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Original Article

RELATIVE ABUNDANCE OF CELL ORGANELLES IN STEROIDOGENIC CELLS OF CORPUS LUTEUM OF GOAT IN DIFFERENT REPRODUCTIVE PHASES

BATRA, S.[1] ? AND SHARMA, R. K.[2]

[1] Department of Zoology, University College and [2]Department of Zoology, Kurukshetra University, Kurukshetra 136119. Haryana, E mail: soniabatra2dec@rediffmail.com

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Abstract: Variations in relative abundance of different cell organelles of steroidogenic cells from seven categories of goat corpus luteum viz. small (1-5 days), medium (6-10 days), large (11-15 days), regressing (16-21 days), previous cycle, penultimate cycle and pregnancy (<30 days), have been analysed. In granulosa luteal cells, smooth endoplasmic reticulum variations were not prominent. Rough endoplasmic reticulum increased from small to large category and was most abundant in corpus luteum of pregnancy. The number of mitochondria, Golgi complex and secretory granules increased from small to large category. The maximum frequency was observed during the pregnancy. Vesicles were prominent in the previous and penultimate categories. The numbers of lipid droplets were maximum in corpus luteum of the previous cycle followed by the regressing category while lysosomes out numbered in corpus luteum of the penultimate cycle. In theca luteal cells, smooth endoplasmic reticulum and number of mitochondria increased from the small to large category and maximum numbers were observed during the pregnancy. Golgi complexes were abundant in corpora lutea of the large category and that of the pregnancy while vesicles showed their preponderance in corpus luteum of the penultimate category. Numbers of lipid droplets were more in corpus luteum of the previous cycle. Lysosomes were abundant in number in theca luteal cells of corpus luteum of the regressing category. Secretory granules were higher in number in corpora lutea of the large and pregnancy categories. These variations in relative abundance of cell organelles in steroidogenic cells shall be discussed in relation to their physiological obligations and endocrine profile at specific stage of reproductive cycle.

Key words: Corpus luteum, Cell organelles, Goat ovary

INTRODUCTION

The corpus luteum is a transient ovarian endocrine gland formed from the wall of the Graafian follicle after the release of the ova. It is a dynamic endocrine gland showing variations in its size, structure and steroidogenic activities during different phases of the estrous cycle and pregnancy [1]. In mammals, corpus luteum consists of steroidogenic cells i.e. large granulosa and small theca luteal cells and non steroidogenic cells i.e. endothelial cells, pericytes, smooth muscle cells, fibrocytes, macrophages, leucocytes and occasional plasma cells which show variations in their numbers [1-4]. Corpus luteum secretes progesterone as the primary endocrine secretory product [3,5]. In addition to this, corpus luteum also secretes prostaglandins, oestradiol 17 b and variety of proteins and peptide hormones like oxytocin, oxytocin related neurophysin 1, vasopressin, relaxin and inhibin [6]. Variations in the steroidogenic and non-steroidogenic activities of corpus luteum have been recorded in large and small ruminants [1,7]. Till date systematic correlative investigation has been on variations in fine structure of corpus

luteum of goat vis-à-vis its functions. Keeping in view, this lacunae in literature, present investigation was undertaken to analyse the relative abundance of cell organelles in small and large steroidogenic cells of goat corpus luteum in different phases of reproduction.

MATERIALS AND METHODS

Goat (*Capra hircus*) ovaries were procured from the slaughter house of Delhi ($28^0$38'N, $77^0$12'E) and brought to laboratory at 0^0C. Corpora lutea were dissected out and classified into seven categories on the basis of size, colour and texture (Table 1 Figs. 1,2). The corpora lutea of all the seven categories were subjected to ultrastructural studies.

For ultrastructural studies, corpus luteum was fIxed in Karnovsky fixative in 0.1M phosphate buffer (pH 7.2 to 7.4) at 4°C for 24 hours. The tissue was processed [8]. The sections were cut at 60-90nm mounted on 100 mesh grids and were stained with uranyl acetate followed by lead citrate [9].The sections were examined and photographed under electron microscope, CM-10 Philips installed at All India Institute of Medical Sciences, New Delhi.

RESULTS AND DISCUSSION

In large granulosa luteal cells the smooth endoplasmic reticulum variations were not prominent as these cells are primarily the protein synthesizing cells (Table 2). The maximum abundance of rough endoplasmic reticulum was observed in the large category followed by the pregnancy. However, the minimum occurrence of rough endoplasmic reticulum was recorded in the penultimate category (Table 2). The mitochondria were oval in shape in corpus luteum of the small category; oval to elongated in the medium category; spherical to oval shaped in the large category and swollen to vacuolated in the regressing category (Table 2). The corpus luteum of the previous and penultimate showed the presence of oval and swollen mitochondria respectively. Spherical shaped mitochondria with crecentric cristae were observed in corpus luteum of the pregnancy. The mitochondria number was maximum during the pregnancy followed by the large category. The minimum number pf mitochondria were recorded in the penultimate category (Table 2). The Golgi complexes were maximum in abundance in corpus luteum of the pregnancy and minimum in the small and penultimate categories of corpus luteum (Table 2). The numbers of vesicles were frequent in the previous and penultimate categories and minimum in the small, medium and pregnancy categories (Table 2). A large number of lipid droplets were observed in the previous category followed by regressing category. Whereas the minimum number of lipid droplets were observed in the large category (Table 2). The maximum numbers of lysosomes were observed in the penultimate category followed by the previous category. The minimum frequency of lysosomes was observed in the medium category whereas only a negligible numbers of lysosomes were seen in corpus luteum of the pregnancy (Table 2). A large number of secretory granules were observed in corpus luteum of the pregnancy followed by the large category. A small number of secretory granules were observed in the previous category (Table 2). The secretory granules were more concentrated towards the peripheral cytoplasm in corpus luteum of the medium category (Fig.3). In small theca luteal cells, smooth endoplasmic reticulum was maximum in the pregnancy and the minimum in the penultimate

Explanation of figures

Figs. 1 and 2: Seven categories of goat corpus luteum viz. I small (1-5 days), II medium (6-10 days), III large (11-15 days), IV regressing (16-21 days), V previous cycle, VI penultimate cycle and VII pregnancy (<30 days), showing different sizes colour and texture.

Fig. 3: Fine morphology of granulosa luteal cell from medium category of corpus luteum (6-10 days) revealing indented nucleus(nu), numerous elongated to oval shaped mitochondria(m), Golgi complex. Note the concentration of the secretory granules(s) towards the peripheral cytoplasm. X 10080

Fig. 4: A magnified view of theca luteal cell from large category of corpus luteum (11-15 days) revealing abundant smooth endoplasmic reticulum, numerous spherical mitochondria (mt) with fully distended swollen cristae and lipid droplets (l).X48300

Fig. 5: Ultraphotograph showing fine morphology of theca luteal cell from 11-15 days of corpus luteum revealing the presence of lysosomes (ls), and microtubules (mt). X26460

Fig. 6: Electronmicrophotograph revealing fine morphology of theca luteal cell of corpus luteum of previous cycle showing spherical to elongated mitochondria(m), vacuoles of varied sizes(v),increased number of lipid droplets and a few secretory granules (s). X 8610

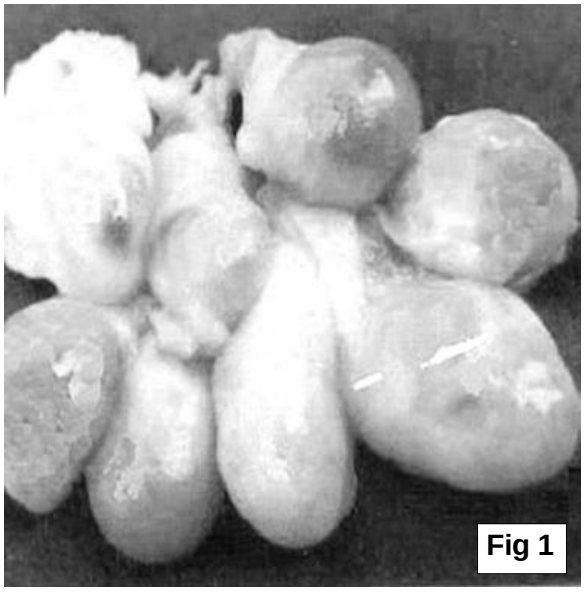
Fig 1

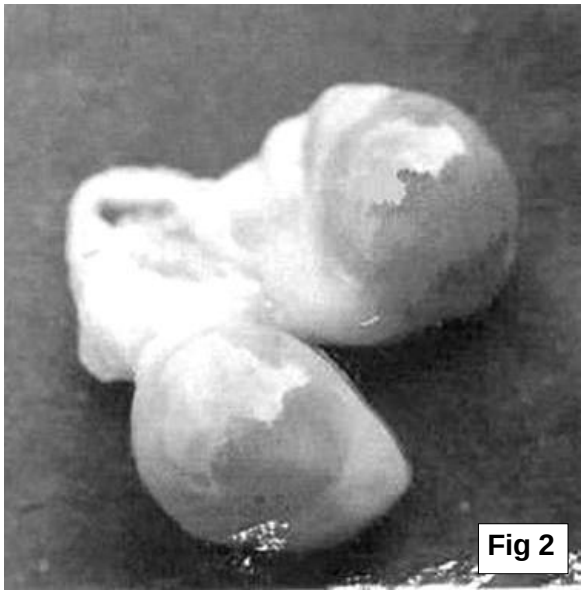
Fig 2

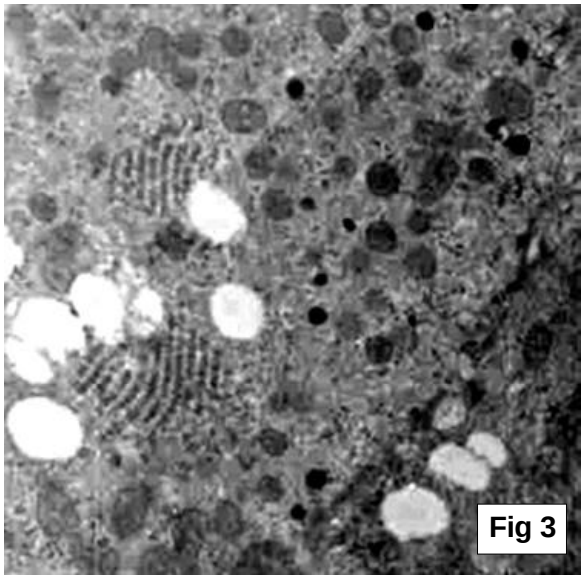
Fig 3

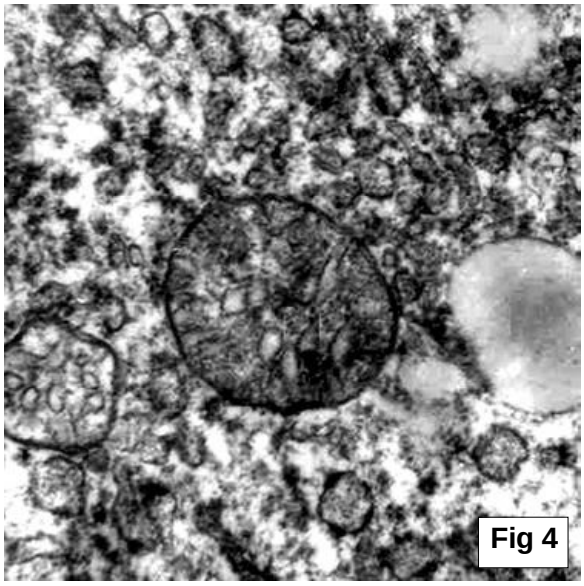
Fig 4

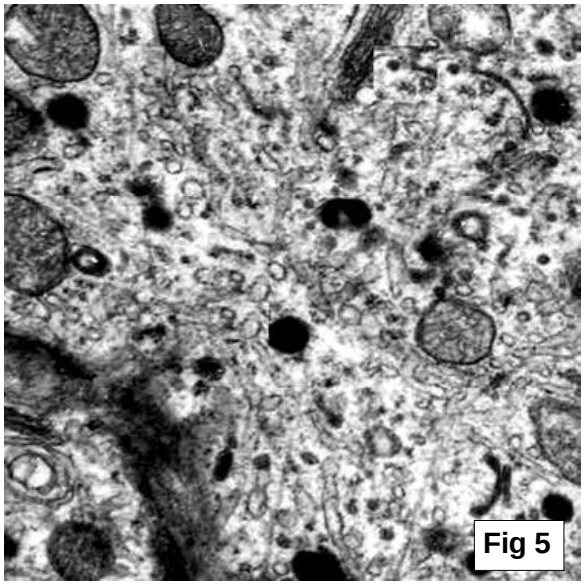
Fig 5

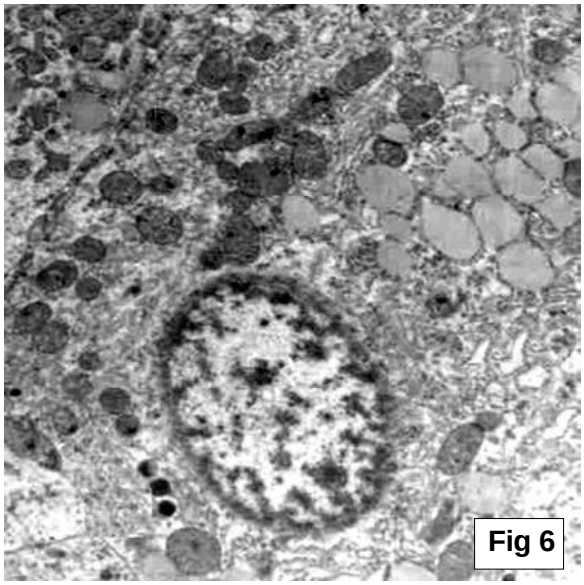
Fig 6

Table 1: Classification of Corpus luteum

Sr.No.	Size (mm) and colour	Name of category		Stage
1.	<2 Pink with red blood clot	Small	I	1-5 days
2.	2-5 Pink	Medium	II	6-10 days
3.	>5 Red	Large	III	11-15 days
4.	2-4 Brown	Regressing	IV	16-21 days
5.	≈ 2 Yellow	Previous	V	Pr
6.	<2 White	Penultimate	VI	Pn
7.	> 6 Dark Red	Pregnancy	VII	< 30 days

Table 2: Variation in the distribution of cell organelles in Granulosa Luteal cells of different categories of corpus luteum. + Minimum, ++ Less frequent, +++Frequent, ++++Abundant, +++++ Maximum (covers ± = .10%, + = 15% , ++ =30%, +++ = 45%, ++++ = 60%, +++++ = 75% of cytoplasm)

Cells Organelles	I Small (1-5 days)	II Medium (6-10 days)	III Large (11-15days)	IV Regressing (16-21 days)	V Previous (Pr)	VI Penultimate (Pn)	PregnancyVIIth (< 30 days)
Smooth endoplasmic reticulum	+	+	+	+	+	+	+
Rough endoplasmic reticulum	++	+++	+++++	++	+++	+	++++
Mitochondria	++ Oval	+++ Oval and elongated	++++ Spherical, oval	++ Swollen vacuolated	++ Oval	+ Swollen shaped	+++++ Spherical, oval
Golgi complex	+	++	++++	+++	+++	+	+++++
Vesicle	+	+	+	++	+++	+++	+
Lipid droplets	++	++	+	++++	+++++	Not observed	++
Lysosomes	++	+	+	+++	++++	+++++	–
Secretory granules	++	+++	++++	++	+	+	+++++

Table 3: Variation in the distribution of cell organelles in Theca Luteal Cells of different categories of corpus luteum of goat. + Minimum, ++ Less frequent, +++ Frequent,++++ Abundant,+++++ Maximum (covers ± = .10%, + = 15% , ++ = 30%, +++ = 45%, ++++ = 60%, +++++ = 75% of cytoplasm)

Cells Organelles	I Small (1-5 days)	II Medium (6-10 days)	III Large (11-15days)	IV Regressing (16-21 days)	V Previous (Pr)	VI Penultimate (Pn)	VII Pregnancy (< 30 days)
Smooth endoplasmic reticulum	++	+++	++++	++	++	+	+++++
Rough endoplasmic reticulum	+	+	–	+	+	+	–
Mitochondria	++	+++	++++	+++	++	+	+++++
Golgi complex	+	++	+++	++	+	+	+++
Vesicle	+	–	–	+	++	+++	++
Lipid droplets	+++	+	+	++	+++	++	++
Lysosomes	++	–	+	++++	+++	++	++
Secretory granules	+	+++	++++	++	++	+	++++

category (Table 3). The mitochondria, Golgi complex, secretory vesicles, lysosomes and lipid droplets revealed a trend similar to the large luteal cells (Table 3; Figs. 4,5,6).

Ultrastructurally, the presence of rough endoplasmic reticulum and smooth endoplasmic reticulum in granulosa luteal cells indicate that these cells have exceptional steroidogenic and protein synthesizing potential as reported in sheep and other bovine species [1,2,4,10,11]. The relative abundance of mitochondria in large and small luteal cells increased from category I (1-5 days) to category III (11-15 days) and decreased thereafter. In corpus luteum of the pregnancy the mitochondria were in maximum abundance. The increased abundance of mitochondria, is an index of increased steroidogenic activities [1,4,7,12]. Variation recorded in the shape and size of mitochondria during different stages of oestrous cycle and pregnancy indicate the metabolic status of the cell. The more secretory cells have a tendency to harbor more number of mitochondria with higher number of cristae to lodge greater quantities of mitochondrial enzymes [13-16]. The corpora lutea of 11-15 days and that of the pregnancy revealed higher preponderance of Golgi complexes and

associated secretory activities in both types of the steroidogenic cells. The large luteal cells had higher frequency of Golgi complexes than the small luteal cells. It is possibly because of different functional obligations of these two cell populations. The large luteal cells are good producers of peptide hormones or secretions while smaller cell population is largely steroidogenic in function. Similar structural and functional relationships have already been documented in the steroid hormone synthesizing cells of a number of domestic animals [13,14,16-18]. The relative frequency of vesicles and lipid droplets in large luteal cells declined from category I (1-5 days) to category III (11-15 days) and increased in the regressing and the previous category. In corpus luteum of pregnancy number of vesicles and lipid droplets were comparable to those of the category I. The variations recorded in the vesicle abundance and lipid droplets revealed a negative correlation with steroidogenic out put. In bovine corpus luteum very little lipids were observed on day 10 to 13, their amount increased during regression [13,14,19,20]. During the luteal phase and early pregnancy the small luteal cells showed minimum to less frequent number of lipid droplets, this may be because the cholesterol and its esters were utilized during active synthesis of progesterone [15,23]. The increase in the lipid droplets and vesicles during later phase of the estrous cycle is because of poor mobilization of lipids, and decline in progesterone synthesis [14,20,22,23].

The lysosomes were maximum in corpora lutea of the penultimate cycle while minimum in category III (11-15 days). The reverse pattern was revealed by the secretory vesicles. The maximum numbers of secretory vesicles were reported in corpus luteum of pregnancy. These secretory bodies were designated as dense luteal bodies in cow [24]. These granules were renamed as secretory granules [25].

During pregnancy and oestrous cycle (15 to 21 days), development of diffuse lipoproteins is correlated to the highest rates of conversion of acetate-1-14C into progesterone *in vitro* [3,20,26-28]. Membranes of agranular endoplasmic reticulum besides acting as a site for enzymes involved in the biosynthesis of steroids, also accumulate and store as a constituent of their lipid component like cholesterol which act as a precursor for the biosynthesis of steroids [29-31].

In the regressing steroidogenic cells, number of lipid droplets and lysosomes increased. The presence of lysosomes is positively correlated to the appearance of autophagocytotic bodies with the declining level of progesterone as observed in most of the bovine species [32-34].

Thus it is evident from the present studies that corpus luteum of goat shares most of its structural organization and variations therein with other bovine species suggesting their close evolutionary relatedness and a common regulatory mechanism.

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