level was depressed but increased by acute cold exposure. It has been shown that catecholamines inhibit insulin release mediated by an α-receptor- and stimulate it by a β-receptor-mechanism. Furthermore, insulin release is known to be enhanced by glucagon[17]. The diminution of basal insulin level by ACS in warm controls may be caused by the elimination of stimulatory action of catecholamines through a β-receptor and by the decrease of plasma glucagon. The increased glucagon release induced by acute cold exposure, together with the lack of α-receptor-mediated inhibitory action of catecholamines due to ACS in cold-exposed ACS rats, may augment insulin release.

Table 2 shows the changes in the levels of blood metabolites. The significant increase in the hematocrit was observed when both groups of rats were exposed to a cold environment, indicating hemoconcentration due to cold exposure[18]. ACS caused a significant decrease in the hematocrit in the warm controls but did not affect the cold-induced increase in the hematocrit.

Acute cold exposure induced a significant decrease of blood glucose level, suggesting that plasma glucose was utilized as the energy source in the cold as previously reported[7]. Plasma glycerol was significantly elevated when the animals in both groups were acutely exposed to cold. However, the extent of increment was significantly smaller in the ACS group than in the SV one. Plasma FFA was also increased by acute cold exposure in both groups. Its increment in the ACS group, however, was significantly larger than in the SV one ($p < 0.001$). The change in plasma glycerol level is considered to be a better index of magnitude of overall lipolysis, since plasma FFA released together with glycerol is metabolized more rapidly than glycerol[19]. Therefore, the plasma glycerol level is believed to reflect a degree of magnitude of lipolysis, that is, an activation of lipolysis is accompanied by a corresponding increase in the plasma glycerol level. From the present results, it is suggested that mobilization as well as utilization of lipids is significantly suppressed in the cold-exposed ACS rats. Although there is no doubt that sympatho-adrenal system is important for the cold-induced responses of animals and for cold acclimation, the increase in the plasma glucagon level by cold exposure may be also closely associated, at least in part, with the cold-induced responses, especially nonshivering thermogenesis, through its lipolytic action.

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Effect of dimethindene, an antihistaminic drug, on the transmembrane potentials of mammalian myocardium

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Summary. Dimethindene (DMI) decreased the maximum rate of rise of action potential (AP) without changing the resting potential in cat ventricular myocardium. DMI abolished the histamine-induced slow APs in left atria but not in right ventricular papillary muscles of guinea-pig, suggesting that DMI blocked the histamine H_1-receptors.

DMI (Forhistal®, Fenistil®) is a well-known antihistaminic drug used in the treatment of different allergies[3]. This activity of DMI has been attributed to antagonize the effects of histamine at the H_1-receptors[4]. It was demonstrated that the drug exerted many other actions, such as induction of histamine release, decrease of arterial blood pressure and peripheral resistance in anesthetized dogs[5]. Recently, it has been shown that DMI has an antiarrhythmic activity[6].

The aim of the present work was to study the effect of DMI on the normal and slow APs of the mammalian myocardium.

Methods. Experiments were carried out on isolated right ventricular papillary muscle of cat and guinea-pig, and on left atrial muscle of guinea-pig. The animals were anesthetized with ether, and the muscles were dissected from the heart as quickly as possible and mounted in an organ chamber. The preparations were driven electrically at 2.0 or 0.5 Hz. The transmembrane potentials were recorded by means of conventional glass microelectrode technique[7].

The slow response APs were elicited with histamine (10^{-5} M)) or caffeine (2 mM) in partially depolarized (up to -40 mV) left atrial and right ventricular myocardium of guinea-pigs. The membrane was depolarized by means of elevated K^+ (26 mM)-Krebs solution (an isosmolar substitution of K^+ for Na^+)[8]. DMI (Fenistil®, Zyma-Biogal) was freshly dissolved and added to the organ chamber containing Krebs solution (composition in mM): NaCl 118, KCl 4.7, $CaCl_2$ 2.5, NaH_2PO_4 1.0, $MgCl_2$ 1.2, $NaHCO_3$ 24.9, glucose 11.5, which was gassed with 95% O_2 and 5% CO_2 and kept at 37 °C.

Results. In the 1st series of experiments, the effect of DMI was examined on normal APs in right ventricular papillary muscle of cat. Figure 1 shows the dose-dependent effect of DMI. The control APs obtained in 5 preparations had the following parametes: overshoot $+21.4 \pm 0.7$ mV, the maximum rate of rise of AP ($\dot{V}_{max}$) 128.3 ± 0.5 V/sec, duration at 50% repolarization 170.5 ± 0.9 msec. The resting potential was -79.3 ± 1.2 mV (means $\pm$SEM of 5 experiments). Application of DMI (5×10^{-6} M) decreased the $\dot{V}_{max}$ to a

value of 58.1±0.6 V/sec, without changing the resting potential, at 2 Hz stimulation frequency. The DMI effect became more pronounced at higher concentrations. At a lower stimulation frequency (0.5 Hz), 5×10^{-6} M DMI decreased the $\dot{V}_{max}$ from 129.7±1.2 V/sec to 87.4±1.9 V/sec.

In the 2nd series of experiments, DMI was tested for its effects on the slow APs induced by histamine or caffeine in K^+-depolarized atrial and ventricular myocardium of guinea-pig. In these preparations the fast Na^+ channels were voltage-inactivated, whereas the slow Ca^{2+} channels remained fully available[8,9]. In left atrial myocardium, DMI (5×10^{-5} M) completely abolished the slow APs induced by 10^{-5} M histamine (fig.2,A), whereas the caffeine (2 mM)-induced ones were only slightly reduced by the same concentration of the drug (fig.2,C). On the other hand, in right ventricular myocardium, DMI (5×10^{-5} M) hardly decreased the amplitude and $\dot{V}_{max}$ of slow APs induced by either 10^{-5} M histamine (fig.2,B) or 2 mM caffeine (fig.2,D).

Discussion. Two main conclusions seem to arise from these experiments: 1. DMI exerts a quinidine-like membrane-stabilizing effect by decreasing the $\dot{V}_{max}$ of AP in cat ventricular myocardium. It has been demonstrated that DMI has a quinidine-like antiarrhythmic effect in different arrhythmia models[6]. The results presented here suggest that this favorable antiarrhythmic effect of DMI can be explained by the Na^+ channel blocking effect of the drug. Several drugs (e.g. quinidine, lidocaine, and procainamide) are known to have an antiarrhythmic activity related to their ability to reduce the $\dot{V}_{max}$ of AP in cardiac muscle (class 1 action)[10-12]. Since DMI more strongly decreases the $\dot{V}_{max}$ of AP at higher stimulation frequency than at lower ones, we suppose, on the basis of Hondeghem-Katzung model[13], that DMI exerts a use-dependent block on the fast Na^+ channels.

2. DMI is capable of selectively blocking the histamine H_1-receptors. Results obtained in experiments with K^+-depolarized heart preparations of guinea-pig indicate that DMI, even at high concentration (5×10^{-5} M), has only a very weak decreasing effect on the slow APs mediated mainly by slow Ca^{2+} channels. The caffeine-induced slow APs were decreased by DMI in neither atrial nor ventricular myocardium. Caffeine stimulates the slow Ca^{2+} channels directly[14]. The histamine-induced slow APs were diminished by DMI in left atrium but not in right ventricle. It has been shown that guinea-pig left atrium contains H_1-receptors, whereas the right ventricle contains H_2-receptors[15]. Therefore, it can be concluded that DMI, like mepyramine[16], has a specific H_1-receptor blocking activity, if we take into account that the slow AP-evoking effect of histamine is mediated by H_1-receptors in guinea-pig left atria and by H_2-receptors in right ventricular myocardium[17] We suppose that membrane-stabilizing property of DMI might not be related to any H_1-receptor blocking activity, and that DMI might release endogenous histamine[4], which can modify the direct effect of the drug.

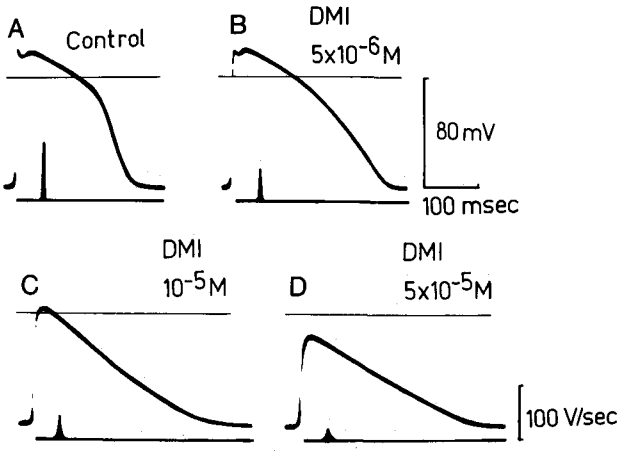

Figure 1. Dose-dependent effect of DMI on the transmembrane action potentials in right ventricular papillary muscle of cat. Tracings from top to bottom: zero potential level, action potential, maximum rate of rise. Bottom tracings were shifted to the right for easier analysis of $\dot{V}_{max}$. Stimulation rate: 2 Hz.

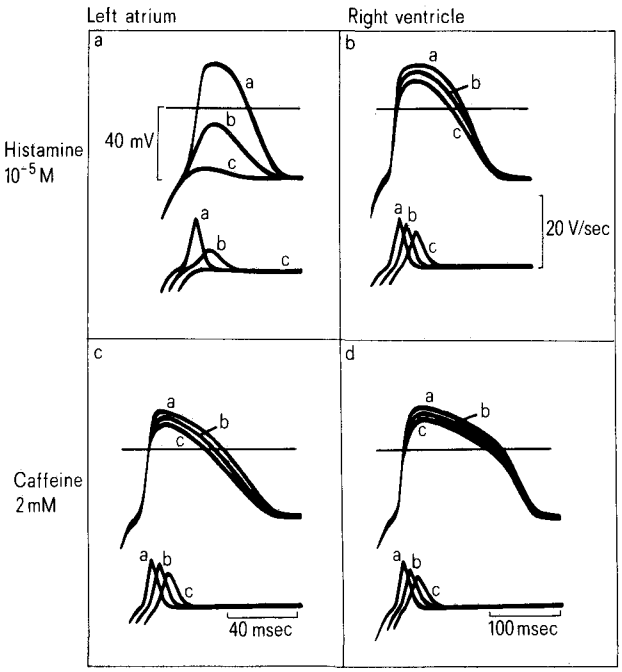

Figure 2. Dose-dependent effect of DMI on the slow APs restored by 10^{-5} M histamine in guinea-pig left atrium (A) and right ventricular papillary muscle (B) depolarized by 26 mm K^+-Krebs solution, and on the caffeine (2 mM)-induced slow APs in left atrium (C) and right ventricular papillary muscle (D). In each panels: a, control, b, 10^{-5} M DMI, c, 5×10^{-5} M DMI. Bottom tracings were gradually shifted to the right for easier analysis of $\dot{V}_{max}$. Stimulation rate: 0.5 Hz.

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The photoovicidal activity of plant components towards *Drosophila melanogaster*[1]

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Summary. Phenylheptatriyne, alpha-terthienyl, and 8-methoxypsoralen are 3 natural products which, in combination with long wavelength ultraviolet light (UVA), prevent the eggs of *Drosophila melanogaster* from hatching. Both phenylheptatriyne and α-terthienyl display ovicidal activity in the dark as well, but the irradiation step increased it 37- and 4333-fold respectively. The photoovicidal activity of 8-MOP was $\frac{1}{10}$ that of PHT. This new type of activity for natural products perhaps contributes to the natural protection of plants from insects.

A simple 'Drosophila test' was reported as a bioassay for insecticide activity in plant extracts, which disclosed that several naturally occurring polyacetylenic molecules had ovicidal activity toward *Drosophila melanogaster*[2-4]. The use of this test has now been extended to the determination of photoovicidal activity, and the feasibility of this method of insect control is illustrated with 3 naturally occurring molecules, phenylheptatriyne (PHT), alpha-terthienyl (αT, 2,2′; 5′, 2″-terthiophene), and 8-methoxypsoralen (8-MOP).

We are not aware of any previous reports on the photosensitized inhibition of the development of eggs in insect species, although short wavelength UV light alone can have this effect[5]. In our experiments, the light source had little emission in the short wavelength range, and most of it was removed by the pyrex filter used. The ovicidal activity of PHT had already been reported, but without the UV light treatment[4]. Both PHT and αT are found in plants of the family Compositae, while 8-MOP and other furanocoumarins are widespread in the families Umbelliferae, Rutaceae, Leguminosae, and Moraceae. All 3 compounds are well known sensitizers[6], particularly 8-MOP, for which a number of medical applications have been suggested[7]. This last compound has also been the topic of studies concerning plant-insect relationships, but the problem of specific toxicity to eggs was not discussed[8,9].

Materials and methods. An alcohol solution of sensitizer (20 µl) was added to a Whatman No. 1 filter paper disc, 1.5 cm in diameter. The solvent was evaporated, and the disc was saturated with distilled water. On a disc, a concentration of 11.5 µg/cm^2 corresponded to a sensitizer concentration of 1 g/l. Eggs less than 4 h old were planted on the discs in a darkroom dimly lit through amber Kodak OC Safelight filters. The eggs were incubated in total darkness, and the results provided the dark controls for the experiments in which the eggs were handled identically, except for a 45-min irradiation with long wavelength UV light (UVA) starting 1 h after the beginning of the incubation. The discs containing 10–40 eggs were in a petri dish covered with pyrex during the irradiations with a bank of 8 low-pressure tubes which had maximum emission at 350 nm (No. RPR-3500A, Southern New England Co, Hamden, Conn.). They were mounted horizontally 5 cm apart, 8.9 cm above the paper discs. At the surface of the discs, the light intensity was 13 J/m^2 · sec, as measured with a Yellow Springs Instruments radiometer Model 65A. The viability of the eggs decreased by less than 10% upon irradiation under the same conditions in the absence of added chemicals. No increased ovicidal activity was observed when the eggs were incubated in the dark over paper discs irradiated after addition of the chemicals. All the experiments were repeated at least 10 times with each compound at each concentration.

8-MOP was purchased from Sigma Chemical Co, PHT and αT were synthesized by Drs J.-P. Beny and S.N. Dhawan in our laboratories.

Results. The results illustrated in the figure indicate that all 3 compounds tested possessed photoovicidal activity toward *D. melanogaster*. In the dark, both PHT and αT displayed ovicidal activity with LD_{50} values corresponding to sensitizer concentrations of 0.13 and 1.26 g/l respectively. The value for PHT corresponds to 1.5 µg/cm^2, and is in good agreement with the published number of 2 µg/cm^2 (Nakajima and Kawazu[4]). In contrast, 8-MOP did not show any ovicidal activity in the dark at concentrations up to 10 times greater.

The increase in activity observed when the eggs were exposed to UV light for a short fraction of their incubation period was dramatic. Taking the concentration of sensitizer solution required for killing 50% of the eggs as the criterium, the activity of PHT was increased 37 times by the exposure to the UV light, from 0.13 to 0.0035 g/l (or from 1.5 to 0.4 µg/cm^2). With αT, the LD_{50} corresponded to a change from 2.6 to 0.0006 g/l (or from 30 to 0.007 µg/cm^2), or an increase in sensitivity greater than 3 orders of magnitude.

Finally, the photoovicidal activity of 8-MOP under the same conditions had a LD_{50} corresponding to a sensitizer concentration of 0.035 g/l (0.4 µg/cm^2). Although 10 times greater than that of PHT, this value is lower than found with either PHT or αT in the dark.

The relationship between the age of the eggs and the timing, duration, wavelength, and intensity of the UVA treatment remains to be determined. While most of the useful phototoxic properties of 8-MOP have been based on its ability to modify DNA, exceptions are known, in which singlet oxygen sensitization is the predominant pathway[10].