Results. Our results indicate that the animal group receiving DMSO alone had no antibacterial activity in their tissues (table).

In the group the received DMSO and gentamicin, tissue gentamicin concentrations were similar to those obtained in the group that received gentamicin only, except for the heart tissue which had increased drug concentrations (table).

Discussion. DMSO is a dipolar hygroscopic solvent. It crosses various biological barriers causing little or no tissue damage[1]. Pharmacokinetic studies in rats demonstrate rapid DMSO absorption and distribution regardless of the route of administration. Serum and tissue peak levels are reached at about 4 h following administration, and decline slowly afterwards. High DMSO concentrations are obtained in the soft tissues, particularly in the spleen, lung, brain, kidney, heart and the liver.

These pharmacokinetic data, together with the ability of DMSO to enhance the penetration of various chemicals through a number of biological membranes such as the bladder[3,8], the skin[3,5] and the blood-brain barrier[7] led us to try and increase the tissue penetration of gentamicin.

Our data indicate that in the rat model, when both DMSO and gentamicin are administered i.p., DMSO fails to enhance the tissue penetration of gentamicin into the brain, lung, liver, spleen, bone and the testicular tissues. A somewhat higher concentration of gentamicin was found in the heart tissue of the animals treated with DMSO and gentamicin. It is doubtful, however, whether this difference is of biological importance. Since the tissue specimens were taken 9 h after the last drug administration it is possible that DMSO facilitated the exit of gentamicin from the tissues. However, the tissues of all the animals still contained the unmistakeable odor of DMSO even after being frozen for several days. Furthermore, previous studies detected appreciable amounts of DMSO 8 h following administration[2].

Gentamicin concentrations in tissues of rats

Organs	Gentamicin (n=6)	DMSO+ gentamicin (n=6)	Statistical significance
	Concentration in μg/ml ± SE		
Brain	0.28±0.07	0.8 ±0.21	$p>0.1$
Heart	0.24±0.09	0.87±0.22	$p<0.01$
Lung	0.66±0.12	0.46±0.12	$p>0.1$
Liver	0.62±0.11	0.27±0.12	$p>0.1$
Spleen	0.30±0.05	0.24±0.09	$p>0.1$
Testes	0.33±0.06	0.1*	
Bone	0.15±0.01	0.13±0.04	$p>0.1$
Muscle	1.1 ±0.01	0.43±0.12	$p>0.1$
Kidney	6.01±2.4	4.13±2.1	$p>0.1$

* Only 3 specimen assayed.

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Increased rate of melanization in hemolymph of American cockroaches *(Periplaneta americana)* and house crickets *(Acheta domesticus)* intoxicated by insecticides

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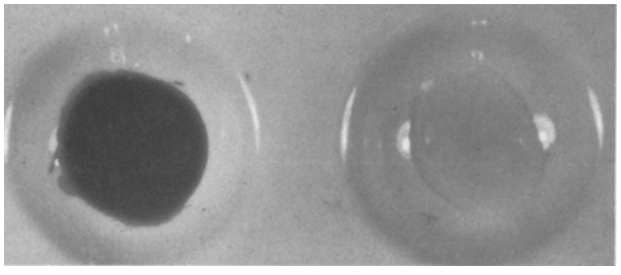
Summary. Treatment of house crickets and American cockroaches with any one of a variety of insecticides increased the rate of melanization in hemolymph incubated with diphenol substrates.

The darkening of insect hemolymph in vitro has commonly been attributed to phenoloxidase (tyrosinase) formation of melanin[1-4]. Hemolymph melanization commonly accompanies pathological conditions[5] and parasitism[4]. Hurst[6] reported increased hemolymph phenoloxidase activity, as measured by in vivo melanization, in *Tenebrio molitor* and *Musca domestica* treated with pyrethrins or DDT. Shorey[7] observed that a topically applied carbamate insecticide (Zectran) caused darkening of cabbage loopers and beet armyworms. We report here the observation that several insecticides (TEPP, dicrotophos, DDVP and DDT) cause an increased rate of melanization in insect hemolymph diphenol solutions.

Each of the following dosages was applied topically to the ventral abdomen of crickets in 2 μl of acetone: TEPP 4 μg, dicrotophos 10 μg, and DDT 50 μg. 30 min after knockdown hemolymph was collected by cutting off the hind legs and collection into 20 μl capillary tubes with gentle squeezing of the abdomen. When sufficient hemolymph was obtained for a test, 2 vol. of Narahashi saline[8] were added. The mixture was sonicated for a period just sufficient to break up the clots and to mix the saline with the hemolymph. The mixture was incubated on spot plates with an equal volume (20 μl) of a 1% solution (w/v) of catechol in Narahashi saline. Control hemolymph from untreated crickets was spotted on the same plate in the same manner. Spot plates were held in closed containers containing damp paper towel to prevent desication. Hemolymph from intoxicated crickets darkened the catechol solution at a much faster rate than control hemolymph. The effect was most obvious at 1-2 h of incubation. Intoxication of American cockroaches with 100 μg of DDT in 2 μl of acetone or with DDVP vapours and collection of their hemolymph by light centrifugation[9] produced similar results when incubated with diphenols (figure).

Iodotyrosine and methyl-para-tyrosine (tyrosine hydroxylase inhibitors) as well as diethyldithiocarbamate and phenylthiourea (phenoloxidase inhibitors) were tested for their ability to inhibit the melanization. The inhibitors of phenoloxidase at a concentration of 10^{-3} M were sufficient to inhibit darkening while the tyrosine hydroxylase inhibitors were ineffective. The diphenols dihydroxyphenylalanine, dopamine, norepinephrine, catechol and epinephrine proved to be effective substrates for the darkening reaction. These results confirm the effect reported by Hurst. The effects of the inhibitors and substrates provide evidence for the involvement of phenoloxidase in the darkening of hemolymph. An increased rate of darkening due to intoxication indicates an increased level of enzyme or enzyme activity. Phenoloxidase has been localized in the hemocytes of various orthopterans by histochemical methods[5,10,11]. Cell lysis and/or increased numbers of cells in the hemolymph of intoxicated individuals may account for the increased darkening. The release of catecholamines from the nervous system of crickets intoxicated by insecticides[12] may raise the redox potential of the hemolymph sufficiently to affect phenoloxidase activity[13]. The possibility that phenoloxidase serves a defensive function and is activated by insecticide produced stress is also under consideration.



Hemolymph from American cockroaches incubated with a 1% DOPA solution in Narahashi saline for 2 h. Left: hemolymph from cockroaches treated with DDVP vapor for 30 min and drawn 1 h later, right: hemolymph from untreated control roaches.

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Orotic acid prevents changes in cardiac sarcolemmal glycoproteins and contractility associated with muscular dystrophy in hamsters[1]

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Summary. Orotic acid included in the diet of cardiomyopathic hamsters during the myolytic phase of the disease (30–60 days of age) prevented the reduction in cardiac sarcolemmal sialic acid, calcium binding, sialyltransferase activity and contractile activity associated with the cardiomyopathy.

The cardiomyopathy associated with Duchenne's muscular dystrophy in man is characterized by focal myolytic lesions and ultimately, development of congestive heart failure. The course of the myopathy in the dystrophic hamster model of this disease follows a similar pattern[2,3] and is associated with a deficiency in the incorporation of sialic acid into the sarcolemmal and interstitial glycoproteins of the myocardium[4]. This defect results in decreased contractility detectable as early as 30 days of age, and moreover is implicated in the pathogenesis of the disease[4]. Mortality is greatly increased by hypokalemia in dystrophic hamsters, however, only 2 potassium salts, orotate and aspartate effectively prevented ventricular hypertrophy and congestive heart failure[5]. I now report that orotic acid rather than potassium prevents the deficiencies in sarcolemmal glycoprotein composition associated with reduced contractility in the dystrophic hamster.

Cardiomyopathic (BIO 14.6; Trenton Experimental Laboratory Animal Co., Bar Harbour, ME) and age-matched normal (BIO.RB) Syrian hamsters, 30 days of age, were segregated into 12 groups of 5 animals each. Each group received ad libitum ground laboratory chow or ground laboratory chow containing either 10% sodium or 10% potassium orotate (Sigma Chemical Corp., St. Louis, MO) for either 30 or 60 days. Neither the intake of food nor body weight differed between groups during the treatment period. Statistical analysis indicated that tissue water, sialic acid content and sialyltransferase activity were not affected by duration of treatment or by the different salts of orotic acid. The results from these groups were pooled to yield comparisons between treated and untreated normal and myopathic animals. The hearts were removed and the ventricles homogenized to produce 1-5 cell fragments. These fragments were subjected to hypoosmotic shock and salt extraction to produce sarcolemmal 'ghosts'[6]. Enzymatic digestion was not used and all media contained a protease inhibitor (aprotinin, 1000 units $\cdot$ l^{-1}). A probability level of 0.05 was preselected as the criterion of statistical significance.

The activity of sialyltransferase in sarcolemma from untreated myopathic hamster hearts was significantly less than that of normal animals (table 1). The activity of this enzyme in sarcolemma from myopathic hearts after treatment with orotic acid did not differ from untreated or treated normal hearts. As reported previously, there was a significant decrease in the sialic acid content of sarcolemma from hearts of dystrophic hamsters[4] (table 1). 10% orotic acid in the diet of the myopathic animals restored the membrane content of sialic acid to levels not different from normal. ATP-independent calcium binding to neuramini-