MUSCLE AND BLOOD HEMOGLOBIN IN THE DOLPHIN

ROBINSON[1] has recently reported the muscle hemoglobin of harbor seals (*Phoca vitulina*) as being 7.715 gm per 100 gm of fresh tissue. We are reporting our results for skeletal muscle and blood from dolphins (*Turciops truncatus*) studied during the past summer through the courtesy of the Marine Studios of St. Augustine, Florida.

Muscle and blood samples were obtained simultaneously from a living female dolphin which weighed 149 kg. Muscle hemoglobin was determined by the method of Whipple[2] and compared colorimetrically with the blood hemoglobin.

The oxygen capacity of the blood was 19.1 volumes per cent., or 14.25 gm hemoglobin per 100 cc. The hemoglobin content of the sacro-spinalis muscle was 3.534 gm per 100 gm tissue. The muscle hemoglobin concentration in the dolphin is thus less than that found in the seal by Robinson, but greater than that in the dog (0.85 to 1.00 gm per 100 gm rectus abdominis muscle).

It should be noted that the common submersion time of the dolphin is rarely over a few minutes; therefore the oxygen storage of the dolphin need not be as great as that of the seal. It will also be necessary to determine the amount of myoglobin in dolphin skeletal muscle in order to determine the oxygen storage more exactly; work on this problem is now in progress.

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BIOGRAPHY OF DR. HARVEY CUSHING

MRS. CUSHING has requested me to prepare a biography of her husband, and I should be most grateful to any one who wishes to make letters, anecdotes or other memorabilia available.

Copies of all letters, no matter how brief, are desired, and if dates are omitted it is hoped that, when possible, these may be supplied (*e.g.*, from the postmark). If original letters or other documents are submitted, they will be copied and returned promptly.

A new Medical Library building is being erected at the Yale University School of Medicine to receive Dr. Cushing's library and collections, including his letters, diaries and manuscripts. Any of his friends who wish, now or later, to present correspondence, photographs or other memorabilia for permanent preservation among the Cushing papers will receive the appreciative thanks of the university.

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SPECIAL ARTICLES

THE ACTION OF CERTAIN HORMONES AS DIETARY CONSTITUENTS

THE present study was prompted by the following theoretical consideration.[1] We know that normal food contains small quantities of hormonic substances, which may exert certain activity and may perhaps be utilized by the organism as precursors of other hormones. On the other hand, a deficient diet may lack certain building stones from which the animal organism synthesizes its own hormones.

Judging from our present knowledge, the various hormones belong mainly to three chemical classes, *viz.*, simple nitrogenous substances, derived mostly from the amino acids of the diet, hormones belonging to the steroid group and finally hormones of protein nature. As far as the first two groups are concerned, it is obvious that the organism prepares these substances from material present in the food. More complicated is the origin of activity of the protein hormones. These substances when hydrolyzed do not seem to differ in structure and composition from the usual proteins; yet no known protein exhibits any definite hormonal activity. Whether the hormonal action is associated with a definite constituent or whether this activity depends on certain spatial configuration of the amino acids, must remain unanswered for the present.

Further, it seems possible that on deficient diets, the synthetic capacity of the organism may become impaired, even though the actual precursors are present. It is hoped that further work will give an answer to the problems outlined.

The present study deals with the action of estrin, testosterone propionate and gonadotropic hormone (of pregnant mare serum)[2] when added to the diet. The problem was approached in this way: a diet was chosen which causes a definite retrogression of the organs to be studied; then a regeneration was sought by inclusion in the diet of the corresponding hormone. It was soon found out, however, that most of the problems raised require special dietary studies: for instance, a diet which is responsible for marked retrogression of the female sex apparatus exerts no influence on the corresponding organs of the male.

In order not to complicate our task, we have used

1 D. Robinson, SCIENCE, 90: 276, 1939.

2 G. H. Whipple, *Am. Jour. Physiol.*, 76: 693, 1926.

1 The present work was outlined by the senior author in an address given on November 10, 1938, at the Hotel Pierre, New York City.

3 These names are listed alphabetically because the work was carried out by the authors as a group. No indication of seniority is implied.

2 We are indebted for the hormones to Roussel Laboratory, Paris, and Dr. André Girard.

TABLE I

EXPERIMENTS WITH ESTRIN*

Diet	Nature of experiment	Number of animals	Duration days	Dose mg	Weight	Loss or gain per cent.	Food	Ovar. mg	Uteri mg
I	Controls	8	26		920	0	1,595	541	897
I	Estrin	8	26	17.6	928	−10.7	1,467	476	*1,797*
I	Controls	8	34		1,151	+ 2.5	2,307	458	971
I	Estrin	8	34	10.27	1,126	− 1.5	2,055	535	*2,047*
I	Estrin plus Gonadotr. H.	8	30	10.27 E 5.2 G. H.	1,097	+ 0.4	2,021	555	*1,674*
II	C.	8	33		1,069	+11.8	2,221	765	1,907
II	Estrin	8	33	6.3	1,049	+11.9	2,115	682	1,864

* The doses of hormones and other data are calculated for 8 rats for the whole period throughout all the tables.

largely a purified diet, to which only those vitamins were added which were available in a pure form. This procedure presented the disadvantage that the diet was incomplete and that the experiments had often to be interrupted before a complete exhaustion of the dietary reserves could be expected. This study was carried out on some 240 piebald rats and the composition of the diets used was as follows:

	Diet I	*Diet II*	*Diet III*
Casein	Non-pur. 200	Non-pur. 180	Same as
Starch	" 500	" 470	Diet I, but
Lard	" 250	" 250	casein and
Salts	40	40	starch
Cod liver oil	20	15	washed
Yeast	—	60	with dil.
Whole wheat	—	200	HCl and
Daily additions:			extracted
B_1	5 γ		with alcohol.
Lactoflavin	10"		
Adenosine	50"		
Nicotinic ac. amide	100"		

From the above results we may conclude that a deficient diet, as used above, produces a marked retrogression of female sex organs; and that this is definitely influenced by an oral administration of estrin. On less deficient diet (Diet II), retrogression does not take place, and estrin *per os* has no effect. Estrin in female rats depresses the food intake and causes loss in weight. Simultaneous administration of estrin and gonadotropic hormone slightly inhibits the estrin action on the uterus. The activity of estrin incorporated into a deficient diet, and its inactivity on a more complete diet supports our view of the possible importance of this substance as a food constituent (as outlined in the introduction).

TABLE II

EXPERIMENTS WITH TESTOSTERONE PROPIONATE

Diet	Nature of experiment	Number of animals	Duration days	Dose mg	Weight	Loss or gain per cent.	Food	Test. g	S.V. g
I	C.	3 × 8 = 24	39		1,001	+ 3.9	2,308	24.8	2.08
I	Testost. ..	3 × 8 = 24	39	17.5	1,002	+ 0.6	2,500	24.0	2.2
II	C.	8	40		1,199	+43.4	3,593	23.9	7.9
II	Testost. ..	8	40	15.45	1,212	+52.7	3,716	24.4	8.5
III	C.	8	28		1,146	−24.1	1,719	22.4	1.0
III	Testost. ..	8	28	16.75	1,147	−14.8	1,820	*26.4*	*1.8*

In discussing the Tables II and III we may conclude that a diet which produces sex organ degeneration in females, leaves the male practically untouched. No changes take place unless the animals lose considerable weight. The action of male and gonadotropic hormones seems to be fairly definite. In contrast to the estrin action in females, the male and gonadotropic hormones exert a rather beneficial action on the food intake and weight, which is perhaps another evidence of their value as food constituents.

Having established that the degeneration of the male sex organs takes place on purified casein and starch,

TABLE III

EXPERIMENTS WITH GONADOTROPIC HORMONE

Diet	Nature of experiment	Number of animals	Duration days	Dose mg	Weight	Loss or gain per cent.	Food	Test. g	S.V. g
I	C.	3 × 8 = 24	39		1,001	+ 3.9	2,308	24.8	2.08
I	G. H.	3 × 8 = 24	39	22.3	1,008	+ 1.2	2,486	23.2	2.34
III	C.	8	28		1,146	−24.1	1,719	22.4	1.0
III	G. H.	8	28	16.75	1,147	−21.5	1,676	*26.2*	*1.4*

we have initiated dietary studies in this direction, keeping in mind, however, that the above effect might be due to loss in weight, rather than to dietary deficiencies.

TABLE IV

INFLUENCE OF DIET ON SEX ORGANS OF THE MALE

Diet	Nature of experiment	Number of animals	Duration days	Weight	Loss or gain in per cent.	Food	Test. g	S.V. g
I	C.	8	39	1,314	+11	2,449	23.7	5.75
I	60 g. yeast per kg food	8	39	1,241	+46	3.359	22.8	8.33
I	200 g. whole wheat added to kg food..	8	39	1,327	+27	3,246	22.6	7.31
I	C.	8	32	1,070	− 6.3	1.977	26.9	1.5
I	Casein extr.*	8	32	1,069	− 5.2	2.297	28.7	1.8
I	C.	8	35	841	+ 9.5	1,844	26.2	1.24
I	Casein extr.†	8	35	834	+ 1.6	1,710	22.6	0.85

* Acetone, acid alcohol and aqueous HCl extract added to imperfectly purified casein.
† Acetone and alcohol extract added to imperfectly purified casein.

From the above table we gather that the nature of dietary additions plays an important role in maintaining the normal aspect of the sex apparatus of the male rat. Additions and extracts containing undefined substances of yeast, whole wheat and aqueous casein extracts exert a favorable action on the food intake and weight, as well as on the picture of the seminal vesicles. Contrary to this, the only experiment with fatty extract of casein was unfavorable in these respects.

Finally, experiments were performed for the purpose of ascertaining the action of synthetic estrogens (dihydro-stilbestrol) which have nothing in common in chemical structure to natural estrogens. Further, we investigated whether testosterone can replace estrin as a hormonic dietary precursor.

Both groups of rats receiving dihydro-stilbestrol in their food showed continuous estrus. The action was practically the same on an incomplete and on a more complete diet. This is in marked contrast to the estrin action, which is only evident on an incomplete diet. As compared with the controls, testosterone initiated in several instances the beginning of an estrus, but it could not be substituted as a dietary estrin precursor. Stilbestrol on a more incomplete diet was somewhat toxic, but less so on a more complete dietary. Testosterone acted adversely in female rats regarding the food intake and therefore weight.

On a deficient diet female sex organs undergo a marked retrogression with cessation of estrus. Inclusion of estrin in such cases shows evidence of definite activity. On the other hand, a more complete diet causes no retrogression and estrin activity is not noticeable. Dihydro-stilbestrol, on the contrary, exerted the same activity on both types of food. Testosterone can not replace estrin as a dietary hormonal precursor.

TABLE V

ACTION OF DIHYDRO-STILBESTROL AND TESTOSTERONE PROPIONATE ON FEMALE RATS

Diet	Nature of experiment	Number of animals	Duration days	Dose mg	Weight	Loss or gain per cent.	Food	Ovar. mg	Uteri mg
I	C.	8	34		779	− 8.9	1,183	294	542
I	Stilbestrol.	8	34	4.7	774	−27.9	906	273	1,779
I	Testost. ..	8	34	9.1	777	−12.1	1,143	315	541
I	Stilbestrol.	8	34	5.3	754	− 5.8	1,319	369	1,716

SUMMARY

The sex organs of the male are more resistant to dietary deficiencies, the animals showing only retrogression along with a significant loss in weight. Testosterone and gonadotropic hormone (of pregnant mare serum) produce a small but definite restorative effect.

Oral administration of estrin to females depresses the food intake, while testosterone, and less so the gonadotropic hormone in males produces the opposite effect. This effect can be regarded as evidence of the dietary significance of these two hormones.

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THE EXCRETION OF CORTIN AFTER SURGICAL OPERATION

A METHOD for the extraction and determination of cortin in human urine has been described.[1] The determination is based on a biological assay for cortin described by Selye and Schenker.[2] Using the method in a study of a group of cases, including one appendectomy, one dilatation and curettage, two gastric resections and one colostomy (none of whom developed post-operative shock), it was found that after operation there is an increased amount of cortin in the urine. Two cases are illustrated in the accompanying table. The excretion of cortin increases to a maximum between the third and fifth post-operative day and then gradually declines to pre-operative levels.

It had previously been shown[1] that cortin appeared in the urine during convalescence from influenza in persons who normally excrete none and that in the presence of chronic suppurative infection cortin is

[1] P. Weil and J. S. L. Browne, Proc. Am. Physiol. Soc., Toronto, 1939, Vol. 121, p. 652.
[2] Hans Selye and V. Schenker, *Proc. Soc. Exp. Biol. and Med.*, 39: 518, 1938.