Br. J. exp. Path. (1980) **61**, 195

PATHOPHYSIOLOGICAL DIFFERENCES BETWEEN OBESE AND NON-OBESE SPONTANEOUSLY HYPERTENSIVE RATS

B. C. WEXLER, S. G. IAMS AND J. P. McMURTRY

From the May Institute for Medical Research of the Jewish Hospital and the Departments of Medicine, Pathology, and Physiology, University of Cincinnati College of Medicine, Cincinnati, Ohio 45229, *U.S.A.*

Received for publication December 7, 1979

Summary.—A genetic variant of the spontaneously hypertensive rat (SHR) has been produced which becomes markedly obese as well as hypertensive, i.e. Obese/SHR weigh 800 g as against 300 g for non-obese cohorts. Serum enzymes (CPK, SGOT, SGPT and LDH) are frequently abnormally elevated, concomitantly with a high incidence of myocardial necrosis. Obese/SHR are hyperlipidaemic with severe fatty infiltration of the liver; they are hyperglycaemic with enormous islets of Langerhans and extensive beta-cell degranulation; despite elevated blood urea nitrogen (BUN) levels, they manifest little or no renal damage. Measurement of corticosterone, deoxycorticosterone (DOC) and aldosterone in Obese/SHR demonstrate marked hyper-responsiveness to moderate stress. Circulating prolactin levels are lower in Obese and non-obese/SHR compared to SHR, but Obese/SHR manifest unusually high increases in circulating prolactin levels in response to stress. Obese/SHR are hyperinsulinaemic and have subnormal growth-hormone levels. Despite mild hypertension, hyperglycaemia and hyperlipidaemia, Obese/SHR show no evidence of atheromatous change but do develop early polyarteritis nodosa. It is believed that the genetically programmed hypertension and hyperglycaemia is mediated by increased DOC, aldosterone and corticosterone production respectively, and that the obesity, hypertension, and diabetes in Obese/SHR may be likened to human Cushing's disease.

THE SPONTANEOUSLY HYPERTENSIVE rat (SHR) is under intensive investigation. This experimental model is intriguing because these animals are normotensive when young and develop hypertension spontaneously as they mature (Okamoto and Aoki, 1963). Apart from the genetic input, the aetiology of their hypertensive disease remains an enigma. In 1973, Koletsky described a genetic variant of the original SHR rat which developed spontaneous obesity in addition to hypertension, accompanied by hyperlipaemia, premature atherosclerosis and death (Koletsky, 1973, 1975). Through the kind auspices of N.I.H., our laboratory has been able to maintain a colony of Obese and non-obese/SHR. This report describes the pathophysiological differences between hypertensive Obese/SHR, non-obese SHR and SHR, as well as differences between our sub-strain of Obese/SHR (N.I.H.-derived) and the Obese/SH rats of Koletsky (1973, 1975).

METHODS

The original breeder stock from which our colony of Obese/SHR were derived are called "Corpulent-SHR" and were kindly provided by Dr Carl T. Hansen, Animal Genetics Division, N.I.H. From birth to weaning it is not possible to determine which of the progeny of Obese/SHR will become obese. At 5 weeks of age, about 25% of the young become hyperphagic, gain weight rapidly, and promptly develop

Reprint requests to Dr Bernard C. Wexler, May Institute for Medical Research, 421 Ridgeway Avenue, Cincinnati, Ohio 45229, U.S.A.

rounded body contour. Both male and female Obese/SH rats gain weight at the rate of 50–60 g per week. By 6–7 months their average body weight is 700–800 g compared to 300 g and 235 g for non-obese/SHR males and females respectively. The non-obese/SH controls cited in this report were the litter-mates of the Obese/SHR. In order to compare strains of rat, male and female normotensive Wistar: Kyoto rats (from which SHR were originally derived), SHR, and non-obese and Obese/SHR of various ages were used. (The Obese/SHR sub-strain was derived from the mating of a normotensive male Sprague–Dawley rat and a hypertensive female SH rat.) All of the animals were housed in our air-conditioned, humidity- and light-controlled Animal Research Colony. The animals were fed a commercial rat chow (Purina) which is relatively low in fat (4%) and were given tap water to drink *ad libitum*. The Obese/SHR consumed 2–3 times the amount of food eaten by their non-obese litter mates.

Blood samples were collected between 0800–1100 hours in deference to the diurnal rhythm of adrenal steroids. In order to determine hormonal levels under minimal conditions of stress, blood was withdrawn by cardiac puncture 1 min after removing the animal from its cage. None of the animals struggled. Several days later, the endocrine response to moderate stress in the same animals was evaluated by obtaining blood from the aorta under light Seconal anaesthesia. Blood pressure (systolic) was recorded using the indirect tail-cuff procedure just before exsanguination. Blood samples were centrifuged (refrigerated) and assayed for creatine phosphokinase (CPK), glutamic oxaloacetic and glutamic pyruvic transaminase (SGOT and SGPT), lactic dehydrogenase (LDH), glucose, free fatty acids, triglycerides, cholesterol, and blood urea nitrogen (BUN), using the automated techniques prescribed for the Auto-Analyzer (Technicon Instruments). Corticosterone, deoxycorticosterone (DOC), and aldosterone were measured by a radioimmunoassay method of Iams and Wexler (1976). Circulating prolactin levels were measured by a double-antibody radioimmunoassay system using materials and methods kindly provided by A. Parlow and the NIAMDD Hormone Distribution Program, N.I.H. All results are expressed in terms of the NIAMDD Rat Prolactin RP-1. Plasma samples were assayed in duplicate at 2 dilutions for insulin and growth hormone. Growth hormone concentrations were determined using a double-antibody radioimmunoassay kit and protocol supplied by NIAMDD (Dr A. Parlow). A second antibody (goat anti-rhesus monkey globulin) was purchased from Research Products International Corp. Growth-hormone values are expressed in terms of NIAMDD-Rat-RP-1 standard. Plasma insulin was determined by a double-antibody procedure described by Morishige *et al.* (1977) except that the insulin standard (porcine) was purchased from Novo Research (Denmark).

The hearts and aortae of each animal were carefully examined at necropsy for gross evidence of vascular disease. Pertinent organs from each rat were trimmed and weighed. The hearts were dropped into saline and allowed to pump out residual blood, blotted, and weighed, *i.e.* as a gravimetric index of cardiac hypertrophy. Organs and tissues were embedded in paraffin and sectioned at 3 μm. Frozen sections used for lipid studies were cut at 5–10 μm. Adjacent sections were stained with haematoxylin–eosin for routine analysis; Verhoef van Gieson and Gomori's aldehyde fuchsin stains for elastic tissue; Alcian blue and toluidine blue for metachromasia; the Hale stain for glycosaminoglycans; the von Kossa method for calcium. Sudan II and III, oil red O, and Sudan black B were used to demonstrate lipids in both frozen and paraffin sections. Statistical analysis of results was performed using a one-way analysis of variance, chi-square test, or Student's t test.

RESULTS

General observations

At birth until weaning, it is not possible to distinguish non-obese from Obese siblings. At 5 weeks of age, some of the animals become hyperphagic, there is rapid development of obesity, and the animals become sedentary. The Obese/SHR become exceptionally rotund with the head and paws barely discernible (Fig. 1). The ubiquitous, grossly visible, adiposity involving the s.c. tissues, mesentery, liver, and carcass makes dissection at necropsy very difficult. Obese/SHR do not survive beyond 12 months of age; the few exceptions are those whose hyperphagia becomes abated and adiposity becomes reduced.

Blood pressure

The systolic blood pressure of SHR weanlings is significantly ($P < 0{\cdot}001$) elevated above normotensive WKY matched controls (Fig. 2). At 5 weeks of age, the spontaneous hypertension of SHR begins to rise briskly and by 4 months of age reaches a zenith of 200 ± 5

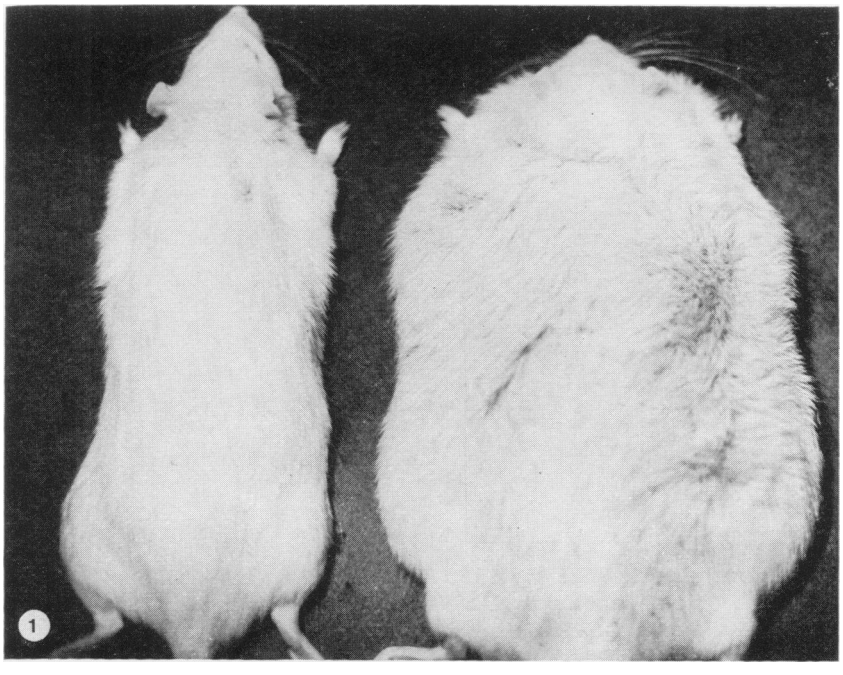


FIG. 1.—Photo of a male, obese/SH rat compared with its non-obese SHR littermate. At 6 months of age, the obese/SH rat weighed 810 g as against 325 g for the non-obese/SH rat.

mmHg with a gradual increase thereafter. The non-obese/SHR do not manifest as brisk an increase in blood pressure as SHR (Fig. 2) but eventually do attain severely elevated blood pressure (200 ± 5 mmHg) comparable to SHR. By direct contrast, Obese/SHR develop abnormally elevated blood pressure (160 ± 5 mmHg) but significantly ($P < 0{\cdot}001$) below their non-obese cohorts (Fig. 2).

Gravimetric data

The male and female Obese/SHR become 2–3 times heavier than their non-obese brothers and sisters (Fig. 3). Obese/SHR eat, drink, and urinate 3 times more than their non-obese litter mates, which continues unabated until the animals become 12 months old. Despite outward appearances of being lean, the non-obese SHR became much heavier than SHR (Fig. 3).

The pituitary glands of SHR gradually increase in size and weight with age, but are smaller and weigh less than those of normotensive WKY (Fig. 4). The pituitary glands of non-obese/SHR are much heavier than those of their obese counterparts ($P < 0{\cdot}001$; Fig. 4). The pituitary glands of female rats are considerably heavier than those of males, as is also the case with adrenal glands. These relationships obtain in terms of absolute glandular weight and become even more accentuated when expressed on the basis of organ-wt: body-wt ratio.

The adrenal glands of Obese/SHR are significantly heavier than their non-obese confrères (Table I). Although an enlarged adrenal gland is usually associated with an involuted thymus gland, a striking enlargement of the thymus glands of male and female obese rats was observed (Table I). Despite the corpulence and larger body surface area of the obese rats, the heart weights of the severely hypertensive SHR weighed much more than

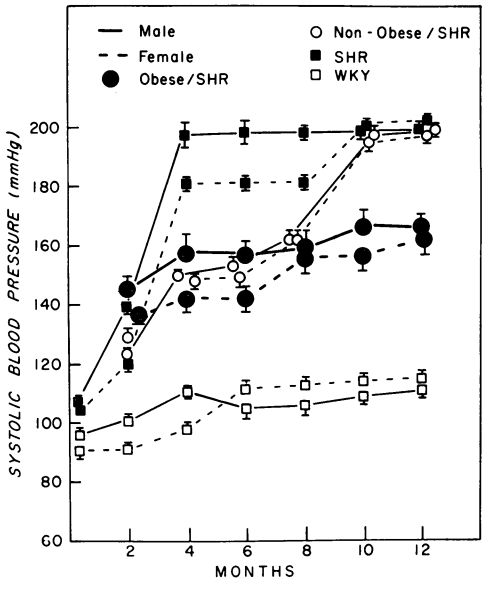


FIG. 2.—Changes in systolic blood pressure levels of male and female, normotensive Wistar: Kyoto rats (WKY) and hypertensive SHR, non-obese SHR, and obese/SHR from weaning until 12 months of age. Each point depicted is the mean ± s.e. mean, (n) = 24 or number of samples for each determination. The same protocol applies to Figs 3 and 4.

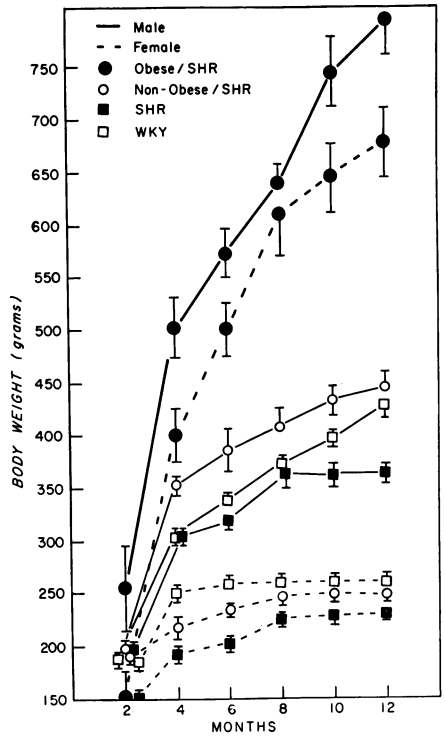


FIG. 3.—Changes in body weight.

those of their obese cousins (Table I). Although cardiac and renal hypertrophy are a good index of elevated blood pressure, the kidneys of the severely hypertensive SHR were considerably smaller than either the non-obese or Obese/SHR (Table I). The testes and ovaries were reduced in size in SHR and Obese/SHR (Table I). A few Obese/SHR manifested arrested growth, *i.e.* decreased crown-rump length, but they did become exceptionally rotund.

Biochemistry

Enzymes.—In most cases, the serum enzymes indicative of myocardial or hepatic damage, *i.e.* CPK, SGOT, SGPT and LDH, were considerably elevated in all of the sub-strains of SHR (Table II) with the exception of SGOT. The serum enzyme levels of Obese/SHR tended to be most abnormally elevated (Table II).

Lipids.—All the sub-strains of SHR were hyperlipidaemic (Table II). The Obese/SHR manifested relatively moderate hypertriglyceridaemia and hypercholesterolaemia compared to their non-obese cohorts, and the hypertensive SHR manifested an equivalent degree of hyperlipidaemia (Table II).

Glucose.—All of the SHR were hyperglycaemic, *i.e.* the average blood sugar of normotensive rats is 120 ± 5 mg/100 ml. The Obese/SHR had the highest glucose levels (Table II).

Blood urea nitrogen.—The Obese and non-obese/SHR exhibited elevated BUN levels (Table II).

Hormones: corticosterone, deoxycorticosterone, aldosterone, prolactin, growth hormone and insulin

SHR are hyper-responsive to stressful stimuli, females being most responsive, as is characteristic of most rodents. Obese/

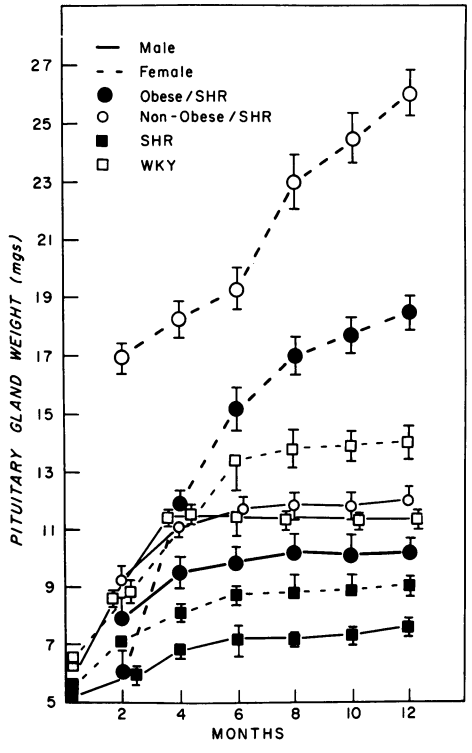


FIG. 4.—Changes in pituitary-gland weight.

SHR have significantly ($P < 0{\cdot}001$) higher resting corticosterone levels than any of the other SHR sub-strains and concomitantly manifest the most brisk increase in corticosterone secretion in response to a mild stress (Fig. 5).

Despite their comparatively lower blood pressure, the Obese and non-obese/SHR had significantly higher ($P < 0{\cdot}001$) resting levels of DOC than their severely hypertensive SHR cohorts. Female non-obese and Obese/SHR manifested a considerable increase in DOC secretion in response to mild stress (Fig. 5). Blood aldosterone levels are characteristically higher in female than in male rodents, as observed in this experiment (Fig. 5). Again, the females of the several sub-strains of SHR produced much more aldosterone than males in response to stress. The Obese/SHR produced the greatest amount of DOC and aldosterone (and corticosterone) in response to stress (Fig. 5).

It is well established that blood prolactin levels, like adrenal steroids, are greatly increased under conditions of acute stress. Although the obese and non-obese/SHR had significantly ($P < 0{\cdot}001$) lower blood prolactin levels (under quiescent conditions) compared to hypertensive SHR, the Obese/SHR displayed the highest levels of prolactin response to stress (Table III). Although SHR and non-obese/SHR females secreted 2 and 3 times more prolactin than their male counterparts, the prolactin levels of male and female Obese/SHR (under resting conditions) were the same (Table III).

At 6 months of age, when hyperphagia and obesity are at a zenith, circulating growth hormone levels in Obese/SHR were subnormal compared to those of their non-obese/SHR littermates (Table

TABLE I.—*Differences in organ weights of one year old, obese, non-obese, and spontaneously hypertensive rats (SHR)*

	Sub-strain of rat	Adrenal	Thymus	Heart (mg)	Kidney	Testes/ovary
Males	Obese/SHR	28 ± 2	740 ± 64	1652 ± 49	1477 ± 36	1524 ± 48
	Non-obese/SHR	23 ± 1†	215 ± 21*	1555 ± 69	1386 ± 47	1744 ± 36*
	SHR	16 ± 1*	145 ± 10*	1705 ± 15*	1152 ± 23*	1449 ± 19*
Females	Obese/SHR	42 ± 2	671 ± 26	1432 ± 80	1363 ± 82	37 ± 5
	Non-obese/SHR	34 ± 2*	283 ± 9*	1130 ± 41*	828 ± 10*	48 ± 3†
	SHR	22 ± 1*	141 ± 5*	1665 ± 62*	674 ± 29*	28 ± 5*

Values are means ± s.e. mean; (n = 24) or number of samples for each determination. Statistical comparisons are made between Obese/SHR *vs* non-obese/SHR *vs* SHR.

* $P < 0{\cdot}001$.

† $P < 0{\cdot}05$.

TABLE II.—*Comparison of serum enzymes, lipids, glucose, and BUN of 1-year-old, obese, non-obese, and spontaneously hypertensive rats (SHR)*

Sub-strain of rat	CPK μM/l	SGOT μM/l	SGPT μM/l	LDH μM/l	Trigly. mg%	F.F.A. mEq/l	Chol. mg%	Glucose mg%	BUN mg%
Obese/SHR	479±20	93±19	140±7	107±16	107±16	386±70	142±10	205±6	30±1
Non-obese/SHR	561±22*	90±9	90±9*	91±20	91±20	356±44	116±25	178±4*	25±1*
SHR	298±40*	80±9	42±4*	50±8	50±3*	400±26	138±7	124±30*	19±1†
Obese/SHR	198±18	78±2	87±4	210±27	251±13	437±10	137±14	186±17	32±1
Non-obese/SHR	203±23	90±10	56±6*	123±23*	77±11*	427±19	83±1*	144±10*	25±1*
SHR	330±5*	72±4	38±3†	418±2*	130±2*	352±12	138±6*	146±3	16±1*

Values are means ±s.e. mean; (n=24) or number of samples for each determination.
Statistical comparisons are made between obese/SHR *vs* non-obese *vs* SHR.
* $P < 0.001$.
† $P < 0.05$.

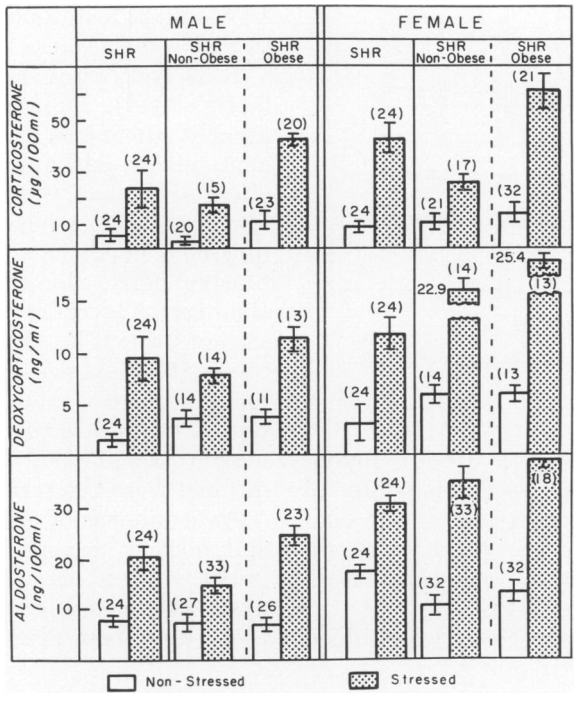


FIG. 5.—Comparison of circulating levels of corticosterone, deoxycorticosterone and aldosterone, the main glucocorticoid and mineralocorticoid secreted by the rat adrenal cortex in male and female, hypertensive, obese/SHR, non-obese/SHR and SHR. The height of each column indicates the mean ±s.e. mean; (n)=number of samples.

IV). Similarly, there is marked hyperinsulinaemia, particularly in Obese/SHR ($P < 0.001$), compared to non-obese/SHR controls (Table IV).

Gross and microscopic pathology

Concomitantly with the copious and ubiquitous deposition of fat in obese rats, their livers are severely infiltrated with fat (Fig. 6). Despite their outward appearance of being lean, the non-obese/SHR and SHR also displayed fatty livers. All of the SHR manifested old and new foci of myocardial necrosis or fibrosis (*vide infra*) but no grossly visible evidence of arteriosclerosis.

Microscopically, the hearts of Obese/

TABLE III.—*Comparison of blood prolactin levels (ng/ml) of 1-year-old, obese, non-obese, and spontaneously hypertensive rats (SHR) under minimal and moderately stressful conditions*

	Obese/SHR		Non-obese/SHR		SHR	
Stress	Minimal	Moderate	Minimal	Moderate	Minimal	Moderate
Male	11 ± 2 (4)	262 ± 11* (6)	9 ± 2 (4)	56 ± 9* (5)	25 ± 8 (11)	124 ± 13* (12)
Female	12 ± 6 (3)	187 ± 13* (6)	26 ± 6 (6)	66 ± 8* (6)	50 ± 12 (11)	173 ± 15* (8)

Values are means ± s.e. mean; (n) = number of samples.
Statistical comparisons are made between animals necropsied under minimal as against moderate conditions of stress.
* $P < 0{\cdot}001$.

TABLE IV.—*Comparison of growth hormone and insulin levels of 6-month-old obese and non-obese, spontaneously hypertensive rats (SHR) when hyperphagia and obesity are intense in obese/SHR*

	Growth hormone (ng/ml)		Insulin (ng/ml)	
	Obese/SHR	Non-obese/SHR	Obese/SHR	Non-obese/SHR
Male	9 ± 1 (10)	28 ± 3 (9)*	87 ± 25 (10)	35 ± 9 (9)*
Female	10 ± 3 (12)	20 ± 4 (13)	44 ± 16 (12)	38 ± 6 (13)

Values are means ± s.e. mean; (n) = number of samples.
Statistical comparisons are made between obese and non-obese animals.
* $P < 0{\cdot}001$.

SHR were hypertrophied with thickened but patent coronary arteries, and showed foci of endocardial fibrosis and myocardial necrosis (Fig. 7). The aortae of these obese and non-obese/SHR were free of arterial disease. However, 2 of the male Obese/SHR had early polyarteritis nodosa (PAN) involving intestinal, mesenteric, and pancreatic arteries (Fig. 8). These PAN lesions consisted of hyalin, fibrous, and glycosaminoglycan material; despite the obesity, fatty liver, and hyperlipidaemia, there was no lipid deposition in the arterial walls of these rats. The pituitary glands manifested intense basophilia, the adrenal glands showed extensive lipid vacuolization and hyperplasia of the zona fasciculata, and extensive lipid depletion of the zona glomerulosa in both obese and non-obese/SHR. The Obese/SHR exhibited giant-sized islets of Langerhans (Fig. 9) and extensive beta-cell degranulation indicative of active insulin production and release.

DISCUSSION

There are several experimental models of obesity produced by hypothalamic lesions, endocrine, nutritional, and drug manipulations, as well as genetic alterations (Bray and York, 1979). Although all of these experimental models share many pathological attributes. the Obese/SHR is unique in that it develops high blood pressure and vascular disease spontaneously. Only 2 spontaneous mutations have been discovered in rats that cause obesity, one described by Zucker and Zucker (1961) and one described by Koletsky (1973). The gene which controls the expression of hyperphagia and obesity is inherited in a recessive manner (Yen, Shaw and Yu, 1977).

The metabolic and pathological changes observed in our Obese/SHR and in those described by Koletsky (1973, 1975) bear a striking resemblance to the pathophysiological changes which we have found in

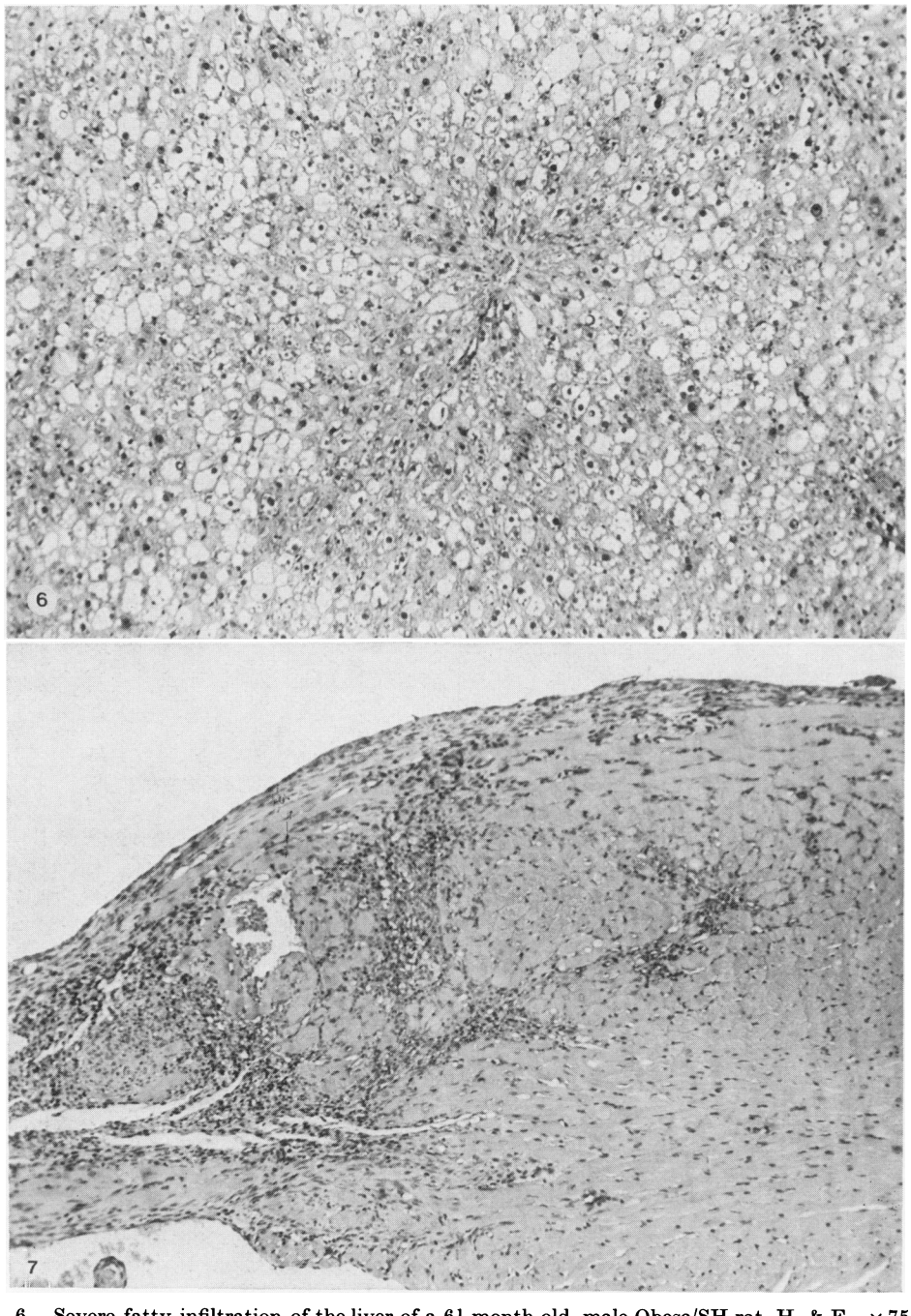
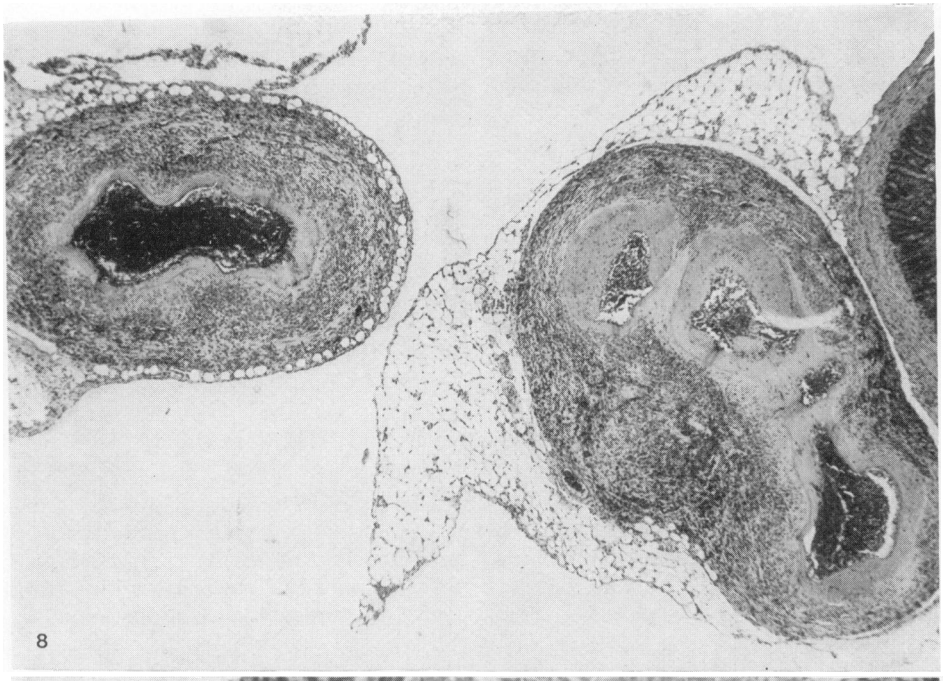


FIG. 6.—Severe fatty infiltration of the liver of a 6½ month old, male Obese/SH rat. H. & E., × 75.

FIG. 7.—Left ventricle of an Obese/SH rat illustrating the patchy necrosis, fibrosis, and white cell infiltration frequently encountered in this obese, genetic variant of the SHR strain. H. & E., × 57.



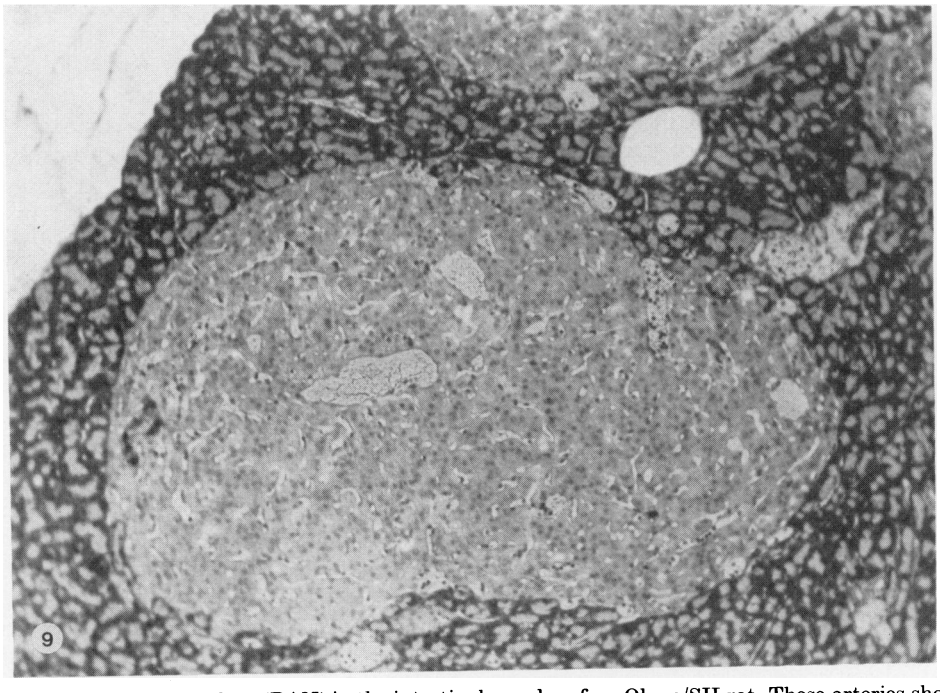


FIG. 8.—Polyarteritis nodosa (PAN) in the intestinal arcades of an Obese/SH rat. These arteries show intimal proliferation with fibrosis, hyalinization, and arteritis. The PAN lesions stain very positively for glycosaminoglycans. H. & E., × 75.

FIG. 9.—Giant-sized islet of Langerhans in an Obese/SH rat. Another giant-sized islet may be seen at the upper edge of the photo. These giant-sized islets manifest complete degranulation of their insulin-producing beta cells. H. & E., × 93.

spawning salmon (Robertson and Wexler, 1957, 1962; Robertson, Wexler and Miller, 1961) and in repeatedly bred male and female rats, *i.e.* obesity, fatty liver, hyperlipidaemia, hyperglycaemia, islet hyperplasia, adrenocortical alterations, renal pathology and lithiasis, arterial degenerative changes, premature aging, and death (Wexler, 1964, 1970). It is our contention that in spawning salmon and in repeatedly bred rats, chronic stimulation of the hypothalamic–pituitary–adrenal–gonadal axis, under the *aegis* of the reproductive effort, will eventually lead to abnormal function of this axis followed by the spectrum of abnormal metabolic and pathological sequelae described by Wexler and Kittinger (1965). For example, hyperadrenocorticism leads to hyperlipidaemia and adiposity, despite the protein catabolic effects of excess glucocorticoids (Hausberger and Hausberger, 1958). The explanation for this apparent paradox is that although glucocorticoids have marked fat-mobilizing effects, hyperadrenocorticism is usually associated with islet hyperplasia, beta-cell degranulation, and hyperinsulinaemia. Hyperinsulinism, in turn, causes hyperphagia and enhances fat deposition, *i.e.* insulin is adipokinetic, and can compensate for the catabolic effects of steroids. We believe that similar abnormal hypothalamic–pituitary regulation is an integral part of the genetically-programmed spontaneous hypertension, hyperglycaemia and hyperlipidaemia of SHR in general, and that in the Cushingoid Obese/SHR the hyperadrenocorticism is intensified. It is well known that adrenal steroid production and its metabolic clearance rate is commensurate with the waxing and waning of body-surface area or obesity and that the signs and symptoms of patients with Cushing's disease or abnormal obesity are hard to differentiate (Poisnick & Di Raimondo, 1956; Cohen, 1958; Schteingart, Gregerman & Conn, 1963). Hypophysectomy or adrenalectomy will ameliorate the obesity of humans, rats and mice (Bray and York, 1979).

Most animal models of hypothalamic obesity manifest hyperphagia, hyperlipidaemia, hyperinsulinaemia and enlarged fat cells (Bray, 1977; Herberg and Coleman, 1977; Morrison, 1977). Our Obese/SHR are no exception. Pituitary-gland size is reduced in experimental models of obesity, which may account for the hypothyroidism, impaired reproductive function and decreased muscle growth, *i.e.* the hypothalamic–pituitary axis is the prime locus of the obesity syndrome. It should be emphasized that blood pressure is best correlated with body weight than with body fat (Mann, 1974). The gross adiposity of these Obese/SHR, in this regard, is misleading.

We have observed consistently elevated serum enzyme levels, *e.g.* CPK, SGOT, SGPT and LDH, in SHR which is indicative of myocardial and hepatic damage. This is consistent with our observation of the frequent occurrence of myocardial necrosis and fatty liver in SHR, as well as in the sub-strain of Obese and non-obese/SHR.

All sub-strains of SHR manifest islet hyperplasia and hyperglycaemia (Wexler, Iams and Judd, 1976, 1977). Obese/SHR have enormous islets compared to non-obese SHR, yet display relatively mild hyperglycaemia. Obese patients and animals characteristically develop hyperinsulinaemia and depressed growth-hormone levels (Mann, 1974; Bray and York, 1979), as we observed in our Obese/SHR. We ascribe the hyperinsulinaemia to the gluconeogenic effects of the hyperadrenocorticism of Obese/SHR. The depressed growth and growth-hormone levels is ascribed to the anti-growth effects of the extra-adrenal steroids, reduced pituitary gland size and reduced growth-hormone secretion resulting from obesity *per se*. Although the BUN levels in our Obese/SHR were definitely abnormal, we did not observe the proteinuria or advanced glomerulopathy that Koletsky (1973, 1975) found in his Obese/SHR. Martin and Gahagan (1977*a*) have found increased amino-acid catabolism and decreased

nitrogen retention in the genetically obese Zucker rat and, like us, ascribe these changes to the reduced growth-hormone secretion and increased corticosterone levels in obesity. Like Cushing's disease patients, the skin of Obese/SHR is very thin, tears easily, and shows poor wound repair. We have found that despite their severe hypertension, SHR maintain low BUN levels and renal lesions do not appear until well after their hypertension has become malignant.

The SHR strain is hyper-responsive to stress, *e.g.* leaping and cavorting in their cages in response to sudden noise, and very combative when handled. We have found that concomitantly with their progressively increasing blood pressure (with age), SHR secrete increased quantities of adrenal steroids in response to stress (Iams, McMurtry and Wexler, 1979). It is intriguing that under quiescent conditions the adrenal steroid levels of SHR are normal and even subnormal compared to those of normotensive strains, of rat but under conditions of acute stress SHR respond by secreting unusually high levels of adrenal steroids, particularly the Cushingoid Obese/SHR. One would expect that the chronic hypercorticism of SHR would be associated with thymus-gland involution, *i.e.* due to the thymolytic effect of glucocorticoids. Although this does occur in SHR, the converse was found in Obese/SHR. Mineralocorticoids, *e.g.* DOC, are thymotrophic. The abnormally enlarged thymi observed in Obese/SHR could be a reflection of their capacity to secrete an extra abundance of thymotrophic mineralocorticoids as distinct from glucocorticoids. It should be emphasized that the increased size and weight of the thymi of Obese/SHR was due to white-cell hyperplasia and was not due to fatty infiltration.

In addition to abnormal pituitary–adrenal function in spawning salmon and breeder rats (Robertson and Wexler, 1957; Wexler, 1964), we have found that there is progressively increasing prolactin secretion concomitant with the pathogenesis and the progressive exacerbation of the Cushing's-disease-like changes (Lewis and Wexler, 1975). Others have reported the induction of obesity, hyperglycaemia, hypertension and related degenerative changes in animals bearing mammotrophic tumours (MtT) which secrete excessive quantities of prolactin (Bates, Garrison and Morris, 1966). Prolactin not only affects lipid and carbohydrate metabolism but causes aldosterone and increased contractility of the arterial wall (Horrobin, 1973). Prolactin and corticosterone act in conjunction to cause fattening of migrating birds and fish. For these reasons, we measured the prolactin levels in these SH rats and found that SHR produce supernormal levels of prolactin in response to stress, in keeping with the observation made by some investigators that pituitary prolactin release and adrenal-gland secretion are interrelated (Boyar and Hellman, 1974). Like the unusual production of adrenal steroids in response to stress, the Obese/SHR secreted much more prolactin than the other sub-strains, underscoring the hyperadrenocorticism and hyperprolactinaemia of obesity. The converse situation of subnormal levels of prolactin in Obese/SHR under quiescent or minimal stress conditions is in keeping with the findings of others (Sinha, Salocks and Vanderlaan, 1975; Martin and Gahagan, 1977*b*).

Perhaps the most salient feature of this investigation was the absence of atherosclerotic lesions both grossly and microscopically in our Obese/SHR and non-obese SHR. This is in direct contrast to Koletsky's finding (1973, 1975) of gross and microscopic atheromatous lesions in his obese and non-obese SHR, *i.e.* grossly visible, nodular, thrombotic and aneurysmal aortic lesions in 50% of his obese rats and 10% of his non-obese controls. Koletsky's obese and non-obese rats also manifested generalized PAN lesions with pools of lipid within the media. Our Obese/SH and non-obese/SHR did not manifest any atheromatous changes grossly or microscopically, but a few of our animals

did develop PAN. We should like to suggest that the *modus operandi* of these diverse genetic vectors may be expressed through sub-strain differences in adrenocortical secretion. For example, we have found that repeatedly bred SHR do not develop arteriosclerosis like all other strains of breeder rats (Wexler, Iams and Judd, 1976), but when repeatedly bred SH rats are subjected to adrenal enucleation the altered spectrum of hypertension-inducing steroids, *i.e.* predominantly DOC, these breeder SHR develop aortic sclerosis identical to the spontaneous arteriosclerosis observed in other strains of breeder rats (Wexler, Iams and Judd, 1977; Wexler, 1979*a*,*b*). This would suggest that the morphological expression of arterial disease in SHR is conditioned by the balance of spectrum of circulating adrenal steroids.

The authors commend the expertise of D. Conatser, E. Domingo, G. Williamson, D. Schaffner, R. Brand, D. Wayne, G. Heap and K. Kern. Dr N. Kazama supervised part of the steroid determinations in this investigation.

This work was supported in part by grants from the National Institute of Heart, Lung and Blood Diseases (HL-21418), the National Institute on Aging (AG-585), the U.S. Public Health Service, and the Southwestern Ohio Heart Association.

The authors express their appreciation to Dr W. K. Morishige, University of Hawaii, for supplying anti-porcine insulin antibody No. 18.

REFERENCES

Bates, R. W., Garrison, M. M. & Morris, H. P. (1966) Comparison of Two Different Transplantable Mammotrophic Pituitary Tumours. Hormone Content and Effect on Host. *Proc. Soc. exp. Biol. Med.*, **123,** 67.

Boyar, R. M. & Hellman, L. (1974) Syndrome of Benign Nodular Adrenal Hyperplasia associated with Feminization and Hyperprolactinemia. *Ann. intern. Med.*, **80,** 389.

Bray, G. A. (1977) The Zucker-fatty Rat: a Review. *Fed. Proc.*, **36,** 148.

Bray, G. A. & York, D. A. (1979) Hypothalamic and Genetic Obesity in Experimental Animals: an Autonomic and Endocrine Hypothesis. *Physiol. Rev.*, **59,** 719.

Cohen, H. (1958) 17-ketogenic Steroid Excretion in Obese Children before and after Weight Reduction. *Br. med. J.*, **1,** 686.

Hausberger, F. X. & Hausberger, B. C. (1958) Effect of Insulin and Cortisone on Weight Gain, Protein and Fat Content of Rats. *Am. J. Physiol.*, **193,** 455.

Herberg, L. & Coleman, D. L. (1977) Laboratory Animals exhibiting Obesity and Diabetes Syndromes. *Metabolism*, **26,** 59.

Horrobin, D. R. (1973) In *Prolactin: Physiology and Clinical Significance.* Lancaster: MTP Medical and Technical Publ. Co. Ltd.

Iams, S. G. & Wexler, B. C. (1976) Aldosterone, Deoxycorticosterone and Corticosterone Differences between Arteriosclerotic Breeder *vs* Non-arteriosclerotic Virgin Rats. *Atherosclerosis*, **24,** 63.

Iams, S. G., McMurty, J. P. & Wexler, B. C. (1979) Aldosterone, Deoxycorticosterone, Corticosterone, and Prolactin Changes during the Lifespan of Chronically and Spontaneously Hypertensive Rats. *Endocrinology*, **104,** 1357.

Koletsky, S. (1973) Obese Spontaneously Hypertensive Rats—A Model for Study of Atherosclerosis. *Exp. mol. Path.*, **19,** 53.

Koletsky, S. (1975) Pathologic Findings and Laboratory Data in a New Strain of Obese Hypertensive Rats. *Am. J. Path.*, **80,** 129.

Lewis, B. K. & Wexler, B. C. (1975) Changes in LH and Prolactin Associated with Arteriosclerosis in Breeder Rats. *Atherosclerosis*, **21,** 301.

Mann, G. V. (1974) The Influence of Obesity on Health. *New Engl. J. Med.*, **291,** 178.

Martin, R. J. & Gahagan, J. H. (1977*a*) The Influence of Age and Fasting on Serum Hormones in the Lean and Obese Zucker Rat. *Proc. Soc. exp. Biol. Med.*, **154,** 610.

Martin, R. J. & Gahagan, J. H. (1977*b*) Serum Hormone Levels and Tissue Metabolism in Pair-fed Lean and Obese Zucker Rats. *Horm. Metab. Res.*, **9,** 181.

Morishige, W. K., Uetake, C. A., Greenwood, F. C. & Akaka, J. (1977) Pulmonary Insulin Responsivity: *In Vivo* Effects of Insulin on the Diabetic Rat Lung and Specific Insulin Binding to Lung Receptors in Normal Rats. *Endocrinology*, **100,** 1710.

Morrison, S. D. (1977) The Hypothalamic Syndrome in Rats. *Fed. Proc.*, **36,** 139.

Okamoto, K. & Aoki, K. (1963) Development of a Strain of Spontaneously Hypertensive Rats. *Jap. Circulation J.*, **27,** 282.

Poisnick, J. & di Raimondo, V. (1956) Adrenocortical Function in Obese Women. *J. clin. Endocrinol.*, **16,** 957.

Robertson, O. H. & Wexler, B. C. (1957) Pituitary Degeneration and Adrenal Tissue Hyperplasia in Spawning Pacific Salmon. *Science*, **125,** 1295.

Robertson, O. H. & Wexler, B. C. (1962) Histological Changes in the Pituitary Gland of the Rainbow Trout (*Salmo gairdnerii*) accompanying Sexual Maturation and Spawning. *J. Morphology*, **110,** 157.

Robertson, O. H., Wexler, B. C. & Miller, B. F. (1961) Degenerative Changes in the Cardiovascular System of the Spawning Pacific Salmon (*Oncorhynchus tshawytscha*). *Circulation Res.*, **9,** 826.

Schteingart, D. E., Gregerman, R. I. & Conn,

J. W. (1963) A Comparison of the Characteristics of Increased Adrenocortical Function in Obesity and in Cushing's Syndrome. *Metabolism*, **12**, 484.

SINHA, Y. N., SALOCKS, C. B. & VANDERLAAN, W. P. (1975) Prolactin and Growth Hormone Secretion in Chemically Induced and Genetically Obese Mice. *Endocrinology*, **97**, 1386.

WEXLER, B. C. (1964) Spontaneous Arteriosclerosis in Repeatedly Bred Male and Female Rats. *J. Atheroscler. Res.*, **4**, 57.

WEXLER, B. C. (1970) Arteriosclerosis of the Pancreas and Changes in the Islets of Langerhans of Repeatedly Bred Rats. *Br. J. exp. Path.*, **51**, 107.

WEXLER, B. C., IAMS, S. G. & JUDD, J. T. (1976) Arterial Lesions in Repeatedly Bred Spotaneously Hypertensive Rats. *Circulation Res.*, **38**, 494.

WEXLER, B. C., IAMS, S. G. & JUDD, J. T. (1977) Comparative Effects of Adrenal Regeneration Hypertension on Non-arteriosclerotic and Arteriosclerotic Sprague–Dawley *vs* Spontaneously Hypertensive Rats. *Atherosclerosis*, **26**, 1.

WEXLER, B. C. & KITTINGER, G. W. (1965) Adrenocortical Function in Arteriosclerotic Female Breeder Rats. *J. Atheroscler. Res.*, **5**, 317.

WEXLER, B. C. (1979*a*) Myocardial Infarction in Spontaneously Hypertensive Rats (SHR) with Superimposed Adrenal Regeneration Hypertension. *Br. J. exp. Path.*, **60**, 366.

WEXLER, B. C. (1979*b*) Induction of Diverse Arterial and Myocardial Lesions by Adrenal Regeneration Hypertension in Sprague–Dawley *vs* Spontaneously Hypertensive Rats. *Endocrinology*, **106**, 935.

YEN, T. T., SHAW, W. N. & YU, P. L. (1977) Genetics of Obesity in Zucker Rats and Koletsky Rats. *J. Heredity*, **38**, 373.

ZUCKER, L. M. & ZUCKER, T. F. (1961) Fatty, a New Mutation in the Rat. *J. Heredity*, **52**, 275.