

THE EFFECTS OF HS-3 AND HS-6
ON CARDIOVASCULAR CHANGES
IN RATS CAUSED BY SOMAN

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The effects of HS-3 and HS-6 on cardiovascular changes due to poisoning with the LD₅₀ of soman were studied in anaesthetized rats. Both HS-3 and HS-6 were found to antagonize the changes in arterial blood pressure and heart rate. HS-3 was more effective than HS-6 in neutralizing the changes in cardiovascular performance induced by the central action of soman. The two oximes produced an antagonistic effect on some abnormalities in the electrocardiogram caused by soman. The primary haemodynamic effect of soman and its antidotes reflected itself on peripheral vascular resistance, the alteration in cardiac output being secondary, in the course of the homeostatic control of arterial pressure.

The effects of soman are characterized by a great variety of changes in cardiovascular performance. The unique »biochemical lesion« in soman poisoning, i. e. the blockade of acetylcholinesterase which takes place in nicotinic and muscarinic peripheral synapses as well as in cholinergic synapses in central structures engaged in both sympathetic and parasympathetic control, produces diverse final effects on cardiovascular performance (1).

The conventional oxime antidotes for organophosphorous compounds are almost completely ineffective in soman poisoning since they are incapable of reactivating the soman inhibited cholinesterase after dealkylation has occurred (»ageing« of the enzyme) and since most of them do not pass the blood-brain barrier (2). Among the few that are effective are bisquaternary mono-oximes HS-3 (N, N-oxydimethylene-bis (pyridinium-2,4-aldoxime) — dichloride) and HS-6 (N, N-oxydimethylene-bis /pyridinium-2 aldoxime 3 — carbamido/-dichloride) (3).

The aim of this work was to study the effects of HS-3 and HS-6 on the cardiovascular changes in rats poisoned with an LD₅₀ of soman,

in particular changes in blood pressure, heart rate, cardiac output and total peripheral resistance. As soman phosphorylated acetylcholinesterase »ages« very rapidly investigations were also undertaken to gain an insight into the antidotal and protective effects of HS-3 and HS-6, if any, in soman poisoning, with regard to the cardiovascular changes that occur during the first ten minutes of organophosphorus compounds administration.

MATERIAL AND METHODS

Wistar rats of both sexes from a substrain raised in our laboratory were used in the experiment. Cardiac output and total peripheral resistance were determined in male rats, weighing 300 — 400 g. In all other experiments female rats (180 — 220 g) were used. Animals were anaesthetized with sodium pentobarbital (Nembutal, 35 mg/kg) intraperitoneally. The rats were ventilated with a pump with intermittent positive pressure through a tracheal cannula. Arterial pressure tracing was recorded on one channel of a direct writing recorder (Physiograph Four, Narco Bio Systems) through a PE-50 catheter introduced into the femoral artery and connected to a low volume displacement mechanoelectric transducer (Statham P₂₃ Db). The mean blood pressure was obtained by electronic integration. Cardiac output was measured with a modification of dye dilution method on a Beckman cardiodensitometer (4). Total peripheral resistance was calculated by dividing the mean femoral artery pressure by the cardiac output. The LD₅₀ of soman (80 µg/kg) was injected through a PE-20 catheter inserted into the jugular vein. A separate PE-20 catheter placed in the opposite jugular vein was used for the injection of antidotes. Antidotes were stored as a powder and dissolved in saline immediately before use. HS-3 (50 mg/kg) and HS-6 (100 mg/kg) were injected i.v. 1 or 10 minutes before or 10 minutes after the administration of the LD₅₀ of soman. Relevant haemodynamic variables were measured immediately before the injection of soman or antidote, one minute after and at 5-minute intervals thereafter for half an hour. The electrocardiogram was recorded by means of standard bipolar leads.

The rats were divided into 11 groups, of 10 animals each.

Female rats

- I soman plus HS-3, 1 minute after soman
- II soman plus HS-3, 10 minutes after soman
- III soman plus HS-6, 1 minute after soman
- IV soman plus HS-6, 10 minutes after soman
- V HS-3 plus soman, 1 minute after HS-3
- VI HS-3 plus soman, 10 minutes after HS-3
- VII HS-6 plus soman, 1 minute after HS-6
- VIII HS-6 plus soman, 10 minutes after HS-6

Male rats (cardiac output)

IX soman

X soman plus HS-3, 30 seconds after soman

XI soman plus HS-6, 30 seconds after soman

The statistical treatment of the results included the determination of the means, the calculation of the standard error of the means and the testing of differences by means of unpaired Student's t-test (5).

RESULTS

Figure 1 shows typical changes produced by an injection of the LD₅₀ of soman: a clear rise of arterial pressure and a small and short-lasting decrease in the heart rate. The injection of an antidote one minute

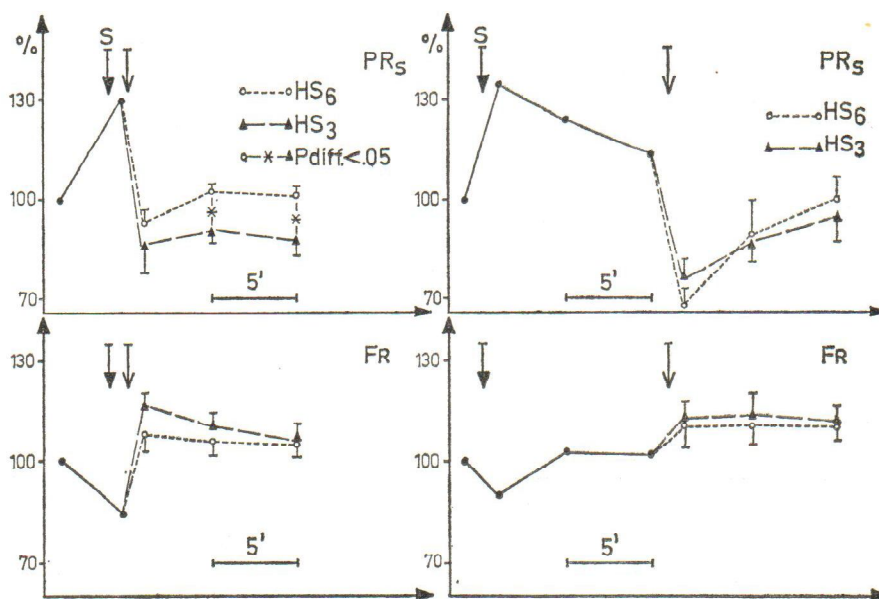


Fig. 1. Effects of HS-3 and HS-6 administered 1 (left side) or 10 minutes (right side) after the LD₅₀ of soman. PR_s — systolic arterial pressure. FR — heart rate. Values are expressed as a percentage of the control values. 100% systolic arterial pressure equals 17.8 kPa (134 mm Hg). 100% mean heart rate corresponds to 5.83 Hz (350 beats/min). The arrow marked S indicates the moment of injection of soman; the unmarked arrow indicates the time of injection of HS-3 or HS-6. Relevant variables were measured immediately before an injection of soman or antidote, one minute after and at 5-minute intervals thereafter

after soman (Fig. 1, top, right side) decreased the hypertension induced by soman to the control level (HS-6) or even lower (HS-3). The blood pressure after HS-3 was significantly lower than the arterial tension after HS-6. Both HS-3 and HS-6 produced a prompt corrective on the bradycardia produced by soman (Fig. 1, bottom, right side). When administered 10 minutes after the injection of soman, HS-3 and HS-6 caused a pronounced and statistically significant drop in blood pressure (Fig. 1, top, left side). There was some acceleratory action on the heart rate (Fig. 1, bottom, left side). Differences between the effects of HS-3 and HS-6 when administered 10 minutes after soman were negligible.

The pretreatment with HS-3 or HS-6 affected the cardiovascular action of soman in the following way: both antidotes produced a significant decrease in blood pressure when given before soman (Fig. 2), which persisted throughout the observation period. An injection of soman 1 or 10 minutes after HS-3 or HS-6 led to a hypertensive response similar, essentially to soman's effect in previously nontreated rats. However, in this series of experiments the bradycardic effect of soman was completely absent (Fig. 2).

When administered immediately after the LD₅₀ of soman, HS-3 and HS-6 abolished the significant rise in total vascular resistance occur-

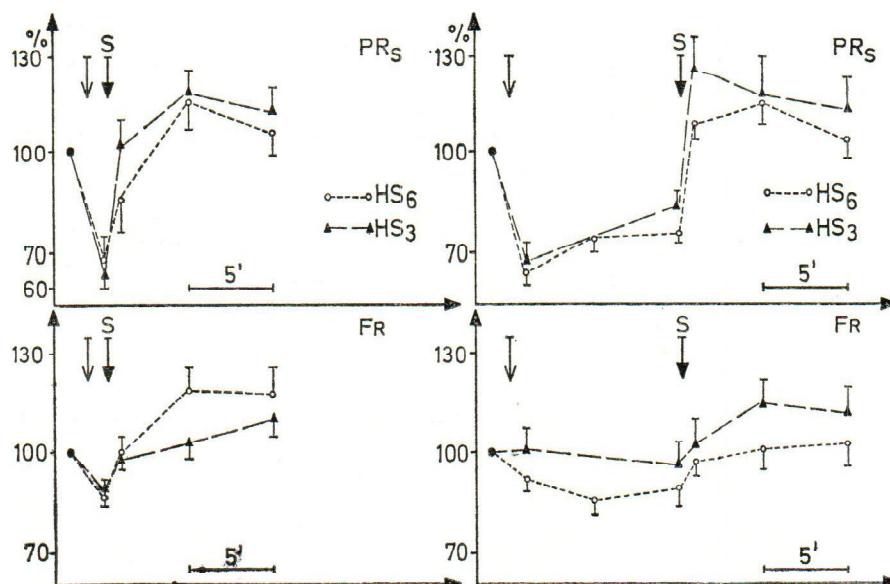


Fig. 2. Effects of HS-3 and HS-6 administered 1 or 10 minutes before the LD₅₀ of soman. The symbols are the same as in Fig. 1.

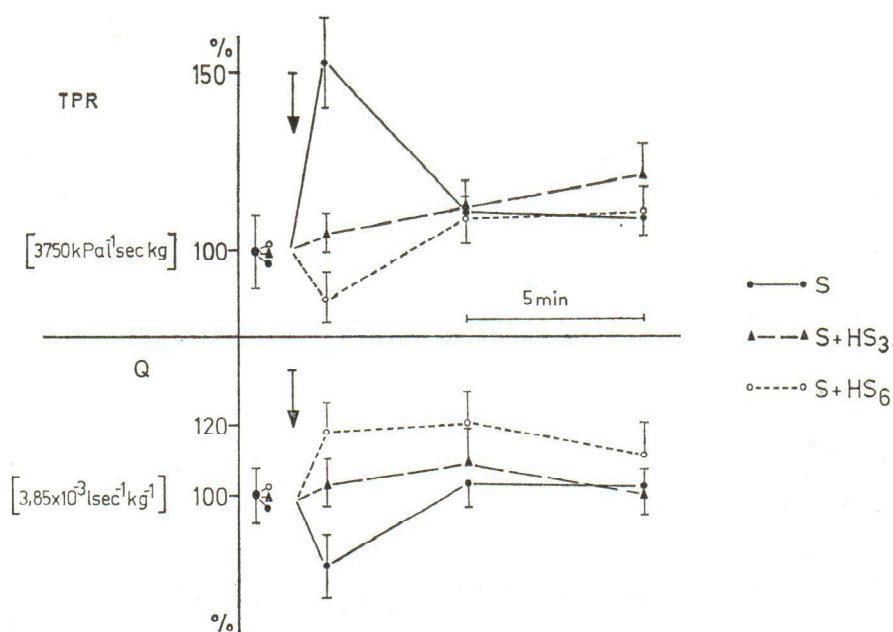


Fig. 3. Effects of soman and its antidotes on cardiac output (Q) and total peripheral resistance (TPR). S — rats treated only with soman; S + HS-3 — rats receiving HS-3 30 seconds after the LD₅₀ of Soman; S + HS-6 — rats receiving HS-6 30 seconds after the LD₅₀ of soman. Measurements were made before an injection of soman, one minute after and at 5-minute intervals thereafter

ring after the injection of soman alone. A decreasing effect on total peripheral resistance was more pronounced in rats treated with HS-6. With respect to the arterial mean pressure this effect was compensated by a rise in cardiac output. In rats treated only with soman the increase in peripheral resistance was of a magnitude sufficient to cause a rise in mean arterial pressure in spite of a drop in cardiac output (Fig. 3).

Both HS-3 and HS-6 were able to correct the high amplitude of the T wave in the electrocardiogram of rats treated with soman (Fig. 4). Antidotes often transformed the bradycardia with a frequency of about 2 Hz, which had been elicited by soman, to the previously existing heart rate (frequency around 6 Hz) (Fig. 5).

DISCUSSION

Both oximes used in this study, HS-3 and HS-6, proved their effectiveness against cardiovascular changes induced by the LD₅₀ of so-

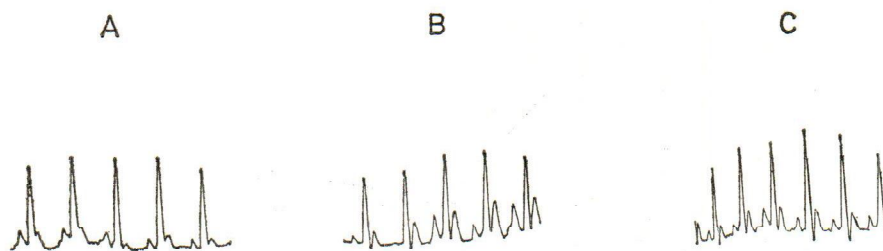


Fig. 4. Electrocardiogram. II standard lead. A — before treatment. B — 5 minutes after an injection of soman. C — 1 minute after an injection of HS-6

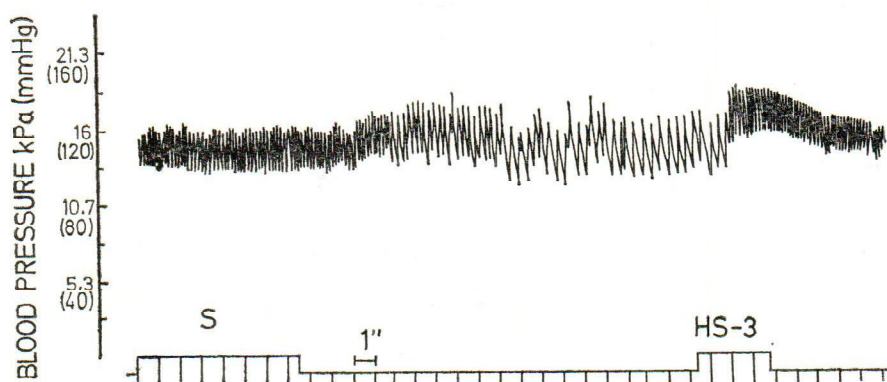


Fig. 5. Transcript of arterial pressure tracing. S — injection of soman. HS-3 — injection of HS-3

man. The fact that both antidotes were also effective against hypertension due mainly to the action of soman on central structures engaged in pressure control (1), could be considered as indirect evidence that HS-3 and HS-6 pass the blood-brain barrier, as suggested by *Kepner* and *Wolthuis* (2). In the absence of a significant difference in the effects of the two antidotes on the heart rate the significantly stronger anti-hypertensive effect of HS-3 compared to that of HS-6, could be explained by a greater permeability of the blood-brain barrier to HS-3, since the heart rate is a parameter affected in soman poisoned rats predominantly through cholinesterase blockade in peripheral parasympathetic synapses. Such a conclusion is not in accordance with that of *Schenk* and co-workers (3) drawn from a follow-up of the effects of

HS-3 and HS-6 on clinical symptoms in soman poisoned beagles. Their results suggested stronger anti-soman effects of HS-6 compared to HS-3.

Both HS-3 and HS-6 when used alone induced a significant decrease in blood pressure and a transient bradycardia. This finding suggests that at least a part of the protective effect of HS-3 and HS-6 on soman induced hypertension might result from direct oxime effects and not from the specific oxime induced reactivation of cholinesterase. However, since no difference in the hypotensive effect of the two oximes was noted but HS-3 exerted a significantly greater effect on the soman induced hypertension, it seems very likely that a part of the protective effect of the two antidotes resides on the specific oxime effect, i. e. on cholinesterase reactivation. This is further supported by the fact that both oximes readily corrected the soman induced decrease in the heart rate, although when applied alone both induced transient bradycardia.

Haemodynamically, the increase in blood pressure induced by soman is brought about by an increase in total peripheral resistance. A transient decrease in cardiac output after an injection of soman is most likely a result of the soman induced decrease in the heart rate. This conclusion is further supported by the finding that the correction by HS-3 and HS-6 of the soman induced bradycardia returned cardiac output to the control level. Both antidotes reduced the soman induced hypertension by an effect on total peripheral resistance. Thus, present findings suggest that vascular resistance is the primary target both with the poison and with the antidotes and that changes in cardiac output are secondary due to the homeostatic control of blood pressure.

Changes in repolarisation, i. e. in the T wave of the electrocardiogram elicited by soman, which were reversed by its antidotes favour the opinion that soman might also have some direct action on the myocardial cell and its membrane, besides the effects exerted on cholinergic synapses.

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Sažetak

EFEKTI HS-3 I HS-6 NA KARDIOVASKULARNE PROMENE
IZAZVANE SOMANOM U PACOVA

Dejstvo HS-3 i HS-6 na kardiovaskularne promene izazvane sa LD₅₀ somana ispitivano je u anestetsanih, veštački ventilisanih pacova. Rezultati ovih istraživanja upućuju na to da i HS-3 i HS-6 smanjuju promene arterijskih pritisaka u pacova tretiranih somanom te da je HS-3 efikasniji od HS-6 u neutralisanju promena u funkciji kardiovaskularnog sistema koje su izazvane centralnim dejstvom somana. HS-3 i HS-6 pokazuju antagonistički efekat na neke poremećaje u EKG-u do kojih dolazi kod trovanja somanom. Primarni hemodinamski efekat somana i njegovih antidota je na perifernom sudovnom otporu, dok su promene u minutnom volumenu srca sekundarne i deo su homeostatske kontrole krvnog pritiska.

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