different concentrations and control for group. Lengthening of developmental time is a fairly good indication of somatic effects caused by the chemical in test substrate[6]. It is evident that the rate of development is prolonged even at the lowest concentration tested. The highest developmental delay was noticed in 30-mg concentration. Thus the mean developmental time is significantly different when compared to controls ($p < 0.05$, table). However, none of the concentrations had any discernible effect on the mean developmental time in either of the sexes. In most groups, the means for males and females did differ by less than 1 SD. The SD were similar in both sexes. Hence, these results clearly indicate that Dithane M-45 has a pronounced effect on the rate of development in *D. melanogaster*. Such a type of chemical effect on the rate of development was also demonstrated by other workers[6-10]. Bhowmik[11] has shown that Dithane M-45 inhibits the mycelial growth and sporulation in *Alternaria triticina*.

In addition to the rate of development, the other usable parameter for evaluating toxicity is lethality. Comparison between the control and treated series clearly shows that even the lowest concentration used caused about 17.86% lethality (table). The extent of lethality is directly proportional to the concentration. The male and female larvae exhibited the same sensitivity in each concentration, except in 20 mg, to which female larvae were more sensitive than males ($\chi^2 = 4.84$, $p < 0.05$; table). By calculating the analysis of variance, it has been shown that Dithane M-45 induced significant effect on viability in concentrations above 5 mg/100 ml food medium ($p < 0.05$). The present findings have revealed that the LC_{50} for Dithane M-45 on *D. melanogaster* is 17.5 mg/100 ml. Experiments with *Phytophthora* has revealed that Dithane M-45 at 5 ppm killed the zoospores[3].

The authors have observed that mortality occurs mostly during the larval stages and that first instar larvae are the most sensitive to 25- and 30-mg concentrations. This is strong evidence to indicate that concentrations of Dithane M-45 greater than 0.025% are lethal to various larval stages of fruit fly and the greatest fungicidal potency is expressed in the larval stages.

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An adrenergic participation subserving a positive inotropism and chronotropism of prostacyclin on isolated rat atria[1]

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Summary. The effects of prostacyclin (PGI_2) on the contractile frequency and on the isometric developed tension of spontaneously beating or paced isolated rat atria were explored. PGI_2 enhanced frequency and contractile tension, both effects being blocked by the presence of propranolol, or following a pretreatment with 6-OHDopamine.

Prostacyclin (PGI_2) a nobel prostaglandin formed by blood vessel walls, has been recently identified and proved to have powerful inhibitory properties of arachidonic acid and adenosine-diphosphate platelet aggregation[2-4]. PGI_2 has also been found to relax isolated strips of coronary arteries and to be the main prostaglandin released by the rat, rabbit and guinea-pig heart[5,6]. Furthermore other cardiovascular actions, namely lowering of systemic blood pressure, coronary vascular and total peripheral resistance without cardiostimulatory actions or changes in heart rate, have been described in the cat[7]. However, augmentation of heart rate and perfusion pressure in the isolated rat heart, and depression of diastolic blood pressure accompanied by tachycardia in rats and rabbits, were also documented[8,9]. Lefer et al.[7] suggested that the increased cardiac output in response to PGI_2 is an indirect (increased cardiac ejection in the face of increased venous return) rather than a direct cardiostimulatory effect[7]. In view of these contradictory findings, it was decided to explore the direct influences of PGI_2 on the isolated rat atria.

Methods. Male albino rats of the Wistar strain were decapitated; their atria removed and suspended in a modified Krebs-Ringer-Bicarbonate media gassed with 95% O_2–5% CO_2; maintained at a constant pH and temperature of 7.4 and 30 °C, respectively, and composed as reported elsewhere[10]. After 1 h of equilibrium, initial atrial isometric developed tension (IDT) and contractile frequency (CF) were recorded as previously described[11,12]. Forthwith cumulative dose-response curves for PGI_2 were constructed for untreated atrial controls as well as for auricles exposed to propranolol or obtained from chemical sympathectomized animals injected 24 h prior sacrifice with 6-hydroxydopamine (6-OHDA). PGI_2 was kindly provided by Dr John Pike (Upjohn Laboratories, Kalamazoo Michigan, USA). L-Propranolol (for delivery into the tissue bath prior PGI_2 at a final concentration of 10^{-7} M) and 6-OHDA (for i.v. injection with 16.5 mg kg^{-1}) were obtained from standard commercial sources. The effect of PGI_2 on IDT was also tested on atria driven with slightly suprathreshold (+10%) square pulses of 0.5 msec duration and 3.3 Hz of frequency delivered by a conventional stimulator and conveyed to the tissue via 2 platinum electrodes. Experimental records were compared with initial control ones and expressed as percent changes. Differences between mean values were considered significant if $p = 0.05$ or less.

Results. As can be seen in figure 1, A, the IDT of spontaneously beating isolated rat atria exhibited a distinct increasing magnitude following increasing concentrations of PGI_2 within a wide range from of 1×10^{-15} to 1×10^{-9} M. A similar situation, i.e. a significant enhancement, was

observable in atrial contractile frequency (figure 1, B). The maximal effect of each concentration was observed within 90 sec–2 min following the delivery of the drug. The figure also depicts the almost complete blockage of the positive inotropic and chronotropic effects of PGI_2 on preparations exposed to the previous presence of propranolol (10^{-7} M), as well as in those obtained from rats injected with 6-OHDA (16.5 mg kg^{-1}) 1 day before killing. Such a treatment with 6-OHDA has been shown to produce an effective chemical sympathectomy of rat atria[13]. The contractile peak tension of isolated atria driven at a constant frequency of 3.3 Hz was also stimulated by PGI_2 (figure 2).

Discussion. The present report documents that PGI_2 is a powerful stimulating agent of the inotropism and chronotropism of isolated rat atria. However, inasmuch as it was also evident that both actions of PGI_2 are blunted following beta-adrenoceptor blockage as well as after a treatment with 6-OHDA, it is plausible that PGI_2 acts indirectly on rat atrial cells, i.e. via a mechanism involving the release of presynaptic catecholamines and the activation of beta-adrenergic postsynaptic sites. It is not plausible to consider the increase in tension as a secondary effect depending of the enhancement in frequency. Indeed it is known that rat atria presents a negative staircase[10,15,16], and, in addition, preparations paced at a single fixed rate were equaly stimulated as spontaneously beating ones. There are indications that prostacyclin released from the heart increase following noradrenaline[14], but not currently available studies described the opposite situation. Existing reports indicate a direct dilating influence of PGI_2 on the intact coronaries of the cat[7], guinea-pig and rabbit[6]. However, it has also been shown, that there is an increase in coronary vascular tone with very low concentrations of the agent[8]. In addition it is known that PGI_2 abolished platelet aggregation in response to arachidonic acid[2,3] and suppresed the formation of thromboxane A_2 by platelets, which is thought to induce constriction of the coronary vasculature[18,19]. The combination of hemodynamic actions; antagonism on platelet aggregation and the inhibition of thromboxane production are cellular and systemic effects of great importance for avoiding the spread of ischemic damage within myocardium. Prostacyclin release from the heart is increased after anoxia[5], and it has been suggested that the compound may be useful in the prevention of secondary ischemic episodes once the initial ischemic event occurs in the heart[7], mainly because its ability to decrease coronary vascular resistance (i.e., lowering coronary perfusion pressure at a constant coronary flow).

However, existing reports claimed the existence of an augmentation of coronary perfusion pressure following PGI_2, accompanied by a higher heart rate. Our results proved a profound catecholamine-mediated influence of PGI_2 on the IDT as well as on the beating rate of the rat myocardium. If the influence on coronary perfusion and the chronotropic and inotropic effects elicited on the normal rat myocardium occured to a significant degree after human cardiac ischemia, a reduction in myocardial oxygen supply relative to tissue oxygen demand, and a net harmful result, may be expected.

In view of all the present findings, it is difficult to ascertain at the moment whether PGI_2 is a true potentially valuable agent for preserving myocardial tissue in acute ischemia, as it has been claimed to be the case for PGE_1[19,20].

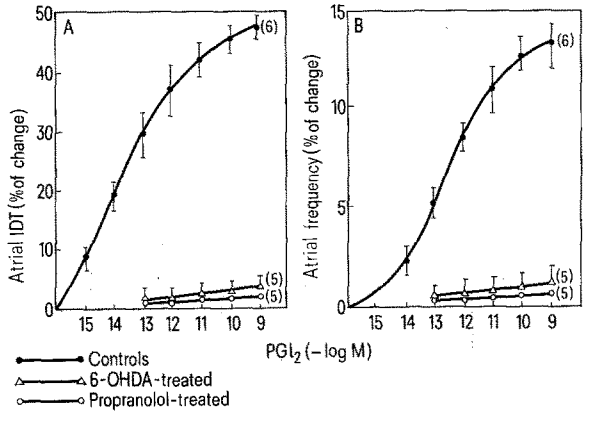

Fig. 1. Cumulative dose-response curves of PGI_2 on spontaneously beating isolated rat atria. IDT = isometric developed tension. Propranolol was used at 10^{-7} M; 6-hydroxydopamine (6-OHDA) was injected i.v. (16.5 mg kg^{-1}) 24 h before sacrifice. Points and bars represents the mean and the SEM, respectively. Figures in the parentheses indicate the number of preparations.

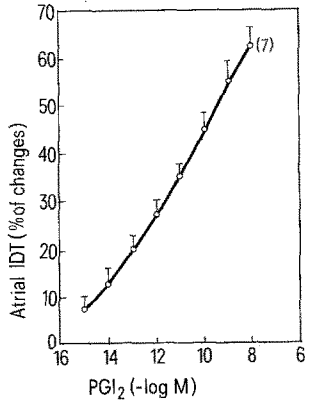

Fig. 2. Cumulative dose-response curve of PGI_2 on driven (3.3 Hz) isolated rat atria. Other conditions and details as in figure 1.

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