of glycine, participating in the following metabolic pathways:

(tartronate $\xrightarrow{10}$) ketomalonate $\xrightarrow{transaminase}$ aminomalonate $\xrightarrow{decarboxylase}$ glycine

The transaminase activity was also demonstrated in the homogenates of rat liver, rabbit liver and rabbit heart. The highest activity was observed with the rat-liver enzyme.

The enzyme was partially purified from the rat liver homogenate by fractionating with ammonium sulphate. The preparation was about 18 times as active as the starting material, but pyridoxal phosphate had no effect on the activity at this stage of purification.

Full details of this work will be reported in a future publication. Our thanks are due to Miss Aiko Kikuchi for her skilful technical assistance in this work.

Hideo Nagayama
Michiko Muramatsu
Kensuke Shimura

Laboratory of Biochemistry,
Department of Agricultural Chemistry,
Tohoku University,
Sendai. Nov. 6.

[1] Shimura, K., Nagayama, H., and Kikuchi, A., *Nature*, 177, 935 (1956).
[2] Koide, F., Shishido, T., Nagayama, H., and Shimura, K., *J. Agric. Chem. Soc. Japan*, 30, 283 (1956).
[3] Braunstein, A. E., *Enzymologia*, 7, 25 (1939).
[4] Green, D. E., Leloir, L. F., and Nocito, V., *J. Biol. Chem.*, 161, 559 (1945).
[5] Krueger, R., *Helv. Chim. Acta*, 32, 238 (1949).
[6] Cavallini, D., and Frontali, N., *Biochim. Biophys. Acta*, 13, 439 (1954).
[7] Braunstein, A. E., *Adv. Protein Chem.*, 3, 1 (1947).
[8] Campbell, jun., L. L., *J. Bacteriol.*, 71, 81 (1956).
[9] Meister, A., *J. Biol. Chem.*, 206, 587 (1954).
[10] Stafford, H. A., *Plant Physiol.*, 31, 135 (1956).

Insulin - Cortisone Relationship in Experimental Teratogenesis

It is well known that the administration of large amounts of cortisone to experimental animals is associated with the production of hyperglycæmia[1]. There is also evidence from clinical cases that in diabetes the therapeutic use of cortisone increases considerably the daily amount of insulin required to maintain the blood-sugar within normal limits[2]. The nature of this contra-insulin effect of cortisone is uncertain, but it has been suggested that it may be due to excessive destruction of insulin since there is no definite evidence that contra-insulin factors are responsible for a marked increase in the peripheral requirements for insulin[3].

In a series of experiments reported recently we used the teratogenic effects of hypervitaminosis A as a base line for the study of the action of cortisone during pregnancy[4,5]. In these experiments one group of pregnant female rats of the Wistar strain received 60,000 I.U. vitamin A acetate daily by gastric intubation from the eighth to thirteenth days of pregnancy. Another group received the same treatment with, in addition, 20 mgm. of cortisone acetate daily by subcutaneous injection from the ninth to twelfth days. A third group received only the cortisone treatment. It was found that in the young obtained from these rats on the twentieth day of gestation there was an incidence of deformities of the brain and calvaria of 7·8 per cent when the rats had received vitamin A alone during pregnancy, and of 36·6 per cent when they had received both vitamin A and cortisone. There were no deformities in the young from those mothers which received cortisone alone (Table 1). These results were interpreted as suggesting that the cortisone potentiated the teratogenic effects of the hypervitaminosis A, and it appeared that this potentiating action might in some way be related to a disturbance of carbohydrate metabolism.

Table 1

Treatment	No. of dams	Total No. of young	No. of young with brain deformities	Percentage of young with brain deformities
Vitamin A	12	77	6	7·8
Vitamin A, cortisone	12	41	15	36·6
Cortisone	12	73	0	0

We decided therefore, in view of the known contra-insulin effects of cortisone, to carry out further experiments in order to examine the possibility that insulin might exert a contra-cortisone effect in relation to the production of malformations. In these experiments one group of pregnant female rats was given vitamin A for the same period and in the same doses as in the earlier experiments. In addition, they received 1·5 units of protamine zinc insulin by subcutaneous injection, daily from the ninth to twelfth days of pregnancy. Another group received both vitamin A and cortisone in the same quantities as previously administered, but were also given 1·5 units of protamine zinc insulin daily by subcutaneous injection from the ninth to twelfth days. The results of these experiments are shown in Table 2. It will be seen that the incidence of deformities in the young fell from 36·6 to 1·2 per cent when insulin was given in addition to vitamin A and cortisone; and that while there was an incidence of 7·8 per cent in the young of dams which received vitamin A alone, no young with brain deformities were obtained from the dams which received insulin in addition to vitamin A.

Table 2

Treatment	No. of dams	Total No. of young	No. of young with brain deformities	Percentag young with brain deformities
Vitamin A, insulin	12	47	0	0
Vitamin A, cortisone, insulin	12	79	1	1·2

These findings indicate that the potentiating action of cortisone in relation to the production of malformations by hypervitaminosis A is wholly prevented by the administration of insulin. Further, it appears possible that insulin may have some effect in protecting the young against the teratogenic effects of hypervitaminosis A.

We acknowledge with gratitude the assistance of a grant from the Nuffield Foundation.

J. W. Millen
D. H. M. Woollam

Department of Anatomy,
University of Cambridge.
Nov. 12.

[1] Ingle, D. J., *Endocrinol.*, 29, 649 (1941).
[2] Boland, E. W., and Headley, N. E., *J. Amer. Med. Assoc.*, 141, 301 (1949).
[3] Mirsky, I. A., *Recent Prog. Hormone Res.*, 13, 429 (1957).
[4] Millen, J. W., and Woollam, D. H. M., *Brit. Med. J.*, ii, 196 (1957).
[5] Woollam, D. H. M., and Millen, J. W., *Brit. Med. J.*, ii, 197 (1957).

Reversibility of the Inactivation of Rous Sarcoma Extracts by Detergents

THE infectivity of an aqueous extract from a Rous sarcoma is lost if cetyl trimethyl ammonium bromide ('Cetavlon'), a cationic detergent, is added to the active extract[1]. By further addition of an acid colloid substance, like ribonucleic acid[2] or heparin, to the non-infective system infectivity is restored.

From the results obtained by Pirie and co-workers[3-6] on some plant viruses, including the tobacco mosaic virus, treated with anionic detergents, and by Pfankuch and Kausche[7] on tobacco mosaic virus with cationic detergents, the mechanisms of the actions of the two types of detergents on tobacco mosaic virus appear to be different. We have explored the possibility of an analogous behaviour of Rous sarcoma extract towards the two types of detergent, with particular reference to the reversibility of inactivation. The results are given in Tables 1 and 2.

The extract was prepared with a Miller and Golder's buffer[8] 0·14 *M*, *p*H 7·0. The initial concentration of extract was 5 per cent. After centrifuging at 1,500 *g* for 60 min., hyaluronidase, 5 U.V./ml. (Ialovis Vister) was added as solubilizer, and after 30 min. the extract was centrifuged at 15,000 *g*. All operations were carried out at 3° C.

In the experiments with 'Cetavlon' the proportion detergent/infective extract, was 2 mgm./ml., and the amount of acid colloid (heparin) needed to restore infectivity was equal by weight to the amount of 'Cetavlon'. In the experiments with the anionic detergent (sodium lauryl sulphate), the proportions were 0·4 mgm./ml., and the amount of basic colloid (protamine) added was equal to the weight of anionic detergent. Inoculations (standard amount, 2 ml.) were made in the breast muscles of 8-week-old chickens, all from the same breed. They were killed between the thirteenth and fifteenth day of the experiment, at which time the first control animals were beginning to die.

It appears from the results obtained that, whereas the cationic detergent has a reversible action, as is shown by the restoration of infectivity in the inactivated system (extract + 'Cetavlon'), such a reversibility does not occur with the anionic detergent, even after addition of a basic colloid, like protamine.

DINO GUERRITORE

Istituto Regina Elena
per lo studio dei tumori,
Divisione Scientifica,
Viale Regina Elena 291.
Rome.

[1] Guerritore, D., *Z. Krebsfor.*, **61**, 649 (1957).
[2] Guerritore, D., *Z. Krebsfor.*, **61**, 655 (1957).
[3] Bawden, F. C., and Pirie, N. W., *Brit. J. Exp. Path.*, **19**, 66 (1938).
[4] Bawden, F. C., and Pirie, N. W., *Brit. J. Exp. Path.*, **19**, 251 (1938).
[5] Sreenivasaya, M., and Pirie, N. W., *Biochem. J.*, **32**, 1707 (1938).
[6] Bawden, F. C., and Pirie, N. W., *Biochem. J.*, **34**, 1278 (1940).
[7] Pfankuch, E., and Kausche, G. Z., *Biochem. Z.*, **312**, 72 (1942).
[8] Miller, G. L., and Golder, R. H., *Arch. Biochem.*, **29**, 420 (1950).

Effect of Creatine Phosphate Administration on the Cardiac Adenosine Triphosphate of Thyroxinized Rats

THE amounts of adenosine triphosphate, creatine phosphate and inorganic phosphorus in the heart of hyperthyroid animals are below normal. These amounts are restored to about the normal level by administration of adenosine triphosphate, whereas with cocarboxylase and organic phosphates restoration is incomplete[1]. Moreover, thyroxin intoxication reduces the creatine content of the skeletal muscle and the rate of creatine synthesis by liver[2].

We have sought to establish if creatine phosphate, which is essential in oxidative phosphorylation, affects the reduction of this process in the heart, induced *in vivo* by thyroxin in doses of 0·01 mgm. per animal.

0·01 mgm. D,L-thyroxin was daily injected intramuscularly into each of twenty-four rats, which were then divided into four lots. Lot *B* received no further treatment; lot *C* daily received 0·1 mgm. of buffered creatine phosphate per animal injected intramuscularly; lot *D* received daily 5 mgm. of buffered creatine per animal, injected intramuscularly; the rats in lot *E* each received a single injection of both creatine phosphate and creatine at the same doses as above. A fifth lot (*A*) of normal animals was used as control. After fifteen days the animals

Table 1. INACTIVATION AND REACTIVATION OF EXTRACTS FROM ROUS SARCOMA TREATED WITH CATIONIC DETERGENT ('CETAVLON')

Extract + heparin				Extract + 'Cetavlon'				Extract + 'Cetavlon' + heparin			
Inoculations in chickens	Developed tumours	Positive inoculations (per cent)	Mean weight developed tumours (gm.)	Inoculations in chickens	Developed tumours	Positive inoculations (per cent)	Mean weight developed tumours (gm.)	Inoculations in chickens	Developed tumours	Positive inoculations (per cent)	Mean weight developed tumours (gm.)
28	25	89	28	57	20	35	5	30	26	86	16

In the control chickens the ratio, developed tumours/inoculations was 70/76, with a positive inoculation of 92 per cent.

Table 2. INACTIVATION AND REACTIVATION OF EXTRACTS FROM ROUS SARCOMA TREATED WITH ANIONIC DETERGENT (SODIUM LAURYL SULPHATE)

Extract + heparin				Extract + sodium lauryl sulphate				Extract + sodium lauryl sulphate + protamine			
Inoculations in chickens	Developed tumours	Positive inoculations (per cent)	Mean weight developed tumours (gm.)	Inoculations in chickens	Developed tumours	Positive inoculations (per cent)	Mean weight developed tumours (gm.)	Inoculations in chickens	Developed tumours	Positive inoculations (per cent)	Mean weight developed tumours (gm.)
14	12	85	24	28	7	25	2	25	3	12	0·500

In the control chickens the ratio, developed tumours/inoculations was 70/76, with a positive inoculation of 92 per cent.