Obesity and reproduction

By ELAINE M. WHITAKER, *Department of Physiology, Leeds University, Leeds LS2 9JT*

An association between obesity and reproductive abnormalities has been reported in a variety of species including rodents and humans. At present it is difficult to provide a unifying theory to explain the association. Furthermore the experimental approaches have been different in the two species. Investigations into the reproductive abnormalities of obese rodents have been mainly concerned with hypothalamic and pituitary functions. Most of the clinical studies have been directed towards circulating steroid hormones.

Rodents

Abnormal reproductive function has been described in experimental obesity as well as in several inherited forms of obesity. Obesity due to overfeeding is associated with prolonged oestrus cycles and reduced rates of pregnancy (Innami *et al.* 1973; Rolls & Rowe, 1982). Reproductive abnormalities have also been reported in animals made obese by damage to the hypothalamus (Bray & York, 1979). Most of the studies of obesity and reproduction, however, have been described in recessively inherited forms of obesity such as the *fa/fa* (Zucker) rat and the *ob/ob* mouse.

fa/fa *Zucker rat.* The obese (*fa/fa*) Zucker rat was first described by Zucker & Zucker (1961), who reported that obese females were always sterile and obese males occasionally fertile. Several reproductive abnormalities have been described. Reproductive behaviour is reported to be abnormal in obese males (Edmonds & Withyachumnarnkul, 1980; Withyachumnarnkul & Edmonds, 1982). Obese male Zucker rats show reduced weights of seminal vesicles (Edmonds *et al.* 1982) and dorsal bulbocavernosus (Whitaker *et al.* 1983); weights of testes and ventral prostate are reported to be normal (Edmonds *et al.* 1982; Whitaker *et al.* 1983; Bannister & Whitaker, 1985). A study of testis morphology in obese males, however, showed hypertrophy of the Leydig cells which contained many lipid droplets (Young *et al.* 1982).

Plasma testosterone is reduced in obese males when compared with non-obese males (Edmonds *et al.* 1982; Young *et al.* 1982; Withyachumnarnkul & Edmonds, 1982; Whitaker *et al.* 1983); it is unlikely, however, that the small reduction in testosterone is responsible for the various reproductive abnormalities observed. The effect of testosterone treatment on improvement of reproductive function is controversial: Withyachumnarnkul & Edmonds (1982) reported no improvement in reproductive performance in obese males treated with testosterone, whereas

Hemmes *et al.* (1978) showed that large doses of testosterone improved fertility in young obese male Zucker rats. Basal concentrations of luteinizing hormone (LH) and follicle-stimulating hormone (FSH) and responses of these hormones to LH releasing hormone (LHRH) are normal in obese males (Young *et al.* 1982). We have measured androgen receptors in the prostate and liver of male Zucker rats (Table 1) and have found reduced concentrations in both organs in obese males (Bannister & Whitaker, 1985). This reduction in receptor concentration may reflect a widespread lack of sensitivity to circulating androgens in obese male Zucker rats; reduced sensitivity of central nervous tissue to circulating androgens could contribute to the behavioural dysfunction seen in the obese rats.

Female obese Zucker rats are reported to have enlarged ovaries and reduced weights of uteri (Saiduddin *et al.* 1973; Bray *et al.* 1973). Vaginal opening is delayed and oestrus cycles are irregular and prolonged (Saiduddin *et al.* 1973; Hervey *et al.* 1981; Whitaker *et al.* 1983). Saiduddin *et al.* (1973) failed to induce pseudopregnancy in obese females unless the females were treated with reserpine; even with reserpine treatment no decidual reaction was seen in the uterus of pseudopregnant obese rats. Reproductive behaviour is abnormal in obese female rats (Saiduddin *et al.* 1973; Chelich & Edmonds, 1981); Chelich & Edmonds (1981), however, reported that a small number of young obese females did become pregnant. Bray *et al.* (1973) reported that weight reduction did not improve reproductive function. Several investigators have interpreted the reproductive abnormalities seen in obese female rats as evidence of reduced oestrogen concentration (Saiduddin *et al.* 1973; Bray & York, 1979). Saiduddin *et al.* (1973) reported normal circulating gonadotrophins during dioestrus and normal increases after ovariectomy in obese female rats. These findings are not consistent with a

Table 1. *Concentrations of cytoplasmic receptors (fmol/mg protein) in non-obese and obese Zucker rats*

(Values are means with their standard errors; no. of rats in parentheses)

	Androgen receptors				
	Non-obese		Obese		Statistical significance of difference
Male Zucker rats	Mean	SE	Mean	SE	
Liver	18·8(11)	1·5	9·5(10)	1·0	$P<0.001$
Prostate	165·0(11)	20·0	116·0(10)	15·0	$P<0.01$

	Hypothalamic oestrogen receptors				
	Non-obese		Obese		
Female Zucker rats	Mean	SE	Mean	SE	
Pre-optic area	30·2(7)	2·1	20·5(6)	3·1	$P<0.05$
Medial basal hypothalamus	19·0(7)	0·9	16·0(6)	1·8	NS

NS, not significant.

deficiency of oestrogens. We have measured concentrations of plasma oestradiol in Zucker rats and have found a similar pattern of oestradiol concentration over the oestrus cycle in obese and non-obese rats (Whitaker *et al.* 1983). There appears to be no gross deficiency in circulating oestradiol in obese female rats. Thus the most likely causes of the abnormalities are impaired responsiveness of target tissues or a defect in central control of reproduction.

Saiduddin & Zassenhaus (1979) reported reduced concentrations of oestrogen receptors in the uteri of obese female rats and impaired interaction of oestradiol with these receptors. We have found reduced concentrations of oestrogen receptors in the pre-optic area of the hypothalamus of obese rats (Robinson & Whitaker, 1985), though concentrations of oestrogen receptors in the medial basal hypothalamus were similar in both phenotypes (Table 1). The pre-optic area is an important site for the feedback control of ovulation; reduced responsiveness of this target tissue to gonadal hormones could result in impaired control of ovulation.

Abnormal concentrations of hypothalamic catecholamines have been found in obese Zucker rats (Cruce *et al.* 1976); these findings are consistent with a central defect in the control of reproduction. Such a defect could result in abnormal secretion of the pituitary hormones involved in ovulation. We have tested this possibility by estimating concentrations of plasma LH, FSH and prolactin sequentially throughout the oestrus cycle in both phenotypes (Robinson & Whitaker, 1986). LH concentrations were found to be attenuated during pro-oestrus in obese rats when compared with non-obese rats (Fig. 1). FSH concentrations increased during pro-oestrus in both obese and non-obese rats but obese rats showed a slower decline in FSH during oestrus than non-obese rats (Fig. 2). Prolactin concentrations also increased during pro-oestrus in both obese and non-obese rats; prolactin, however, remained elevated during oestrus and early dioestrus in obese rats (Fig. 3). In non-obese rats prolactin decreased during the night of oestrus. Obese rats that had 2 d of oestrus showed elevated prolactin concentrations during both days of oestrus. The results of these studies show that secretion of gonadotrophins and prolactin is abnormal in obese female Zucker rats. The abnormal secretion of prolactin could account for the irregular timing of ovulation and prolongation of the oestrus cycle: increased concentration of prolactin during lactation is believed to suppress ovulation by inhibiting gonadotrophin secretion.

ob/ob *mouse.* Both male and female obese (*ob/ob*) mice show marked hypogonadism and infertility. Male *ob/ob* mice have lower weights of testes and ventral prostate than non-obese mice (Swerdloff *et al.* 1976), and female *ob/ob* mice have reduced weights of ovaries and uteri (Lane, 1959). Peripheral reproductive organs showed normal responses to exogenous steroids in both sexes and both phenotypes (Jones & Ainsworth-Harrison, 1958).

Basal circulating LH is low at all ages in *ob/ob* males, unlike non-obese males who show a peak of LH between 39 and 45 d of age. FSH is also reduced in *ob/ob* males. Both non-obese and *ob/ob* males show a rise in circulating testosterone between 60 and 70 d of age, but the concentrations are always much lower in the
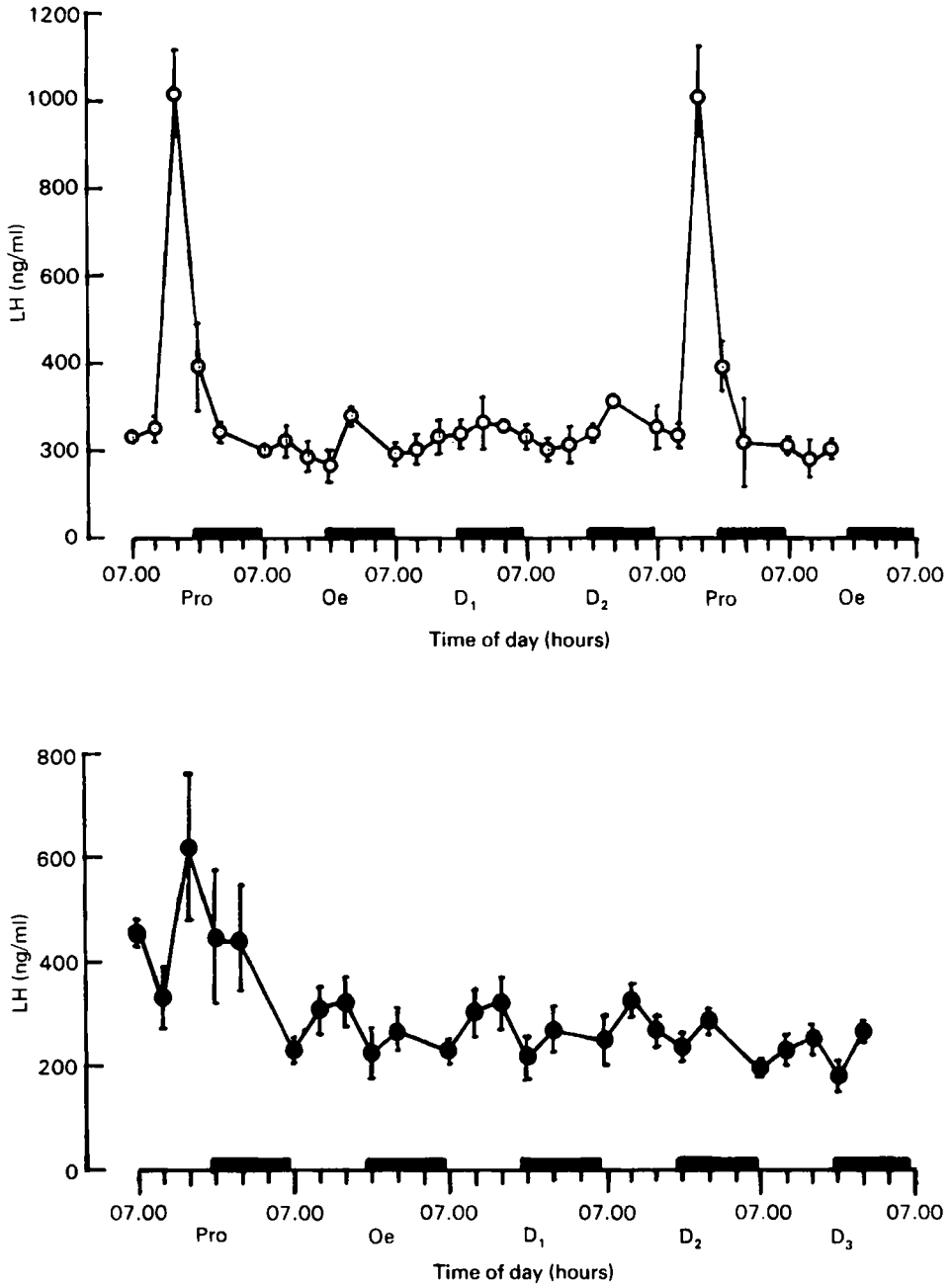
Fig. 1. Plasma luteinizing hormone (LH) concentrations in female Zucker rats throughout the oestrus cycle: non-obese (o), obese (●). Values are means, with their standard errors represented by vertical bars. Blood samples were taken every 4 h between 07.00 and 23.00 hours. (■), Dark period (18.30–06.30 hours); Pro, pro-oestrus; Oe, oestrus; D_1, D_2, D_3, dioestrus 1, 2 and 3 respectively.
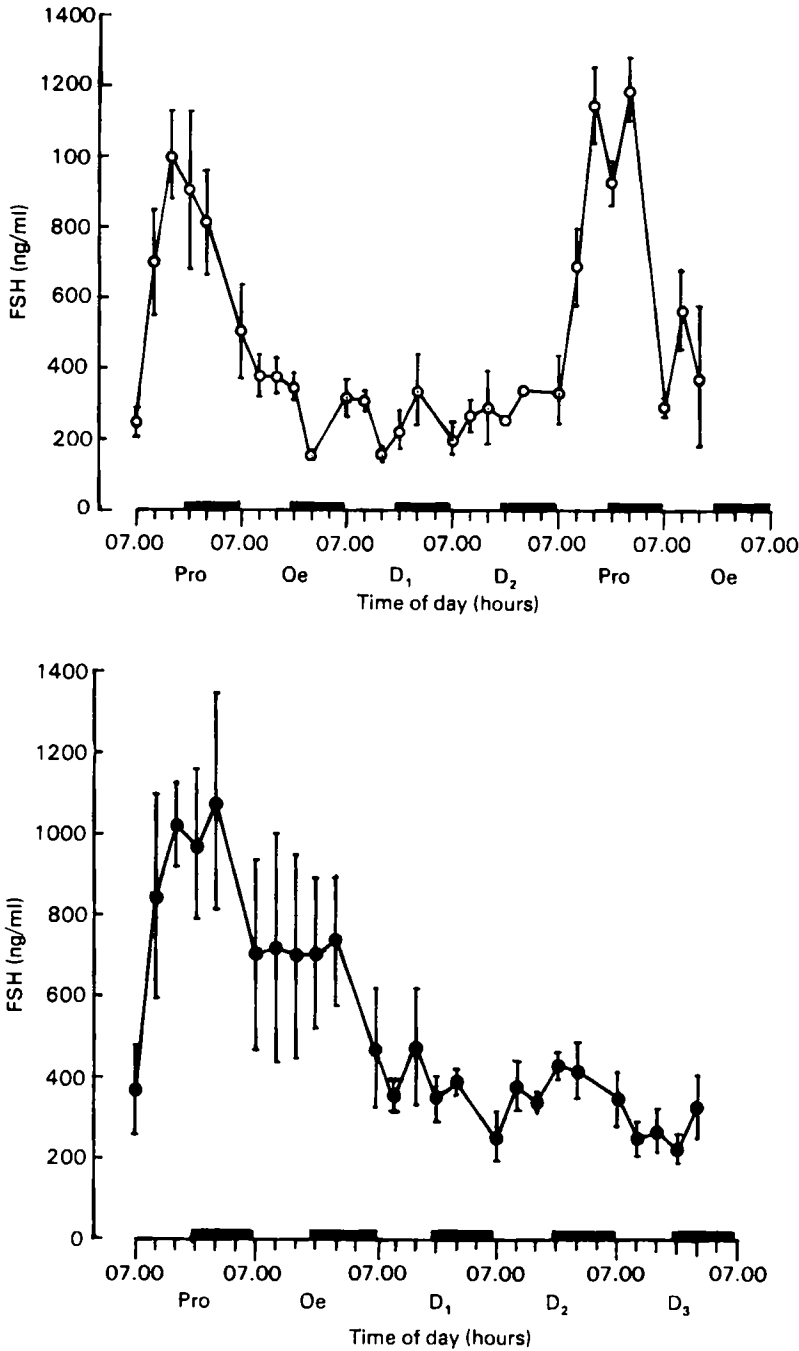
Fig. 2. Plasma follicle stimulating hormone (FSH) concentrations in female Zucker rats throughout the oestrus cycle: non-obese (o), obese (●). Values are means, with their standard errors represented by vertical bars. Blood samples were taken every 4 h between 07.00 and 23.00 hours. (■), Dark period (18.30–06.30 hours); Pro, pro-oestrus; Oe, oestrus; D_1, D_2, D_3, dioestrus 1, 2 and 3 respectively.
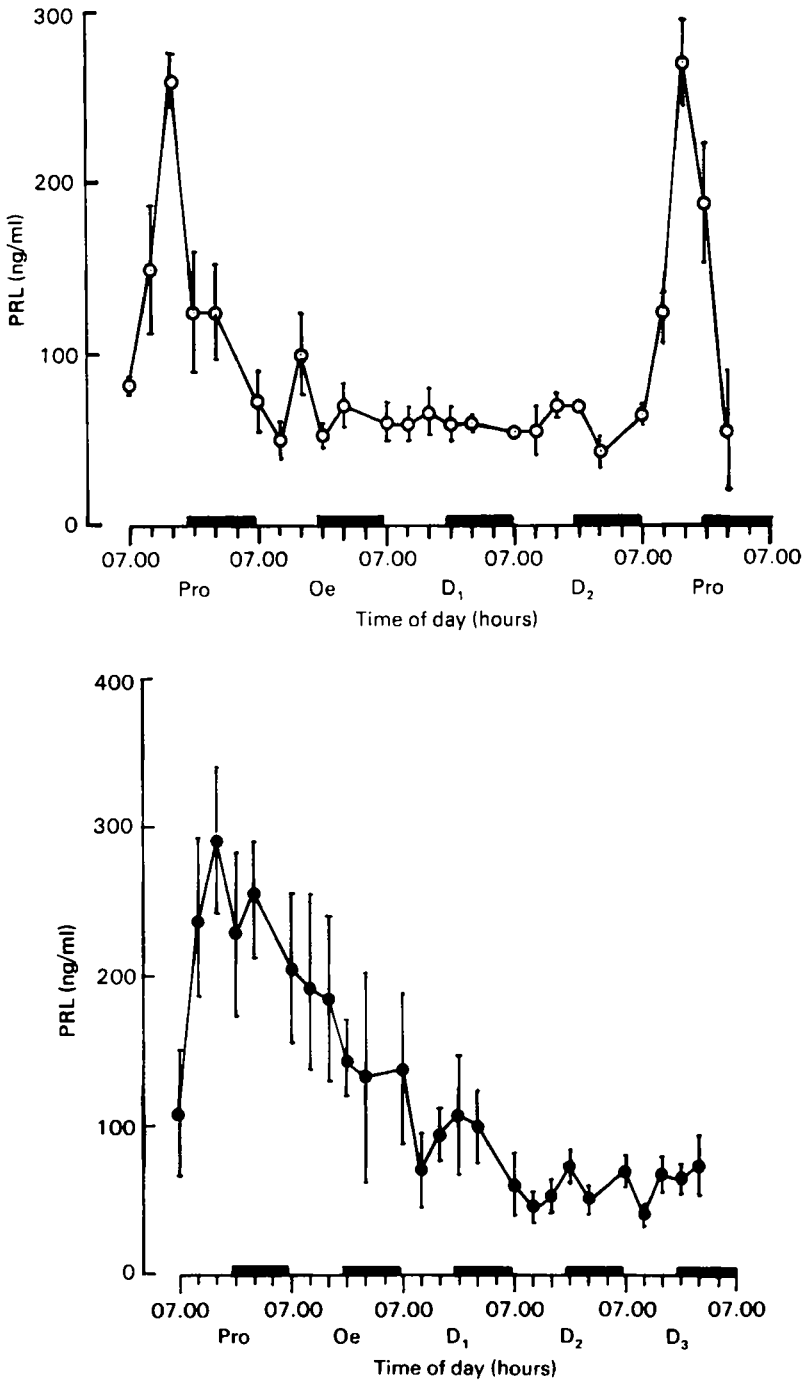
Fig. 3. Plasma prolactin (PRL) concentrations in female Zucker rats throughout the oestrus cycle: non-obese (o), obese (●). Values are means with their standard errors represented by vertical bars. Blood samples were taken every 4 h between 07.00 and 23.00 hours. (■), Dark period (18.30–06.30 hours); Pro, pro-oestrus; Oe, oestrus; D_1, D_2, D_3, dioestrus 1, 2, and 3 respectively.

ob/ob than in the non-obese mice. After gonadectomy, LH and FSH increase in both phenotypes, but the pre- and post-castration concentrations of both hormones are significantly lower in the *ob/ob* males. Administration of testosterone to castrated *ob/ob* male mice results in a more rapid suppression of LH and FSH than in non-obese males (Swerdloff *et al.* 1976). Circulating prolactin is reported to be reduced in male and female *ob/ob* mice (Sinha *et al.* 1975).

The reduction in basal concentrations of gonadotrophins and increased sensitivity of gonadotrophins to exogenous testosterone in *ob/ob* males suggest an abnormality in hypothalamic–pituitary function. Reduced hypothalamic content of LHRH has been shown in *ob/ob* mice (Nemeroff *et al.* 1978); Edwardson & Donaldson (1979), however, reported no such reduction. Attenuated responses of LH and FSH to acute and chronic administration of LHRH in *ob/ob* males indicate that the abnormality in reproductive function seen in *ob/ob* mice involves the pituitary as well as the hypothalamus (Swerdloff *et al.* 1978).

Central catecholamines are believed to be important in the control of reproductive hormones. Studies of catecholamine content in hypothalami and pituitaries of non-obese and *ob/ob* mice have shown increased concentrations of hypothalamic noradrenaline and dopamine, and decreased concentration of pituitary dopamine in *ob/ob* mice (Lorden & Oltmans, 1977; Edwardson & Donaldson, 1979). These findings support the proposition that a central defect in the control of reproduction exists in *ob/ob* mice.

Recessively inherited diseases are of great interest. An autosomal recessive trait implies a single gene mutation which is believed to result in a single defective peptide or protein. Multiple abnormalities are seen in recessively inherited diseases but they are presumed to arise as a consequence of the primary lesion. The reproductive abnormalities described in the obese (*fa/fa*) Zucker rat and the *ob/ob* mouse point to a defect in hypothalamic function. Other abnormalities have been described in the *fa/fa* rat and *ob/ob* mouse which are also indicative of hypothalamic dysfunction: hyperphagia, impaired temperature regulation and impaired secretion of growth hormone. It is tempting to speculate that these abnormalities reflect a primary defect within the hypothalamus. Our evidence from studies in the *fa/fa* Zucker rat leads us to suggest that the defective protein is a receptor protein.

Clinical studies

Obesity and amenorrhoea. There is a widespread clinical impression that obesity is associated with menstrual irregularities such as dysfunctional bleeding, amenorrhoea or oligomenorrhoea. Rogers & Mitchell (1952) reported that the incidence of obesity in women with menstrual abnormalities was 43% compared with 13% in women with no menstrual abnormalities. Women who were 20% or more above their ideal weight were considered to be obese. Several other investigators observed that chronic anovulation (including polycystic ovary syndrome) is two to four times as common in obese women as in women of normal

body-weight (Hartz *et al.* 1979; Yen, 1980). The causes of amenorrhoea in obese women are poorly understood. There have been a number of reports on hormonal changes associated with obesity and the possible role these hormones may play in the etiology of amenorrhoea. Few of these studies have attempted to investigate ovulatory and anovulatory obese women separately.

Studies of obesity and menstrual irregularities have been concerned mainly with measurements of sex steroids and sex-hormone-binding globulin (SHBG) in blood. The evidence suggests that increased amounts of androgens not bound to proteins may contribute to the menstrual abnormalities. A reduction in the concentration of SHBG is the most consistent finding in premenopausal obese women (Hosseinian *et al.* 1976; Kopelman *et al.* 1980a; Nisker *et al.* 1980; Plymate *et al.* 1981; Mathur *et al.* 1982; Sulkes *et al.* 1984; Zhang *et al.* 1984; Loughlin *et al.* 1985). Increased concentration of total testosterone has been reported in obese women by Hosseinian *et al.* (1976), Pasquali *et al.* (1983) and Zhang *et al.* (1984); but Kopelman *et al.* (1980a) and Loughlin *et al.* (1985) reported normal testosterone in their obese subjects. Free testosterone (i.e. non-protein-bound) is reported to be increased in obese women (Hosseinian *et al.* 1976; Zhang *et al.* 1984; Loughlin *et al.* 1985); this observation is consistent with the finding of reduced capacity of SHBG to bind androgens in obese women. Secretion of adrenal androgen androstenedione is also increased (Kopelman *et al.* 1980a; Mathur *et al.* 1982; Pasquali *et al.* 1983; Zhang *et al.* 1984) and this finding probably reflects the increased adrenal activity reported in obese subjects (see Bray, 1976). In two studies normal obese women were compared with anovulatory obese women and normal non-obese women; the greatest reduction in SHBG and the greatest increase in free testosterone occurred in the obese women who were not ovulating (Hosseinian *et al.* 1976; Zhang *et al.* 1984; Loughlin *et al.* 1985). The mechanism whereby increased free androgens contribute to the amenorrhoea observed in obese women is not clear: there is evidence that androgens inhibit follicular maturation (Louvet *et al.* 1975) and increased androgens may also act to alter hypothalamic–pituitary function.

Extraglandular production of oestrone may also be increased in obese women. Adipose tissue is known to be rich in the enzyme aromatase which catalyses conversion of androstenedione to oestrone (Schindler *et al.* 1972). Increased mass of adipose tissue could contribute to increased production of oestrone in these women, particularly since the availability of adrenal androgens is increased. There is evidence to support this possibility. Edman & MacDonald (1978) have reported enhanced conversion of androstenedione to oestrone in both ovulatory and anovulatory obese women. Several investigators have shown that circulating oestrone is increased in young obese women (Pasquali *et al.* 1983; Zhang *et al.* 1984; Loughlin *et al.* 1985), though Kopelman *et al.* (1980a) did not find this. Fishman *et al.* (1975) reported increased production of the more biologically active oestrogens and decreased production of the less biologically active 2-hydroxylated oestrogens in young obese women. The increased oestrogen production may contribute to anovulation by modifying the release of gonadotrophins in a similar

manner to that of raised androgens. Since androgens are believed to suppress and oestrogens stimulate the synthesis of SHBG it is not easy to explain the reduction in circulating SHBG seen in obese women.

Obesity and reproduction in men. Obesity in men rarely results in androgen deficiency despite reported reductions in circulating SHBG (Glass *et al.* 1977; Amatruda *et al.* 1978; Kley *et al.* 1979) and in circulating testosterone (Glass *et al.* 1977; Amatruda *et al.* 1978; Kley *et al.* 1979; Schneider *et al.* 1979). In most obese men free testosterone was normal. However, Barbato & Landau (1974) and Glass *et al.* (1977) reported reduced free testosterone in a few massively obese men. Oestrone secretion is increased in obese men (Kley *et al.* 1979; Schneider *et al.* 1979).

The changes in sex steroid secretion and sex steroid binding seen in obese women and men are reversed by weight reduction, and weight loss resulted in more regular menstrual cycles in the women (Kopelman *et al.* 1981; Stanik *et al.* 1981; Harlass *et al.* 1984). These findings suggest that the steroid hormone abnormalities are secondary to the obesity.

Several investigators have reported abnormal hypothalamic–pituitary function in obese subjects that may reflect a fundamental central defect. Plymate *et al.* (1981) have reported increased basal concentrations of LH in obese women with amenorrhoea, and Zhang *et al.* (1984) have reported similar increases in LH together with reduced basal concentrations of prolactin. Reduced nocturnal secretion of LH has been reported by Kalucy *et al.* (1976), and an exaggerated response of LH to LHRH has been shown in obese anovulatory women (Loughlin *et al.* 1985; Kopelman *et al.* 1980*a*). Prolactin response to insulin-induced hypoglycaemia has been shown to be absent in several groups of obese subjects, particularly in those who have been obese since childhood, (Kopelman *et al.* 1979, 1980*b,c*, 1983; Jung *et al.* 1982). Until there is more knowledge of hypothalamic function one can do no more than speculate on the possible importance of these abnormalities in causing amenorrhoea.

Obesity and puberty. Puberty is reported to be earlier in obese girls (Bruch, 1939; Mossberg, 1948; Wolff, 1955), and in obese boys (Bruch, 1939; Mossberg, 1948). Several possible causes might be envisaged. Frisch and colleagues have proposed that puberty depends on attainment of a critical body-weight or fat:lean tissue value, and that obese children reach this critical state earlier than normal weight children (e.g. Frisch, 1984). Adrenal androgen secretion, however, is increased in obese pubertal children (Garces, 1968; Genazzani *et al.* 1978; Pintor *et al.* 1984). Maturation of the hypothalamic–pituitary adrenal system is known to precede maturation of the hypothalamic–pituitary gonadal system in normal weight children. It is possible, therefore, that the increased adrenal activity observed in obese pubertal children may play a role in the earlier puberty seen.

Conclusion

An association between obesity and defective reproduction has been described in rodents and humans. The evidence from rodents can be taken to suggest that the

reproductive abnormalities and the obesity are both consequences of an underlying hypothalamic defect. This does not seem to be the case for humans: the available evidence suggests that the reproductive abnormalities are secondary to the obesity.

REFERENCES

Amatruda, J. M., Harman, S. M., Pourmotabbed, G. & Lockwood, D. H. (1978). *Journal of Clinical Endocrinology and Metabolism* **47**, 268–271.

Bannister, P. & Whitaker, E. M. (1985). *Journal of Endocrinology* **107**, R13–15.

Barbato, A. L. & Landau, R. L. (1974). *Clinical Research* **22**, 647A.

Bray, G. A. (1976). In *Major Problems in Internal Medicine, The Obese Patient*, vol. 9, pp. 252–299 [L. H. Smith, editor]. Philadelphia, London, Toronto: W. B. Saunders Company.

Bray, G. A. & York, D. A. (1979). *Physiological Reviews* **59**, 719–809.

Bray, G. A., York, D. A. & Swerdloff, R. S. (1973). *Metabolism* **22**, 435–442.

Bruch, H. (1939). *American Journal of Diseases of Childhood* **58**, 457–484.

Chelich, A. M. & Edmonds, E. S. (1981). *Physiology & Behaviour* **27**, 331–335.

Cruce, J. A. F., Thoa, N. B. & Jacobowitz, D. M. (1976). *Brain Research* **101**, 165–170.

Edman, C. D. & MacDonald, P. C. (1978). *American Journal of Obstetrics and Gynaecology* **130**, 456–461.

Edmonds, E. S., Dallie, S. K., Withyachumnarnkul, B. (1982). *Biology of Reproduction* **27**, 891–897.

Edmonds, E. S. & Withyachumnarnkul, B. (1980). *Physiology & Behaviour* **24**, 1139–1141.

Edwardson, J. A. & Donaldson, A. (1979). In *Animal Models of Obesity*, pp. 177–190 [M. W. Festing, editor]. London: Macmillan Press Ltd.

Fishman, J., Boyat, R. M. & Hellman, L. (1975). *Journal of Endocrinology and Metabolism* **41**, 989–991.

Frisch, R. E. (1984). *Biological Reviews* **59**, 161–188.

Garces, L. Y., Kenny, F. M., Drash, A. & Taylor, F. H. (1968). *Journal of Clinical Endocrinology and Metabolism* **28**, 1843–1847.

Genazzani, A. R., Pintor, C. & Corda, R. (1978). *Journal of Clinical Endocrinology and Metabolism* **47**, 974–979.

Glass, A. R., Swerdloff, R. S., Bray, G. A., Dahms, W. T. & Atkinson, R. L. (1977). *Journal of Clinical Endocrinology and Metabolism* **45**, 1211–1219.

Harlass, F. E., Plymate, S. R., Fariss, B. L. & Belts, R. P. (1984). *Fertility and Sterility* **42**, 649–652.

Hartz, A. J., Barboriak, P. N., Wong, A., Katayama, K. P. & Rimm, A. A. (1979). *International Journal of Obesity* **3**, 57–73.

Hemmes, R. B., Hubsch, S. & Pack, H. M. (1978). *Proceedings of the Society for Experimental Biology and Medicine* **159**, 424–427.

Hervey, G. R., Shaw, M. A. & Whitaker, E. M. (1981). *Journal of Physiology* **325**, 67–68P.

Hosseinian, A. H., Kim, M. H. & Rosenfield, R. L. (1976). *Journal of Clinical Endocrinology and Metabolism* **42**, 765–769.

Innami, S., Young, M. G., Mickelsen, D. & Hafs, H. D. (1973). *Proceedings of the Society for Experimental Biology and Medicine* **143**, 63–68.

Jones, N. & Ainsworth-Harrison, G. (1958). *Studies in Fertility* **9**, 51–64.

Jung, R. T., James, W. P. T., Campbell, R. G. & Callingham, B. A. (1982). *Lancet* i, 1043–1046.

Kalucy, R. S., Crisp, A. H., Chard, T., McNeilly, A., Chen, C. N. & Lacey, J. H. (1976). *Journal of Psychosomatic Research* **20**, 595–604.

Kley, H. K., Solbach, H. G., McKinnon, J. C. & Kruskemper, H. L. (1979). *Acta Endocrinologica* **91**, 553–563.

Kopelman, P. G., Apps, M. C., Cope, T. & Empey, D. W. (1983). *British Medical Journal* **287**, 859–861.

Kopelman, P. G., Pilkington, T. R. E., White, N. & Jeffcoate, S. L. (1979). *Lancet* i, 747–750.

Kopelman, P. G., Pilkington, N., White, N. & Jeffcoate, S. L. (1980*a*). *Clinical Endocrinology* **12**, 363–369.

Kopelman, P. G., Pilkington, N., White, N. & Jeffcoate, S. L. (1980*b*). *British Medical Journal* **280**, 82–83.

Kopelman, P. G., Pilkington, N., White, N. & Jeffcoate, S. L. (1980*c*). *British Medical Journal* **281**, 358–359.

Kopelman, P. G., White, N., Pilkington, T. R. E. & Jeffcoate, S. L. (1981). *Clinical Endocrinology* **14**, 113–116.

Lane, P. W. (1959). *Endocrinology* **65**, 863–868.

Lorden, J. F. & Oltmans, G. A. (1977). *Brain Research* **131**, 162–166.

Loughlin, T., Cunningham, S. K., Culliton, M., Smyth, P. P. A., Meagher, D. J. & McKenna, T. J. (1985). *Fertility and Sterility* **43**, 720–725.

Louvet, J. P., Harman, S. M., Schreiber, J. R. & Ross, G. T. (1975). *Endocrinology* **97**, 366–372.

Mathur, R. S., Moody, L. O., Landgrebe, S. C., Peress, M. R., Rust, P. F. & Williamson, H. O. (1982). *Fertility and Sterility* **38**, 207–211.

Mossberg, H. O. (1948). *Acta Pediatrica* **35**, Suppl. 2, 1–122.

Nemeroff, C. B., Bissette G. & Kizer, J. S. (1978). *Brain Research* **146**, 358–387.

Nisker, J. A., Hammond, G. L. & Siiteri, P. K. (1980). *New England Journal of Medicine* **303**, 1124.

Pasquali, R., Casimirri, F., Venturoli, S., Paradisi, R., Mattioli, L., Capelli, M., Melchionda, N. & Labo, G. (1983). *Acta Endocrinologica* **104**, 110–116.

Pintor, C., Loche, S., Faedda, A., Nurchi, A. M. & Corda, R. (1984). *Hormones and Metabolic Research* **16**, 544–548.

Plymate, S. R., Fariss, B. L., Bassett, M. L. & Matej, L. (1981). *Journal of Clinical Endocrinology and Metabolism* **52**, 1246–1248.

Robinson, A. C. & Whitaker, E. M. (1985). *Journal of Physiology* **365**, 115P.

Robinson, A. C. & Whitaker, E. M. (1986). *Journal of Physiology* **381**, 43P.

Rogers, J. & Mitchell, G. W. (1952). *New England Journal of Medicine* **247**, 53–55.

Rolls, B. J. & Rowe, E. A. (1982). *Physiology and Behaviour* **28**, 393–400.

Saiduddin, S., Bray, G. A., York, D. A. & Swerdloff, R. S. (1973). *Endocrinology* **93**, 125–126.

Saiduddin, S. & Zassenhaus, H. P. (1979). *Biology of Reproduction* **20**, 253–260.

Schindler, A. E., Ebert, A. & Friedrich, E. (1972). *Journal of Clinical Endocrinology and Metabolism* **35**, 627–630.

Schneider, G., Kirschner, M. A., Berkowitz, R. & Ertel, N. H. (1979). *Journal of Clinical Endocrinology and Metabolism* **48**, 633–638.

Sinha, Y. N., Salocks, C. B. & Vanderlaan, W. P. (1975). *Endocrinology* **97**, 1386–1393.

Stanik, S., Dornfeld, L. P., Maxwell, M. H., Viosca, S. P. & Korenman, S. G. (1981). *Journal of Clinical Endocrinology and Metabolism* **53**, 828–832.

Sulkes, A., Fuks, Z., Gordon, A. & Gross, J. (1984). *European Journal of Cancer and Clinical Oncology* **20**, 19–23.

Swerdloff, R. S., Batt, R. A. & Bray, G. A. (1976). *Endocrinology* **98**, 1359–1364.

Swerdloff, R. S., Peterson, M., Batt, A. V. R., Heber, D. & Bray, G. A. (1978). *Endocrinology* **103**, 542–547.

Whitaker, E. M., Shaw, M. A. & Hervey, G. R. (1983). *Journal of Endocrinology* **99**, 485–490.

Withyachumnarnkul, B. & Edmonds, E. S. (1982). *Physiology and Behaviour* **29**, 773–777.

Wolff, O. H. (1955). *Quarterly Journal of Medicine* **24**, 109–123.

Young, R. A., Frink, R. & Longcope, C. (1982). *Endocrinology* **111**, 977–981.

Yen, S. S. C. (1980). *Clinical Endocrinology* **12**, 177–208.

Zhang, Y., Stern, B. & Rebar, R. W. (1984). *Journal of Clinical Endocrinology and Metabolism* **58**, 1077–1083.

Zucker, L. M. & Zucker, T. F. (1961). *Journal of Heredity* **52**, 275–278.