TABLE I. Effect of Nalorphine on Gastric Emptying Time of 7 Subjects.

Controls		Experimental			
No. of trials	Avg emptying time, hr	No. of trials	Avg emptying time, hr	Diff.	p*
3	2.67	3	6.83	4.16	.03
4	2.44	3	5.17	2.73	<.01
4	2.25	3	2.83	.58	.10
4	2.06	4	2.81	.75	.05
3	2.08	3	6.42	4.34	.02
3	2.00	2	5.25	3.25	.09
3	3.08	3	6.00	2.92	.01
Avg	2.37		5.04	2.67	<.01

* p according to the Fisher t test.

cept No. 3 and 6, showed a statistically significant delay, and in No. 6 lack of significance is possibly due to the small number of trials since he showed a large difference. The average delay for the series as a whole is significant at the one per cent level.

Discussion. Initially, a dose of 10 mg was tried on 4 of the subjects, but this produced severe malaise, and since at the end of 4 hours practically none of the meal had left the stomach, the experiment was discontinued. The dosage was reduced to 5 mg for all subsequent work, and the results reported here are based entirely on this dose.

According to a recent textbook of pharmacology(5), the effects of nalorphine in man, when morphine or other narcotic has not been given previously, are similar to those of morphine but less intense. The delay in gastric emptying reported here is at least as great as that produced by morphine, and possibly greater. There is some disagreement among various workers about the magnitude of the morphine effect on the stomach(6).

Summary. Five mg of nalorphine administered subcutaneously caused an average delay of over 2½ hours in the emptying time of the stomach of 7 human subjects.

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Effect of Growth Hormone and Testosterone on Induction of Cardiovascular Changes in Choline-Deficient Rats.* (22351)

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Severe choline deficiency may induce fatty and necrotic changes in cardiac muscle fibres, medial coronary lipoidosis and medial sclerosis of the Moenckeberg type in aortas of rats fed a high fat hypolipotropic diet(1-3). It is not known whether those changes are primarily due to choline deficiency or are secondary phenomena associated with the haemorrhagic renal cortical necrosis (HKL). Evidence in favor of the latter explanation is accumulating.

We have found that growth hormone and testosterone administered either alone or together to choline-deficient rats greatly enhances occurrence of HKL in animals which are in a borderline state of choline deficiency. Cardiovascular disease is increased in hormone-treated animals.

Methods. In a pilot experiment using 80 animals, Hall and Bieri's findings(4) were confirmed and borderline conditions for our final experiment were delineated. This re-

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TABLE I. Composition of Diet WCA.

Casein	7
Peanut meal*	28
Soya protein†	5
Salt mixture‡	3
Sucrose vit. mixture‡	1
Sucrose	11
Starch	10
Lard	34
α-tocopherol acetate§	.015
Cod liver oil‖	.01
Corn oil§	1

* Extracted with 50%, 75% and 95% ethanol.
† Water washed "alpha proteins" (Glidden Co.).
‡ For details see: *Canad. J. Med. Sci.*, 1953, v31, 135.
§ Fat-soluble vitamins dissolved in corn oil so that 1% of dietary corn oil will supply the desired amount of fat-soluble vitamins.
‖ Obtained from Ayerst McKenna and Harrison, Montreal; contains 200,000 i.u. vit. A and 50,000 i.u. vit. D/g.

port deals only with this final experiment in which 44 female rats were used. Using our high-fat severely choline-deficient diet, it was found that weight range between 110 and 115 g in female rats was just suitable, at a room temperature of 72 degrees, for production of the desired borderline conditions in control animals. All animals were put on the same choline-deficient diet, the composition of which is shown in Table I. One experimental group was injected with 3 mg of growth hormone subcutaneously once daily; another with 1 mg testosterone (CIBA) subcutaneously once daily, while a third group was injected with both hormones. Choline-deficient animals injected with saline served as controls. The growth hormone used was obtained from the Connaught Laboratories in Toronto, lot 200, 1955 and was prepared by the method described by Campbell and Davidson(5). It contains less than 0.05 US unit TSH, less than 0.005 US unit ACTH and less than 0.2 US unit Prolactin.

The animals were injected for a maximum of 14 days. The survivors in the experimental groups were then kept another 3 weeks on a choline-deficient diet, to detect any late effect of the treatment. They were then sacrificed at the same time as the saline-injected controls.

Results. Table II illustrates the effect of growth hormone and testosterone administered either alone or together to young female choline-deficient rats. The saline-injected females all had fatty livers but the incidence of kidney and heart lesions was very low. There were no deaths from bilateral renal cortical necrosis in the control group. All control animals which showed slight involvement of the kidney due to lack of choline ("renal frosting," Hartroft(6)) were able to sustain a critical period of kidney damage between the 10th and 14th experimental day. Fatty infiltration of cardiac muscle fibres and focal cardiac necrosis were present in mild degree. The aortas of the saline-injected controls did not exhibit any lesions.

All the animals given growth hormone, however, showed a marked degree of kidney and cardiovascular involvement. The livers were fatty. More than half died from acute bilateral haemorrhagic renal cortical necrosis. The survivors in the growth hormone group had advanced degrees of 'renal frosting' as is shown in Table II. The rate and severity of cardiovascular involvement increased with the advent of HKL. At least half the aortas showed the Moenckeberg-type lesion described elsewhere(1).

The findings in the testosterone-injected animals were similar but slightly less advanced. The animals injected with both growth hormone and testosterone exhibited more severe lesions but not more frequently than the animals given growth hormone

TABLE II. Effect of Growth Hormone and Testosterone on Cardiovascular Changes in Choline Deficiency.

Group	Avg wt, g	Avg food intake, g/day	No. of rats	Pathology: Kidney: Frosting	Kidney: HKL	Kidney: Total	C. V. system: Cardiac	C. V. system: Aortic	C. V. system: Total
Control	112	14.5	18	6	0	6	3	0	3
Growth hormone	112	14.3	7	3	4	7	4	3	7
Testosterone	110	11	9	3	6	9	6	3	6
Growth hormone + testosterone	117	12.6	10	3	7	10	5	4	5

alone. The number of animals used does not permit an accurate assessment of the effects of the different hormones. We were primarily interested in elucidating the aetiology of cardiovascular disease in choline deficiency and less so in the ability of different hormones to aggravate it.

The average daily food consumption was calculated for the first week of the experiment only. Thereafter, the experimental animals exhibited loss of weight, reduction and irregularity of food intake. The hormone-injected animals perhaps showed a slight decrease in their food intake in comparison with the saline-injected controls. (Table II).

Discussion. The observation of Hall and Bieri(4), on the aggravating effect of growth hormone on choline deficiency has been confirmed and extended. Under suitable conditions of borderline choline deficiency, growth hormone may force animals into the full-blown picture of severe bilateral renal cortical necrosis (HKL). With the development of HKL, cardiovascular changes make their appearance in increased frequency and severity. The results of our present growth hormone experiment provide further evidence(11) that the appearance of cardiac lipoidosis and necrosis, coronary medial lipoidosis and the Moenckeberg-type of arteriosclerosis in choline deficiency are probably closely associated with HKL. It is probable that growth hormone in our experiments stimulates protein synthesis(7). This would augment the methionine requirement, which in turn would increase the severity of choline deficiency, as less methionine would be available to act as a precursor of choline. The increased severity of choline deficiency may then aggravate the bilateral renal cortical necrosis and this lesion may play its part in the cardiovascular changes. This concept is supported by the fact that results similar to those obtained with growth hormone are secured by testosterone administration. Testosterone may be a powerful protein anabolic hormone(8) in the presence of insulin(9).

Growth hormone did not increase the food intake in our choline-deficient animals. It increases appetite when administered to normal animals, but this effect was not demonstrable in choline deficiency.

Selye(10) was able to produce cardiovascular changes (collagen disease like) by growth hormone and/or DCA injection in rats which were "sensitized" to these two hormones. The nature of the lesions produced does not resemble closely those which we have found. (Moenckeberg type).

Summary. 1. The work of Hall and Bieri on aggravation of choline deficiency by growth hormone has been confirmed. 2. Under suitable borderline conditions, growth hormone precipitates haemorrhagic lesions of the kidney in choline-deficient animals. With the advent of kidney lesions, cardiovascular changes appear. 3. These findings provide further evidence that the aetiology of cardiovascular lesions in choline deficiency is closely associated with kidney damage, but a causal relationship of one lesion to the other can not be established by experiments of this type. 4. Testosterone has effects similar to those of growth hormone in choline deficiency. 5. The mechanism of this aggravation of choline deficiency has been briefly discussed.

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