropineintraperitoneally in rats can be effective as Atropine on preventing mortality resulted from organophosphates in acute

poisonings, and this drug in the form of eye drop can be used as an adequate and available prophylactic treatment in the emergencies of organophosphorus poisoning.

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