

SHORT COMMUNICATIONS

POSTOVULATORY LEVELS OF PROGESTOGENS, OESTROGENS, LUTEINIZING HORMONE AND FOLLICLE-STIMULATING HORMONE IN THE PLASMA OF AGED GOLDEN HAMSTERS EXHIBITING A DELAY IN FERTILIZATION

T. A. PARKENING*, S. K. SAKSENA† AND I. F. LAU†

**Department of Anatomy, University of Texas Medical Branch, Galveston, Texas 77550, U.S.A. and †The Worcester Foundation for Experimental Biology, Shrewsbury, Massachusetts 01545, U.S.A.*

(Received 5 December 1977)

Fertilization is delayed approximately 2–5 h in the majority of ova recovered from aged hamsters (14–17 months old), compared with younger (3–5 months old) animals (Parkening & Soderwall, 1975). This delay may explain the relatively large percentage (40%) of ova which is incapable of developing to the implantation stage in this species (Parkening & Soderwall, 1975). Blaha & Leavitt (1974) have shown that there is a wider variation in the peripheral plasma concentration of progesterone at the time of fertilization in aged than in younger hamsters. A hormonal imbalance in the senescent female hamster may disturb normal gamete interactions, thus causing the delay in fertilization. In order to determine the significance of hormonal levels with respect to fertilization, the concentrations of progestogens, oestrogens, luteinizing hormone (LH) and follicle-stimulating hormone (FSH) in peripheral plasma were analysed by radioimmunoassay in mature and old hamsters.

Mature Syrian hamsters (3–5 months old), which had produced one or two litters and senescent hamsters (14–17 months old), which were received as retired breeders at 10 months of age, were maintained on a 13 h light : 11 h darkness cycle. All female hamsters exhibited three normal 4 day oestrous cycles before being placed with a male hamster of proven fertility in the early evening. Animals were observed to ensure mating had taken place and ovulation was considered to have occurred between 8 and 9 h after the onset of lordosis (Parkening & Soderwall, 1975). Blood samples (2.0–6.0 ml) were collected in heparinized syringes from the abdominal aorta of female hamsters, lightly anaesthetized with ether, between 3 and 5 h after the expected time of ovulation. Immediately after exsanguination, ova were collected from the ampulla of each oviduct and treated with 0.05% hyaluronidase in Hanks' solution to remove the cumulus cells, fixed in 3% glutaraldehyde and examined under a phase-contrast microscope for fertilization.

The concentrations of progesterone, oestradiol-17 β and oestrone in the plasma were determined by radioimmunoassay (Saksena, Lau & Chang, 1976) using 0.5 ml samples of plasma, after separation of the steroids on microcolumns of Celite (Lau, Saksena & Chang, 1976). For the determination of 20 α -hydroxyprogesterone, 0.2 ml plasma was extracted, subjected to chromatography and assayed as described by Maroulis & Abraham (1975). Plasma LH and FSH were determined by post-precipitation double-antibody radioimmunoassays, which were modifications of the procedures described by Niswender, Midgley, Monroe & Reichert (1968) and Gay, Niswender, Midgley & Reichert (1970) respectively.

There were no differences between the numbers of ova from 3- to 5-month-old (14.8 ± 0.80 (S.E.M.); $n = 17$) and 14- to 17-month-old (14.2 ± 0.78 ; $n = 19$) hamsters. Of these ova, 67% from the 3- to 5-month-old and 22% from the 14- to 17-month-old animals either contained a swollen sperm head and a female pronucleus or both male and female pronuclei and were, therefore, considered to be fertilized. All but one mature female hamster had some

ova in various stages of fertilization, but none of the ova from seven aged animals showed signs of becoming fertilized. In the samples of plasma collected from the above animals there were no statistical differences between the age groups in the levels of progesterone, 20 α -hydroxyprogesterone, oestradiol-17 β , oestrone, LH or FSH, as determined by Student's *t*-test (Table 1).

Table 1. Levels of various hormones in peripheral plasma collected from hamsters 3 to 5 h after the expected time of ovulation (means \pm S.E.M.; numbers of animals given in parentheses)

Hormones	Age of hamsters (months)	
	3-5	14-17
Progesterone (ng/ml)	7.1 \pm 0.7 (16)	5.5 \pm 0.6 (16)
20 α -Hydroxyprogesterone (pg/ml)	198.3 \pm 31.0 (9)	217.3 \pm 25.3 (10)
Oestradiol-17 β (pg/ml)	48.4 \pm 2.9 (17)	53.8 \pm 3.6 (18)
Oestrone (pg/ml)	85.6 \pm 4.3 (17)	86.2 \pm 8.0 (18)
LH (ng/ml)	7.4 \pm 1.6 (10)	10.9 \pm 1.7 (10)
FSH (ng/ml)	712.3 \pm 42.3 (10)	651.6 \pm 27.4 (10)

Although there were no statistical differences between the hormone concentrations in peripheral plasma from the two age groups, it is possible that local systemic differences may occur. However, Shaikh & Saksena (1973) indicated that the pattern of progesterone secretion appeared to be similar in mature female hamsters during the oestrous cycle whether plasma was collected from the peripheral system or from the ovarian vein. It seems that the delay in fertilization in aged female hamsters may not be hormone-related and that the interaction or attraction between the gametes may be altered because of a changing milieu in the older animal. *In vitro*, there were no differences in the numbers of ova fertilized or the times required for fertilization when ova from 3- to 5-month-old and 14- to 17-month-old hamsters were compared (Parkening & Chang, 1976). The passage of spermatozoa through the oviduct in senescent female hamsters may be impaired, but a previous study provided little evidence to support this hypothesis (Parkening & Soderwall, 1975). The specific cause(s) of the delay in fertilization in the senescent hamster remains to be determined.

REFERENCES

- Blaha, G. C. & Leavitt, W. W. (1974). *Biology of Reproduction* **11**, 153-161.
 Gay, V. L., Niswender, G. D., Midgley, A. R., Jr & Reichert, L. E., Jr. (1970). *Federation Proceedings* **29**, 439 (Abstr.).
 Lau, I. F., Saksena, S. K. & Chang, M. C. (1976). *Prostaglandins* **11**, 859-869.
 Maroulis, G. B. & Abraham, G. E. (1975). *Analytical Letters* **8**, 377-382.
 Niswender, G. D., Midgley, A. R., Jr, Monroe, S. E. & Reichert, L. E., Jr. (1968). *Proceedings of the Society for Experimental Biology and Medicine* **128**, 807-811.
 Parkening, T. A. & Chang, M. C. (1976). *Journal of Reproduction and Fertility* **48**, 381-383.
 Parkening, T. A. & Soderwall, A. L. (1975). *Biology of Reproduction* **12**, 618-631.
 Saksena, S. K., Lau, I. F. & Chang, M. C. (1976). *Steroids* **27**, 751-757.
 Shaikh, A. A. & Saksena, S. K. (1973). *Advances in the Biosciences* **9**, 635-639.