(6.5%) was digested with commercial trypsin at pH 8.9 in the presence of the rare metal salts and in an untreated control. Even in concentrations as high as 1.1 x 10^{-3} M no effects could be observed.

Experiments with purified lipase and esterase which will be reported in a subsequent communication indicate that these enzymes are not appreciably affected by the salts studied.

It is questionable whether these results obtained *in vitro* conditions represent the effects that would be observed *in vivo*. The results observed by Sure *et al.*[4] are not paralleled completely since we have found that trypsin, lipase and esterase are unaffected or only slightly affected by these metals, one of which has been reported as carcinogenic. A more justifiable comparison must await the results of the effects of these metals on *in vivo* enzymic activity.

8632 C

Sex-Difference in Susceptibility to Dinitrophenol Intoxication in Anesthetized Cats.*

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In the course of an investigation concerning the interaction of the calorigenic actions of 2-4 dinitrophenol and epinephrine, we have observed a marked sex difference in the susceptibility to dinitrophenol intoxication, as judged by the survival time to a dose which was, under our experimental conditions, almost invariably fatal. There do not appear to have been any reports in the literature concerning sex-differences in the action of dinitrophenol.

Adult cats, previously starved for 24 hours, were anesthetized with pentobarbital (37.5 mg. per kilo intraperitoneally), and attached to a closed circuit metabolism apparatus by means of a tracheal cannula. After a preliminary control period of one-half hour, a quantity of sodium 2-4 dinitrophenolate adequate to yield 10 mg. per kilo of dinitrophenol was injected intramuscularly. In certain experiments, an epinephrine solution was infused intravenously at a rate of 0.001 mg. per kilo per minute during the period

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of dinitrophenol action. (The Ringer's solution used contained 1 part of defibrinated blood to 10 parts of saline to protect the epinephrine against oxidation.[1]) In the remaining experiments Ringer's solution was infused at the same rate. Oxygen consumption, rectal temperature, and respiratory rate were followed.

TABLE I.
Survival Time of Cats Receiving Dinitrophenol Alone, and Dinitrophenol Plus Epinephrine.

Males		Females	
DNP alone	DNP plus Epinephrine	DNP alone	DNP plus Epinephrine
283	140	65	16
155	115	140	45
280	107	76	10
245	53	45	60
168	169	25	46
		55	27
		19	
		60	
Av. 226	117	61	34

In the table, the survival times of the animals are given in minutes after the injection of dinitrophenol. Among the cats receiving dinitrophenol alone, the males survived on the average 226 minutes, the females 61 minutes. The reliability of this difference was calculated by the variance method of Fisher.[2] The odds that this difference could have appeared by chance were less than 1 in 5000. Thus, even though the number of cases is small, the reliability of the difference is established beyond reasonable doubt.

Among the cats given epinephrine as well as dinitrophenol, a similar difference appeared, the male survival time averaging 117 minutes, the female 34 minutes. The odds that this difference could have appeared by chance were 8 in 1000, which establishes the difference as reliable.

Within each sex, the addition of epinephrine to the dinitrophenol decreased the survival time by about one-half. The odds that this difference could have occurred by chance were for the males less than 1 in 90. For the females the chances were only 1 in 5 on account of their greater variability.

Lest the differences reported above should be due, not to the sexual factor, but to some factor by chance associated with sex in

1 Wiltshire, *J. Physiol.*, 1931, **72**, 88.

2 Fisher, R. A., Statistical Methods for Research Workers, third edition, London, 1930, p. 107.

this group of animals, we attempted to correlate survival time with body weight, rectal temperature at the time of injection, initial control metabolic rate and dose of pentobarbital required to maintain anesthesia. In none of these cases was any significant correlation found.

However, it was noted that the males showed a greater metabolic rate (per unit surface area) at the moment when the first signs of the fatal collapse (slowing of breathing, decreased oxygen consumption[3]) appeared. Pooling all 4 groups of animals, we find a positive correlation of 0.45 between this "critical" metabolic rate and the logarithm of the survival time, a correlation which might have arisen by chance in this population in 1 case in 20 (Fisher[2]). The existence of this correlation suggests that the sex difference in survival time may be due to the males having a greater capacity to deliver oxygen to the tissues than the females, death resulting when a certain oxygen debt has been incurred. However, the correlation is equally well explained on the assumption that death, occurring as a result of some action of dinitrophenol, relatively early in the course of the rise in metabolism would curtail the time available for the rise and consequently its magnitude.

In seeking a physiological sex difference to which the difference in susceptibility to poisoning with dinitrophenol, a drug which involves particularly actively the metabolism of fats,[3] might be referred, we were reminded of the work of Deuel and associates,[4] who have found that female rats, guinea-pigs and human beings manifest ketosis more readily than do the males. A second case in which shortened survival time after dinitrophenol may well have been accompanied by ketosis occurred in the experiments of Tainter, Boyes and DeEds,[5] who observed that depancreatized dogs succumb more rapidly to the drug than do control operated animals. A third case is constituted by the observations by Baudouin, Bénard, Levin and Sallet,[6] among others, that epinephrine, in doses comparable to those which we found to shorten survival time after dinitrophenol administration, caused a marked ketonuria.

The above considerations have suggested to us the possibility that the metabolic disturbance of which ketosis is the presenting

[3] Hall, Field, Sahyun, Cutting, and Tainter, *Am. J. Physiol.*, 1933, **106**, 432.

[4] Deuel and Gulick, *J. Biol. Chem.*, 1932, **96**, 25; Butts and Deuel, *J. Biol. Chem.*, 1933, **100**, 415.

[5] Tainter, Boyes, and DeEds, *Arch. internat. Pharmacodyn. et Thérap.*, 1933, **45**, 235.

[6] Baudouin, Bénard, Levin, and Sallet, *C. r. S. B.*, 1935, **120**, 860.

manifestation is a factor in predisposing animals to dinitrophenol intoxication. This and other possible explanations of the sex difference reported herein are now being investigated in this laboratory.

8633 P

Comparison of Capacity of Some Nitrated Phenols to Stimulate Respiration of Yeast.*

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We have shown that the action of several of the nitrated phenols on the respiration of yeast may be excitatory or inhibitory, depending on the dosage, and that the undissociated form is the active agent in such stimulation or inhibition.[1, 2, 3] A method for comparing the activities of nitrated phenols as metabolic stimulants has been developed and applied to 3 of the dinitrophenols and to the mononitrophenols.[3] We present herewith the results of experiments designed to compare the activities of 5 nitrated phenols which have been used extensively as metabolic stimulants. These are 2-4 dinitrophenol,[4] 4-6 dinitro-o-cresol,[5] 2-4 dinitro-α-naphthol,[6] 2-4 dinitro-cyclo-pentyl phenol[6] and 2-4 dinitro-cyclo-hexyl phenol.[6] The last 2 compounds were provided through the kindness of Dr. M. L. Tainter, who obtained them from Hoffman, LaRoche and Company. In every case aqueous solutions of the sodium salts were employed.

The pure yeast culture and the experimental methods have been described elsewhere.[2, 3] In every case the yeast, after preliminary washing, was suspended in 1% glucose made up in 0.1M phos-

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1 Field, J., 2nd, Martin, A. W., and Field, S. M., PROC. SOC. EXP. BIOL. AND MED., 1933, **31**, 56.

2 Field, J., 2nd, Martin, A. W., and Field, S. M., *J. Cell. and Comp. Physiol.*, 1934, **4**, 405.

3 Field, J., 2nd, Martin, A. W., and Field, S. M., *J. Pharm. and Exp. Therap.*, 1935, **53**, 314.

4 Magne, H., Mayer, A., and Plantefol, L., *Ann. physiol. physicochim. biol.*, 1931, **7**, 269.

5 Dodds, E. C., and Pope, W. J., *Lancet*, 1933, **2**, 352.

6 Heymans, C., and Casier, H., *Arch. internat. de Pharmacodynamie et de Thér.*, 1935, **50**, 20.