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Effects of Anticholinergic–Antiparkinsonian Drugs on Striatal Neurotransmitter Levels of Rats Intoxicated With Soman

TSUNG-MING SHIH,[1] BENEDICT R. CAPACIO AND LARA A. COOK

Pharmacology Division, U.S. Army Medical Research Institute of Chemical Defense, Aberdeen Proving Ground, MD 21010-5425

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SHIH, T.-M., B. R. CAPACIO AND L. A. COOK. *Effects of anticholinergic–antiparkinsonian drugs on striatal neurotransmitter levels of rats intoxicated with soman.* PHARMACOL BIOCHEM BEHAV 44(3) 615–622, 1993.—Antimuscarinic drugs possessing antiparkinson activity that were effective in preventing convulsions induced by the organophosphorus cholinesterase (ChE) inhibitor soman were studied for their effects on spinal cord ChE activity and striatal levels of acetylcholine (ACh) and catecholamines in soman-intoxicated rats. Either biperiden (BPR) or trihexyphenidyl (THP) was administered to rats at an anticonvulsant dose (0.125 mg/kg, IM) in the presence or absence of soman (100 μg/kg, SC). The time course (up to 2 h) for ChE activity and levels of ACh and catecholamines were measured after soman, BPR, THP, soman and BPR, or soman and THP treatment. Soman rapidly inhibited ChE activity (65–75%; 15–120 min) and increased ACh levels (35%; at 30 min). It did not affect norepinephrine or dopamine (DA), but elevated at later time points (60–120 min) levels of the DA metabolites, 3,4-dihydroxyphenylacetic acid (DOPAC) and homovanillic acid (HVA), thus indicating increased DA turnover. BPR and THP alone reduced striatal ACh level from control, but did not affect any other neurochemical parameters studied. THP and BPR each reversed the effects of soman on DOPAC and HVA levels, but neither affected ChE activity nor ACh level induced by soman. Thus, our findings suggest that the anticonvulsant effects of BPR and THP in soman poisoning may be attributed to their earlier reported muscarinic receptor blocking properties.

Soman Organophosphorus compounds Cholinesterase inhibitors Convulsions Anticonvulsants Anticholinergic compounds Antiparkinsonian drugs Acetylcholine Catecholamines Dopamine metabolism

ORGANOPHOSPHORUS (OP) cholinesterase (ChE) inhibitors such as soman can cause a progression of toxic signs, including hypersecretions, convulsions, and death (71). Pretreatment with carbamate ChE inhibitors (3,15,18,21,35) along with atropine sulfate (ATS) and oxime therapy significantly increases the 24-h survival rate in animals exposed to multiple LD_{50} doses of soman. However, this treatment regimen does not ameliorate soman-induced convulsions (15,21,34). Convulsive activity in OP intoxication creates a problem for medical management of exposed subjects and has been linked to irreversible brain damage (36,41,44,50). Therefore, the most effective employment of a treatment regimen against OP poisoning appears to require concomitant administration of an adjunct compound selected for its anticonvulsant activity (16).

The anticholinergic drug ATS is extremely valuable for treating OP poisoning (9,71). The usefulness of ATS in minimizing OP-induced lethality has been attributed, in the past, to its ability to antagonize muscarinic cholinergic hyperstimulation subsequent to OP-induced ChE inactivation and acetylcholine (ACh) accumulation at synapses (9,56,63,71). However, the ability of ATS to prevent seizures, convulsions, and neuropathology in soman poisoning was observed only at higher doses than have been previously utilized for protection against lethality (11,41,42). Thus, the role of anticholinergics in preventing soman-induced convulsions and brain damage is not entirely clear (22,52).

In the striatum, fine-tuned control of motor function is the result of feedback interactions between ACh and catecholamines (70). It has been suggested that anticholinergic drugs, such as biperiden (BPR) and trihexyphenidyl (THP), exert their antiparkinsonian effects on motor function by acting on

The experiments reported here were conducted according to the "Guide for Care and Use of Laboratory Animals" (1985), as prepared by the Committee on Care and Use of Laboratory Animals, National Research Council, NIH Publication No. 85-23. Portions of this work were presented in abstract form (59). The opinions or assertions contained herein are the private views of the authors and are not to be construed as official or as reflecting views of the Department of the Army or the Department of Defense.

[1] To whom requests for reprints should be addressed.

these striatal neuronal pathways (12). In addition, we previously found that BPR and THP are potent anticonvulsants in soman poisoning (11,61). Thus, the anticonvulsant effects of these drugs may be due to central actions on the cholinergic and catecholaminergic nervous systems. This implication is consistent with that of Shipley et al. (64), who suggested that soman-induced seizures are initiated by a combination of cholinergic and noradrenergic stimulation. To better understand the neurochemical basis for the anticonvulsant efficacy of these compounds in soman poisoning, the present study has been undertaken. Here, we report the effects of THP and BPR on some cholinergic parameters and catecholamine concentrations in the striatum of rats intoxicated with soman. The chemical structures of BPR and THP are shown in Fig. 1.

METHOD

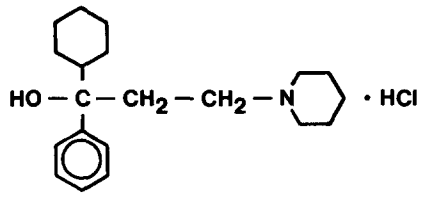
Animals

Male (Crl:CDBR^R VAF/Plus^R) Sprague-Dawley rats (*Rattus norvegicus*) weighing 200–300 g were used for this study. Rats were quarantined on arrival and screened for evidence of disease before they were released for the experiments. They were maintained, under an American Association for Accreditation of Laboratory Animal Care (AAALAC) program, in plastic cages (Lab Products, Inc., Maywood, NJ) on hardwood chip contact bedding (Beta-Chip, Northeastern Products Corp., Warrensburg, NY) changed two times each week, and were provided commercial certified rodent ration (Zeigler Bros., Inc., Gardners, PA) and tapwater ad lib. Animal holding rooms were maintained at 21 ± 2°C with 50 ± 10% relative humidity using at least 10 air changes per hour of 100% conditioned fresh air. Rats were on a 12 L : 12 D full-spectrum lighting cycle that was provided between 0600 and 1800 h. In all experiments, rats were randomly assigned to treatment groups. All drug injections were given between 0900 and 1200 h.

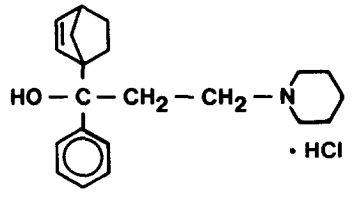
Materials

For drug administrations, saline (0.9% NaCl) injection, USP, was purchased from Cutter Labs., Inc. (Berkeley, CA), trihexyphenidyl HCl (THP) was purchased from Sigma Chemical Co. (St. Louis, MO), and biperiden HCl (BPR) was obtained from Knoll Pharmaceuticals (Whippany, NJ). Soman (pinacolyl methylphosphonofluoridate), obtained from the Chemical Research, Development and Engineering Center (Aberdeen Proving Ground, MD), was diluted in ice-cold saline prior to injection. BPR and THP were prepared and diluted in a vehicle containing 40% propylene glycol, 10% ethanol, 1.5% benzyl alcohol, and 48.5% distilled water. The volume of injection was 0.5 ml/kg. All drug solutions were prepared and injected separately. Soman was administered SC and the anticholinergic compounds IM.

For neurochemical analyses, deuterated ACh (98 atom % deuterium) was purchased from MSD Isotopes (St. Louis, MO), and 3,4-dihydroxybenzylamine HBr (DHBA), 3,4-dihydroxyphenylacetic acid (DOPAC), homovanillic acid (HVA), dopamine HCl (DA), and norepinephrine HCl (NE) were obtained from Research Biochemicals Inc. (Natick, MA). Other chemicals (analytic grades) were obtained commercially and used without further purification.



TRIHEXYPHENIDYL (MW = 337.92)



BIPERIDEN (MW = 347.45)

FIG. 1. Chemical structures of biperiden (BPR) and trihexyphenidyl (THP).

Experimental Design and Procedures

Animal and Tissue Preparations. Groups of rats (n = 6–12 per group) were injected with saline (0.5 ml/kg, IM), soman (100 μg/kg, SC; equivalent to 0.9 × LD_{50}), BPR (0.125 mg/kg, IM), THP (0.125 mg/kg, IM), soman immediately followed by BPR, or soman immediately followed by THP. These studies were carried out to study the effects of each compound alone and see whether BPR and THP could antagonize the neurochemical effects of soman. A saline-treated group served as control for comparison. Separate groups of animals were used for studies of cholinergic and catecholaminergic parameters.

Following drug injection and prior to tissue collections, animals were observed and examined (by palpation and touching) for abnormal gross behavioral changes. These included general activity (grooming, sitting, prostration), coordination (walking, ataxia, rolling, loss of righting reflex), salivation, lacrimation, diarrhea, muscle tone, and motor activity (muscle fasciculations, whole-body tremors, tonic or clonic convulsions). Many of these signs are characteristics of anti-ChE intoxication (4,32). Particular attention was paid to the motor activity because BPR and THP have been shown to prevent or reduce soman-induced convulsions in this species (11,61).

At different times (from 15–120 min) after injection, rats were killed by focusing a beam of microwave radiation (3.0 kW at 2.45 GHz) (Gerling-Moore Metabostat System, Gerling-Moore, Inc., Santa Clara, CA) on the head for 2.9 s for the ACh and catecholamine assay. Microwave irradiation of animals at the termination of the experiment halted brain enzymatic activity and facilitated the accurate determination of neurotransmitter concentrations (5,66). All animal experiments were performed between 0930 and 1200 h each day to minimize effects due to circadian variations in neurochemical measures (25,49,55).

Assay of Spinal Cord ChE Activity. Immediately following head-focused microwave irradiation, the spinal cord was obtained and the cervical segment, which had been affected by the microwave procedure (31), was discarded. We reported earlier that there is an excellent correlation between total ChE activity in the spinal cord and brain regions (31). Thus, the ChE activity measured in the spinal cord can be an indicator of the ChE activity in the brain of microwaved rats. The thoracolumbar segment of the spinal cord was homogenized in

1% Triton X-100 in ice-cold saline (1 : 15; w : v) by 8–10 strokes using a glass/glass Potter-Elvehjem homogenizer. Homogenates were centrifuged at 4°C for 10 min at 15,000 × *g*, and the supernatant was analyzed. It was found that following Triton X-100 solubilization 99.5% of ChE activity was in the supernatant. Total ChE activity (using acetylthiocholine as substrate) and total protein concentration (using biuret reaction) were determined by the automated spectrophotometric method of Groff et al. (23) using a Technicon Autoanalyzer System (Chauncey, NY) described elsewhere (62).

Analysis of ACh. Following head-focused microwave irradiation, the striatal region of the brain was dissected. ACh was extracted from this brain region by the method of Jenden and Hanin (30), and the concentrations were quantitatively determined by a gas chromatograph-mass spectrometric method described previously (56,62), using deuterated analogs of ACh as the internal standards.

Analysis of Catecholamines. Following head-focused microwave irradiation, the striatal region of the brain was dissected, weighed, and immediately homogenized in 2.0 ml perchloric acid (0.05 N) containing DHBA as the internal standard. The homogenates were centrifuged at 4°C for 20 min at 15,000 × *g* and the supernatant was saved for assay. Catecholamines and their metabolites were determined electrochemically following separation by reversed-phase (C_{18} column), ion-pairing high-pressure liquid chromatography (BAS-200A Analyzer, Bioanalytical Systems, Inc., West Lafayette, IN). The mobile phase (pH 3.1) consisted of 0.075 M monochloroacetic acid, containing 1.0 mM EDTA, 1.8 mM sodium octyl sulfate, and 1% acetonitrile in distilled water. The mobile phase was pumped at a flow rate of 1.0 ml/min and the oxidation potential was set at +750 mV.

Data Analysis

Total ChE activity in the spinal cord was calculated as μmol acetylthiocholine hydrolyzed/g protein/min (μmol/g/min). Striatal ACh and catecholamine concentrations were quantitatively determined as nmol/g wet tissue. All data were then expressed as % of control activity. Changes in striatal ACh, catecholamines, and spinal cord ChE activities were analyzed using the two-way analysis of variance (ANOVA) (treatment × time) followed by Duncan's multiple-range test. If significant treatment × time interaction was observed, overall treatment differences were compared at individual time points with a one-way ANOVA followed by Duncan's multiple-range test. A two-sample *t*-test was also used to assess significant changes from control for each time point in each treatment group. Statistical analyses were accomplished with SAS/STAT software (SAS Institute Inc., Cary, NC) and Number Cruncher Statistical System software (Dr. Jerry L. Hintze, Kaysville, UT).

RESULTS

Effects on General Behavior

Soman at a dose of 100 μg/kg SC (0.9 × LD_{50}) produced typical cholinergic signs of OP intoxication, appearing between 5–10 min after injection. Rats were observed to have tremors, muscle fasciculation, salivation, diarrhea, repetitive head bobbing, tonic and overt clonic convulsions, Straub tail, and respiratory distress. Shortly after, animals' limbs and toes became rigid and stiff and they attempted to walk on their toes. Animals with hypersecretions exhibited labored breathing and gasping with head bobbing typical of soman intoxication. No overt disturbances in normal behavioral patterns or physical coordination were observed after treatment with BPR or THP alone (0.125 mg/kg, IM). Therapeutic treatment with BPR or THP appeared to block the occurrence of soman-induced convulsions and their effectiveness was qualitatively similar. At the expected time for convulsions, animals displayed only a lateral (side to side) head movement atypical of soman alone and soman-induced motor signs of intoxication were modified. Some animals showed muscle twitches and occasional whole body tremors, which occurred about 15 min after soman. Muscle fasciculations developed in most animals approximately 1 h after soman and lasted for the duration of the observation period. Subsequently, animals were weak but able to move about. Rats did not show soman-induced salivation, nasal secretion, or diarrhea.

Effects on Spinal Cord ChE Activity

The total ChE activity in the thoracolumbar segment of the spinal cord in control rats was 181.2 ± 6.5 μmol/g/min. Administration of BPR or THP alone did not affect the baseline ChE activity. On the other hand, soman significantly inhibited the total ChE activity by 65–75% beginning at 15 min and throughout the 2-h experimental period ($p < 0.01$). BPR or THP treatment did not reverse the inhibitory effects of soman on spinal cord ChE activity. The typical effects of THP on ChE activity are represented in Fig. 2.

Effects on Striatal ACh Concentrations

The effects of soman, BPR, and soman followed by BPR on striatal ACh concentrations are shown in Fig. 3. The control value for ACh concentrations in the rat striatum was 64.7 nmol/g tissue. ACh concentration measured at 30 min after soman was elevated by 35% above control ($p < 0.05$). Administration of BPR alone (0.125 mg/kg, IM) produced a significant ($p < 0.05$) decrease of ACh concentrations from controls 30, 45, and 60 min after administration. Increases (ranging from 16–30%) of ACh were observed in the group of rats challenged with soman and treated with BPR. At 30 min after administration, the BPR-alone group had a significantly lower ACh concentration from control than soman and

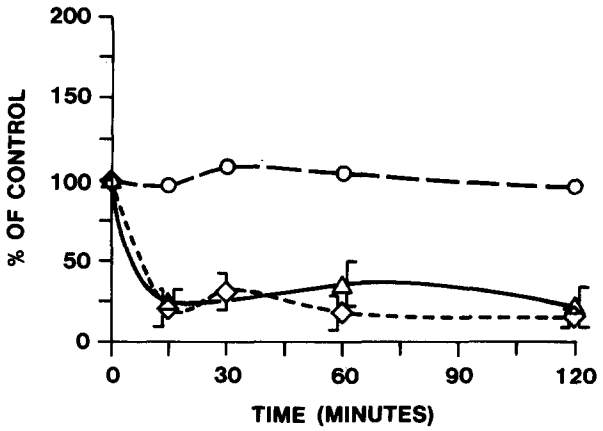


FIG. 2. Time course effects of trihexyphenidyl (THP) (0.125 mg/kg, IM; ○), soman (100 μg/kg, SC; △), and soman plus THP (◇) on cholinesterase (ChE) activity of thoracolumbar segment of the rat spinal cord. Data represented mean ± SEM with 6–12 rats for each time point in each group. See the text for statistical significances.

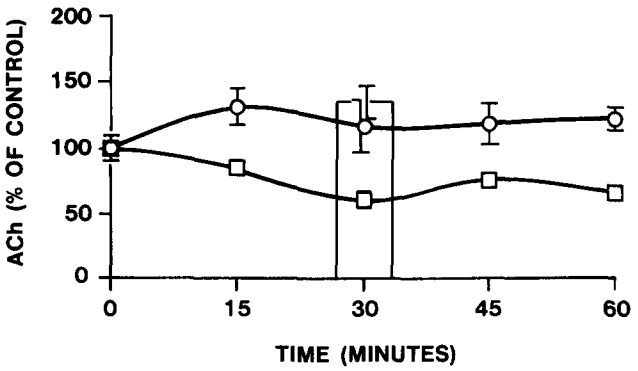


FIG. 3. Time course effects of biperiden (BPR) (0.125 mg/kg, IM; □), soman (100 μg/kg, SC; only 30 min point; bar), and soman plus BPR (○) on striatal ACh concentrations. Data represented mean ± SEM with 6–12 rats for each time point in each group. See the text for statistical significances.

soman + BPR groups ($p < 0.05$). Similar results were obtained with the treatment of THP.

Effects on Striatal Catecholamine Concentrations

The mean control values for NE, DA, DOPAC, and HVA in the rat striatum were 113.6, 4,951.2, 159.3, and 185.5 μg/g, respectively. Figure 4 shows the results obtained with THP. Soman, THP, and soman plus THP treatments did not reliably change striatal baseline NE levels (Fig. 4a). A transient increase of DA (Fig. 4b) from control was observed only at 60 min after soman administration. A significant decrease of DA from control was observed at all time points for soman + THP group ($p < 0.05$). Significant changes from control were observed in striatal DOPAC (Fig. 4c) and HVA (Fig. 4d) concentrations. Soman elevated levels of both DOPAC and HVA, the metabolites of DA, at 60 and 120 min after administration. Although THP treatment alone did not affect the concentrations of DOPAC and HVA, it did reduce the effects of soman on levels of these two DA metabolites. Soman had significantly greater levels of DOPAC at 60 and 120 min than THP treatment ($p < 0.05$). Other significant treatment differences were observed, but none were representative of a consistent change. Similar results were found with BPR treatment.

DISCUSSION

In the past, antidote development for OP poisoning has primarily focused on decreasing lethality after exposure. Although convulsions and seizures have been recognized as toxic signs following OP poisoning, they were considered primarily a factor that complicated definitive treatment of the more immediate life-threatening effects nerve agents can have on respiratory function. The utility of including an effective anticonvulsant in the medical management of nerve agent poisoning has not been a priority until recently, when reports of animal studies showed that carbamate pretreatment and subsequent therapy (ATS alone or together with oxime ChE reactivators) improved significantly the number of survivors after OP intoxication (3,15,18,21,26,35,37). However, this treatment regimen (pyridostigmine, ATS, and oxime) provides only minimal protection against OP-induced tremors and convulsions (21,34). Convulsive activity produced by OP anti-ChEs has been linked to postexposure brain neuropathology (36,41,44,50). Therefore, a thorough understanding of the central actions of OP nerve agents is important for the prevention of convulsive activity and associated brain damage (16).

It has been suggested that diazepam is the drug of choice in addition to traditional ATS and oxime therapy for the symptomatic relief of seizures (38,39,54), convulsions (6,72), and protection against postseizure brain damage (27,40) in OP poisoning. However, diazepam possesses many undesirable side effects (2,14,53).

In OP poisoning, a dual antidotal mechanism of atropine-like compounds has been noted by Green et al. (22), namely, protection against lethality as well as protection against convulsions. Recently, we reported a number of anticholinergic drugs, including BPR and THP, that also possess antiparkinsonian activity, to be much more potent anticonvulsants than ATS against soman (11,61). Thus, these compounds may be more suitable than ATS for OP therapy.

In our earlier studies (11), we found that all of the tertiary antimuscarinic compounds tested demonstrated activity against soman-induced convulsions. The observation that quaternary analogs completely lacked this activity suggested that central cholinergic mechanisms are primarily implicated in soman-induced convulsions. In addition to the observation that both THP and BPR are potent anticonvulsants to soman (11), these drugs have also been utilized to treat disease states that appear to involve neurochemical imbalances between dopaminergic and cholinergic mechanisms of the striatal motor nuclei, including Parkinson's disease (12), drug-induced parkinsonism, and Huntington's disease (70). Hallmark clinical signs of these disorders include muscle rigidity, tremors, and dyskinesias (17,70). Chemical potentiation of central cholinergic mechanisms by compounds such as the carbamate ChE inhibitor physostigmine produces a state of rigidity and tremors that, on neurophysiological grounds, closely resembles symptoms of clinical parkinsonism (1). As we reported earlier (11), soman produced tremors and rigidity in rats similar to, but more persistent than, those produced by physostigmine. Thus, some of the effects observed after soman may be due to actions at striatal cholinergic and catecholaminergic neurotransmitter systems. These observations are further strengthened by the suggestion that soman-induced seizures are initiated by a combination of cholinergic and noradrenergic hyperstimulation (64). Because the striatum possesses a well-characterized cholinergic and catecholaminergic neuronal circuitry and an abundance of both ACh and catecholamine neurotransmitters, this brain region provides an opportunity to study the effects of soman on these two systems concomitantly. Therefore, to elucidate the neurochemical mechanism of these compounds in soman poisoning we further examined the effects of BPR and THP on cholinergic and catecholaminergic neurotransmitter system parameters in the striatum.

Administration of soman at a dose of 0.9 LD_{50} to rats induced within 5–10 min the characteristic cholinergic symptoms that progressed and lasted for the duration of the experiment (up to 2 h). Neurochemically, there was an immediate and precipitous depression of total ChE activity (60). The spinal cord ChE activity, a good indicator of brain ChE activity in head-microwaved rats (31), reached its lowest level of 25% of control at 15 min and remained at that level for the rest of the experimental period. The striatal ACh concentrations were elevated to 35% above control at 30 min, consistent with our earlier report (56), which demonstrated that ACh in this brain region in this species following the same toxic dose of soman occurred 5 min after exposure, reached a plateau

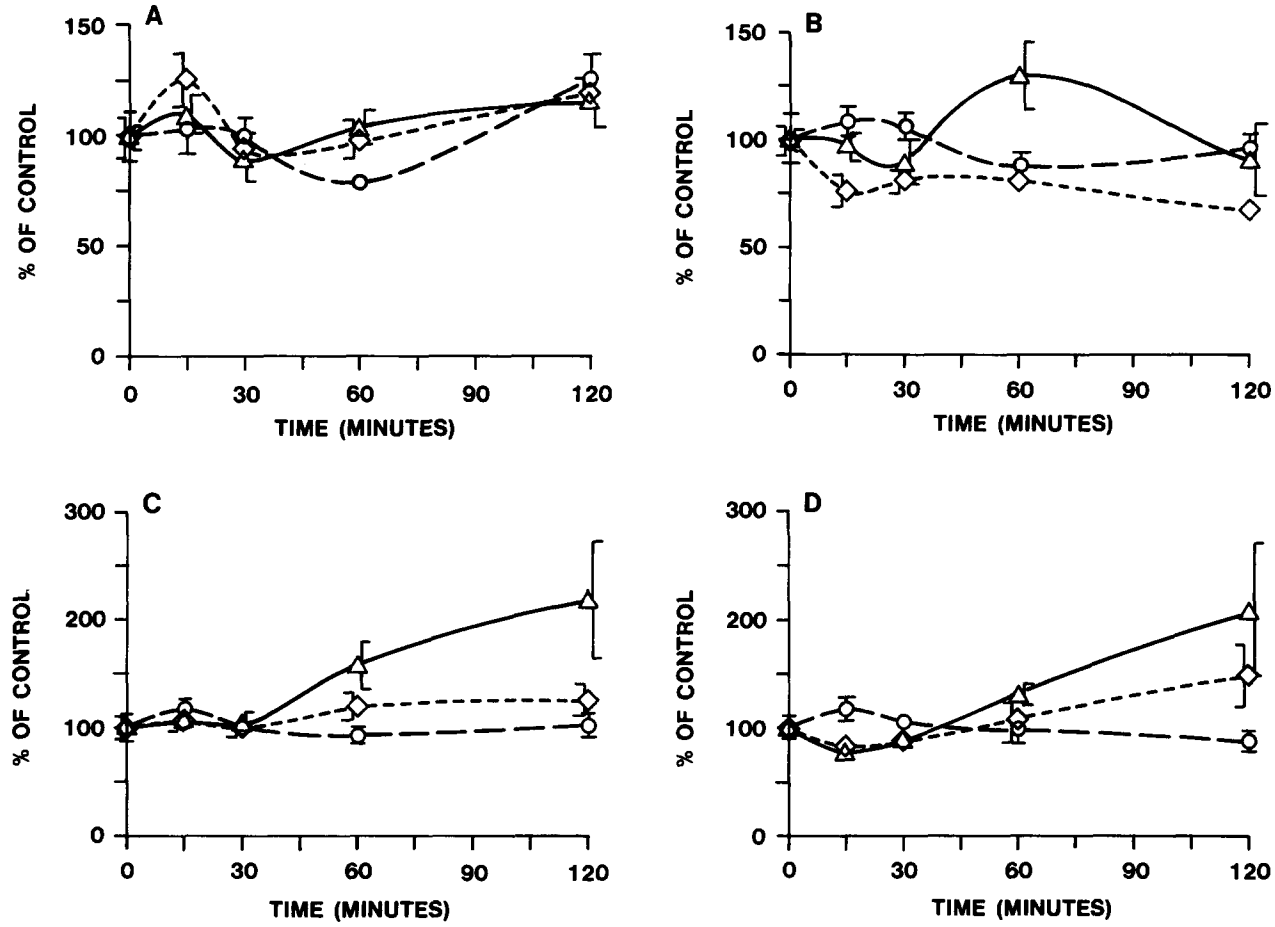


FIG. 4. Time course effects of trihexyphenidyl (THP) (0.125 mg/kg, IM; ○), soman (100 μg/kg, SC; △), and soman plus THP (◇) on striatal norepinephrine (NE) (a), dopamine (DA) (b), dihydroxyphenylacetic acid (DOPAC) (c), and homovanillic acid (HVA) (d), concentrations. Data represented mean ± SEM with 6–12 rats for each time point in each group. See the text for statistical significances.

level at 20 min, and remained there for several hours. Soman intoxication did not change striatal NE concentrations, but there was a transient increase of DA that occurred at 60 min after soman. At that time point (60 min) and beyond (120 min), the levels of DA metabolites DOPAC and HVA were markedly increased in the striatum, suggesting an increased DA utilization and increased turnover of DA. These changes occurred much later in time than the changes in cholinergic parameters and animals' symptoms of intoxication were as debilitating as during the earlier time point. Therefore, these findings are interpreted as a compensatory response of the dopaminergic system to the hypercholinergic activity in the striatum. These neurotransmitter changes obtained in these rat studies are in agreement with those reported in the guinea pig after administration of soman (19).

In the present study, animals treated with BPR or THP completely prevented soman-induced hypersecretions and motor convulsions. The antisecretory and anticonvulsant effects of BPR and THP confirmed our previous report (11), although some differences between the two studies exist. In that earlier study, the anticonvulsant ED_{50} value of BPR and THP was calculated to be 0.125 mg/kg with a soman challenge dose of 1.8 LD_{50}. In the present study, the same dose of BPR and THP was used but the soman dose was reduced by half (0.9 LD_{50}). In the present paradigm, the neurochemical effects induced by soman on DA metabolites, DOPAC and HVA, were reduced by administration of the antimuscarinic-antiparkinsonian drugs BPR and THP. The levels of NE, DA, and its metabolites were essentially the same as the control throughout the 2-h experimental period. Because the enhanced DA utilization caused by soman administration was found late in the neurochemical processes, this reduction may be explained as an indication of the immediate turn-off by BPR and THP of the hypercholinergic activity caused by soman, thereby eliminating the compensatory increase in the striatal dopaminergic activity. Because the antidotal action of ATS and its analogs in OP poisoning is in general attributed to their ability to block the cholinergic muscarinic receptors (10,71), the muscarinic blocking action may serve to preserve dopaminergic-cholinergic equilibrium in the striatum at times when the critical balance is disrupted.

The anticonvulsant activity of BPR and THP could not be attributed to their action on ChE activity because neither BPR or THP administration alone altered the ChE activity nor any of them reversed soman-induced ChE activity. It was found, however, that BPR and THP treatment alone reduced total ACh levels in the striatum. These effects of BPR and THP are typical of anticholinergic compounds. There is strong

pharmacological evidence for the presence of inhibitory presynaptic muscarinic receptors located both in the periphery and the brain (29,68). These receptors are believed to play a regulatory role in ACh release (51,68,69). Thus, blockade at these receptors by antimuscarinic drugs such as ATS, BPR, and THP has been shown to increase the release of ACh from brain both in vivo (24,67) and in vitro (7).

The increased ACh release and subsequent hydrolysis at synapses could account for the decreased total brain regional concentrations of ACh following anticholinergic drug administration observed in the present study and by others (20,28,33,58). However, the effects of anticholinergic compounds on total striatal ACh concentrations may not account for the anticonvulsant activity of BPR or THP because these drugs did not reverse the elevated ACh induced by soman. In addition, a dual and contradictory effect of BPR or THP in OP poisoning might be operating: Antimuscarinic drugs block postsynaptic muscarinic receptors on the one hand while at the same time stimulating presynaptic ACh release. This is consistent with the observations that without concurrent treatment of oxime to reactivate phosphorylated ChE, the benefit and protective potency (protective ratio = 1.2) of ATS therapy against soman lethality are minimal (63). Therefore, the muscarinic receptor antagonist properties of BPR and THP may principally be responsible for their antisecretory and anticonvulsant actions in soman poisoning. It should be noted, however, that even though hypersecretions and convulsions induced by soman may be terminated by the treatment of BPR or THP, other toxic effects of soman, such as muscle fasciculations, still persist. Therefore, to restore fully the functional capacity after OP nerve agent exposure, administration of drugs acting at the neuromuscular junction may be necessary. It has been reported that oximes that reactivate the phosphorylated ChE will restore the functional capacity at the neuromuscular junction (73,75).

BPR and THP utilized in this study have recently been reported to also possess *N*-methyl-*d*-aspartate (NMDA), an excitatory amino acid (EAA) subtype, receptor antagonist properties (48). EAAs, such as glutamate and aspartate, have been associated with brain neurotoxicity by their excitotoxic mechanisms (46,47) and have been identified as playing important roles in neurodegenerative diseases (45) and possibly parkinsonism. Compounds that block the neurotoxic actions of EAA at NMDA subtype receptors are being investigated as potential anticonvulsants and neuroprotective agents (45). For example, the noncompetitive NMDA antagonist MK-801 has been shown to effectively block soman-induced seizure activity, convulsions, and brain neuropathology (8,57). Procyclidine, an antimuscarinic drug that has both antiparkinsonian and NMDA antagonist activity, when administered after onset has been shown to terminate ongoing soman-induced convulsions and prevent brain neuropathology (52). In the literature, there is an inconsistency between the findings of Olney et al. (48) and Cestari et al. (13) with regard to the effect on NMDA receptor activity of scopolamine, which has been shown to be one of the most effective anticonvulsants in soman poisoning (11,26). Thus, although some of the beneficial effects of anticholinergic compounds may be mediated through EAA systems the involvement of NMDA receptors in the protective actions of BPR and THP requires further investigation.

Although cholinergic agents such as soman precipitate convulsive activity, the neuropathology does not appear to be due to a direct neurotoxic action of soman, high levels of ACh, or hypoxic-anoxic injury (42). McDonough et al. (42) and Shih (57) postulated that soman-induced convulsions are initiated by hyperstimulation of the cholinergic system (primarily muscarinic), and propagation of convulsions and subsequent neuropathology occur through the recruitment and cascade of other excitotoxic effects (74). This is consistent with our present findings and those of others (52,65). From the review of available literature, OP-induced neurotoxicity is unique in that the primary pathology involves cholinergic receptors being continuously flooded with ACh. If this primary source of cholinergic excitatory drive is immediately and effectively countered at the cholinergic receptors, pharmacological intervention at other receptor systems is probably not necessary (11,43,52). On the other hand, if this primary source of cholinergic hyperactivity is not effectively countered early in the process, pharmacological intervention at other receptor systems, such as catecholamine and NMDA, may be beneficial in protecting against convulsions and subsequent neuropathology (8,57,65).

In summary, the results from this study demonstrated that the OP nerve agent soman exerts some delayed effects on striatal DA metabolism in addition to its rapid action on the cholinergic nervous system, namely, inhibition of ChE activity with subsequent elevation of ACh at synapses. The increase in level of DA metabolites, DOPAC and HVA, starting at about 1 h, suggests that soman may be increasing the DA turnover rate in compensating for hypercholinergic activity in this brain area. The neurochemical effects induced by soman on DA metabolites were blocked by the therapeutic administration of the antimuscarinic-antiparkinsonian drugs BPR and THP, while at the same time soman-induced convulsive activity was stopped. On the other hand, soman-induced perturbations in the cholinergic parameters such as depressed ChE activity and elevated striatal ACh levels were not reversed by these drugs. Thus, our findings suggest that the anticonvulsant effects of antiparkinson drugs BPR and THP in soman poisoning may be attributed to their ability to block central cholinergic muscarinic receptors that are activated early in the process.

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