

# INFLUENCE OF NOR-ETHISTERONE ENANTHATE ON THE STRUCTURE OF OVARY, PARTICULARLY CORPUS LUTEUM

by

KAMALA ACHARI,\* M.B., M.S., F.I.C.S., F.R.C.O.G.

The female gonadal hormones, oestrogen and progesterone, either singly or in combination, are able to inhibit fertility. There has been considerable conjecture about the mechanism of their antifertility effect but it still remains undetermined. Studies with Chloromidanone acetate, a pure oral progestogen, have been noted to have a weak ovulostatic effect; the main mechanism of its action in the control of fertility is by its action on the endometrium and the cervical mucus. In this analysis, a new injectable contraceptive, nor-ethisterone enanthate (SH-393) has been used for the control of fertility. Ovarian biopsy has been performed to study the gonadal changes under the influence of injectable nor-ethisterone enanthate. This preparation gives contraceptive safety for three months.

## Material and Methods

A total of 50 cases were studied. The trial of nor-ethisterone enanthate (SH-393) was carried out in patients attending the Hospital for Women, Patna Medical College Hospital. Patients were chosen of known high fertility index and who were scheduled for sterilization by the abdominal

route. They had pretreatment cervical mucus, vaginal smear studies and premenstrual endometrial biopsy to exclude any apparent ovarian dysfunction. The criteria to be fulfilled for these cases to be included in this series were:

(1) Premenstrual endometrial biopsy must show a secretory pattern.

(2) Vaginal cytology and cervical mucus must indicate progesterone effect in the luteal phase and oestrogenic reaction in pre-ovulatory phase.

An injection of 200 mg. of SH-393 was given between the 5th and 7th days of menstrual cycle. The age of the patients ranged from 20 to 48 years and parity ranged from 3rd to 9th para. Ten cases were studied in the first month of treatment and 12 cases in the second month of treatment. The remaining 28 cases were operated in the third month of treatment. Every case had an endometrial biopsy and cervical mucus study throughout the treatment period.

The gross appearance of both the ovaries were noted during laparotomy. Wedge resection of the ovary was taken from an area which looked like a corpus luteum. In the absence of a corpus luteum bilateral wedge resection of the ovary was performed making a deep narrow section in the parenchyma. The sections were noted for the presence of recently formed —(1) corpora lutea, (2) presence

\*Prof. of Obst. & Gynec., P. W. Medical College, Patna, (Bihar).

Received for publication on 24-9-1969.

of fresh corpora albicantia which is proof of ovulation in the previous cycle.

#### Observation

This table shows that there was suppression of ovulation in 7 cases and that too in the second and third month of injection. The presence of recent corpora albicantia indicates that there had been recent ovulation in all the cases.

the nuclei are degenerating. Vacuoles are present in the cells. There are very few blood channels in the vacuolated granulosa cell layer. There is none or very little blood in the cavity of the corpus luteum.

Microphotograph 3 shows a normal corpus luteum on the 15th day of menstruation (Cycle 4/27-28 days). Granulosa cells are many layered. There are capillaries in the granulosa cell layer. Many of the granulosa

TABLE I

*Gives the general morphology of the ovaries on different days of the menstrual cycle counted arbitrarily from the first day of menstruation and different days after injection*

No. of cases	Days of menstrual cycle	Days after injection.	Ovarian Morphology		
			Atretic follicles or follicular cysts.	Corpus luteum	Corpus albicans.
4	15-23	9-14	—	+	+
2	24-26	18-20	—	+	+
4	26-30	21-25	—	+	+
10	47-57	40-50	—	+	+
2	47-57	40-50	+	—	+
23	63-97	56-90	—	+	+
5	63-97	56-90	+	—	+

Microphotograph I shows a normal corpus luteum on the 21st day of the menstrual cycle (Cycle 4/29-30). Luteinization of the granulosa cells, which are thick and many layered, is complete. The cytoplasm is abundant. The nuclei of the granulosa cells are vesicular. Each cell is surrounded by capillaries, many of which are dilated. Vacuoles are present along the border of the cavity. There is free blood in the central cavity and coarse fibrin is not present inside the lumen.

Microphotograph 2 shows a corpus luteum on the 22nd day of menstruation or 25th day after injection of SH-393. The granulosa layer is thin, consisting of only 3 to 4 layers and

cells are luteinized with typical lighter staining cytoplasm and large pale nuclei. There is bleeding through the granulosa into the central cavity.

Microphotograph 4 shows a corpus luteum on 16th day of menstrual cycle or 9th day after injection. Both the granulosa and theca layers are thin. The cytoplasm in the granulosa cells is thin, the blood vessels are poorly developed. There is very little blood in the cavity.

Microphotograph 5 shows a normal corpus luteum probably of 24 hours. There is considerable perifollicular haemorrhage and the blood vessels are markedly engorged. There is bleeding in the central cavity.

TABLE II  
Shows the gross appearance of the ovaries as seen during laparotomy on various days of menstrual cycle and various days of injection

Days of menstrual cycle	Days of injection.	Gross appearance of the ovaries				
		General appearance		Size of corpus luteum		
		Size.	Visibility on the surface of ovary.	Presence of dark reddish mound on the surface of the ovary.	Festooning of the corpus luteum into cavity filled with haemorrhagic fluid.	
15-23 24-26	9-14 18-20	Normal Small	Normal Small	Poor Poor	Absent Absent	Poor, small amount of fluid. Absent. Small amount of haemorrhagic fluid.
26-30 47-57	21-25 40-50	Small Small	Small Very small.	Not visible. Not visible.	Absent Absent	Absent, very small amount. Absent, negligible.
47-57 63-97	40-50 56-90	Small Very small.	Very small. Very small.	Not visible. Not visible.	Absent Absent	Absent, negligible. Absent, hardly any.
63-97	56-90	Very small.	Very small.	Not visible.	Absent.	Absent, hardly any.

Though the ovulation took place in 43 out of 50 cases, even on naked eye appearance, the majority of corpora lutea are abnormal. They are smaller than normal and hardly visible on the surface of the ovary except as a patch of yellow fibrous tissue. In none of the cases it projects on the surface as a reddish mound even in young and mature corpora lutea. On cut section there is hardly any blood-filled central cavity. The cavity is filled with small quantity of sero-sanguinous fluid. There is no festooning of the yellow corpus luteal wall into the blood-filled central cavity in any of these cases. So on naked eye appearance, a corpus luteum of 15th to 20th day resembled a degenerated corpus luteum of the day one or two of the last cycle.

TABLE III  
Microscopic appearance of the ovaries in 43 cases where corpus luteum was present

Days of menstrual cycle.	Days of injection.	Microscopic appearance of the ovary					
		Fibrosis in the ovarian cortex.	Vascularity	Corpus luteum.			Vascularity in granulosa cells.
				Central cavity filled with haemorrhagic fluid.	Granulosa cell layer developing into luteal cells.	Theca cells.	
15-24	9-14	+	Diminished.	Small amount	Diminished luteinizing thin layer.	Degenerating nuclei, vacuoles present.	Poor.
24-26	18-20	+	Diminished.	Very little	Thin cytoplasm, thin layer.	Thin layer, degenerating nuclei.	Poor.
26-30 47-57	21-25 40-50	++ ++	Poor Very poor.	Very little Very little fluid and blood.	Poor luteinization. Thin layer, minimal peri-follicular haemorrhage.	Poorly developed. Thin layer.	Very poor. Very poor.
63-97	56-90	++	Very poor	Negligible.	Vacuoles present in the luteal cells.	Very poorly developed.	Very poor.

Table III shows that in histological appearance the corpus luteum is very abnormal. The changes are increased fibrosis, poorly developed granulosa cell and theca cell layers. Evidence of luteinization is poor. Vacuoles appeared very early in the granulosa cell layer in very young corpora lutea. Vascularity and peri-follicular haemorrhage from delivery into the central cavity which are evidences of luteinization is negligible.

Microphotograph 6 shows a corpus luteum 37 days after injection or on 44th day of menstrual cycle. This corpus luteum should be the early corpus luteum of the present cycle. Vascularisation is absent. There is very little blood in the central cavity. Perifollicular haemorrhage is minimal.

#### Discussion

In 10 cases who had been operated in the first month, there was a corpus luteum in all the 10 cases. In the second and third month of treatment in 40 cases, corpus luteum was present in 34 cases. The corpus luteum was fresh in all the cases. There were plenty of corpora albicantia indicating that ovulation had occurred in recent cycles. This shows that the anti-ovulatory property of injectable nor-ethisterone enanthate is very weak.

Sanchez Rivera *et al* (1968) have shown that in 5 cases with the administration of gestagen, a pure progesterone preparation in cyclical fashion, free corpus luteum was present in three cases.

The analysis of corpora lutea of treated patients after various days of injection in different phases of menstrual cycle shows that the vascularization and luteinization are very poorly developed. In gross appearance these abnormal corpora lutea are much smaller than the normal ones. They do not project on the surface of the ovary like reddish mounds. On cut section there is no yellow festooning. Histologically there is marked

deviation from the normal corpus luteum on various days of menstruation. There is poor growth of the granulosa cell layer and poor luteinization. Degenerative changes and early vacuolisation take place in the luteal cells.

From the analysis of the results it seems that nor-ethisterone enanthate did not inhibit ovulation in the majority of the cases. The corpus luteum when formed shows degenerative changes at the early phase. There is thinning of both the granulosa and the luteal layers, irregular luteinization, poor vascularisation, diminution in the amount of cytoplasm of granulosa cells and diminution in the luteinization process.

It is obvious from this study that the fertility control by nor-ethisterone enanthate is not by inhibition of ovulation, but by its action mostly on the endometrium and the cervical mucus. If, however, the ovum is fertilized, the poor corpus luteum formation and its early degeneration interfere with nidation. In addition, the lack of development of granulosa cells and atresia of granulosa layer interferes with endometrial proliferation and maturation.

Therefore, we suggest that incomplete follicular development interference prevents conception.

#### Reference

1. Sanchez Rivera, G.: Merlo, J. G., Escudero, M. and Botella-Llusia, J.: *Am. J. Obst. & Gynec.* Vol. 101: 665, 1968.

*See Figs. on Art Paper V.*