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# USE OF ELECTRON MICROSCOPY IN STUDY OF STEROIDOGENIC CELLS OF NORMAL, REGRESSING AND PREGNANT CAPRINE CORPORA LUTEA

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**ABSTRACT:** Electron microscopic studies revealed variations in different cell organelles in steroidogenic cells of normal, regressing and pregnant caprine corpora lutea. Corpus luteum was comprised of two types of steroidogenic cells *viz.* small theca luteal cells and large granulosa luteal cells. In granulosa luteal cells, rough endoplasmic reticulum was most abundant in corpus luteum of pregnancy. The number of mitochondria, Golgi complex and secretory granules increased from normal to pregnancy category and decreased in regressing category. In theca luteal cells, smooth endoplasmic reticulum, number of mitochondria and Golgi complex were more in pregnancy. During regression, degenerating luteal cells with shrunken cytoplasm, dispersed nuclear membrane, apoptotic granules, increased number of lipid droplets, lysosomes, swollen mitochondria, irregular plasma membrane, disintegrating and vacuolated rough endoplasmic reticulum were observed. The degenerating luteal cells of regressing corpus luteum revealed the patches of chromatin material along the periphery of the uneven nuclear membrane and also within the nucleoplasm. These variations shall be discussed in relation to their physiological obligations.

**INTRODUCTION:** The corpus luteum is a transient ovarian endocrine gland formed by postovulatory follicle wall. It secretes progesterone as primary endocrine secretory product [1-5]. In addition to this, corpus luteum also secretes prostaglandins, Oestradiol 17 β, and a variety of proteins and peptide hormones [6]. In case pregnancy fails, corpus luteum regresses and triggers the development of follicles and estrous cyclicity leading to ovulation [7, 8]. Prostaglandins induce this regression through the constriction of blood vessels and by apoptosis [9-11].

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As corpus luteum is dynamic in nature, therefore, it is used for the study of diverse cellular processes that occur in it *viz.* cell proliferation and apoptosis, immune system activation, downregulation of functional proteins and cellular matrix remodeling events [12]. The various kinds of growth factors are the key regulators of corpus luteum formation, function and regression [13, 14].

Although, morphology (ultrastructure) of corpus luteum of rodents has been carried out [15]. On the contrary, in the caprine, a little information on the morphology of corpus luteum of the luteal phase of the estrous cycle has been recorded [7, 16]. Keeping in view, the lacunae in caprine corpora lutea literature, the present investigation was undertaken to describe the electron microscopic study of steroidogenic cells of normal, regressing and pregnant caprine corpora lutea.

**MATERIALS AND METHODS:** Goat (*Capra hircus*) ovaries were procured from the slaughterhouse (Registration number APEDA/24) of Delhi and brought to the laboratory in an ice bucket at 0 °C. Corpora lutea were dissected out and classified into seven categories on the basis of size, color and texture.

Out of seven categories, corpora lutea of normal, regressing, and pregnancy categories were separated. For electron microscopic studies, corpus luteum was fixed in Karnovsky fixative in 0.1 M phosphate buffer (PH 7.2 to 7.4) at 4 °C for 24 h. The tissue was processed [17]. The sections were cut at 60-90 nm thickness with the help of glass knives and were mounted on 100 mesh grids. The thin sections were stained with uranyl acetate followed by lead citrate [18]. The sections were examined and photographed under an electron microscope, CM-10 Philips installed at All India Institute of Medical Sciences, New Delhi.

**RESULTS AND DISCUSSION:** The transmission electron microscopic analysis of caprine corpora lutea revealed the presence of two dominant cell types that is large granulosa luteal cells and small theca luteal cells. The small theca luteal cells of normal (large category) corpus luteum showed oval-shaped nucleus with patches of electron-dense nuclear material in the nucleosol **Fig. 1**, the narrow and elongated stacks of Golgi complexes and their tips revealed the stages of pinching off the secretory granules **Fig. 2**.

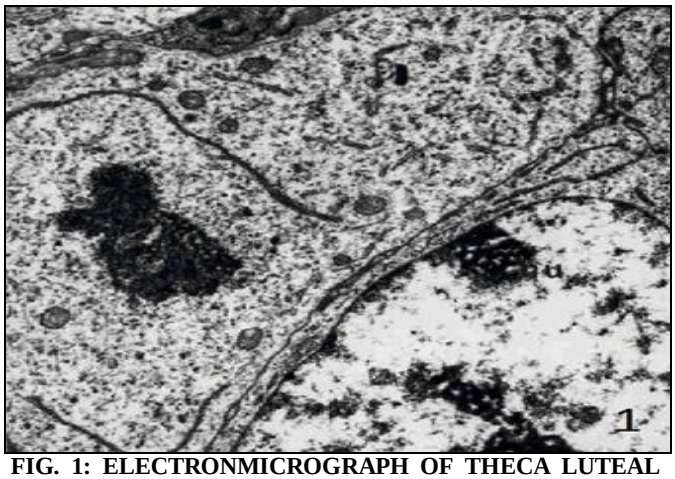


**FIG. 1: ELECTRONMICROGRAPH OF THECA LUTEAL CELL OF LARGE CATEGORY (11-15 DAYS) OF CORPUS LUTEUM SHOWING OVAL SHAPED NUCLEUS WITH PATCHES OF ELECTRON DENSE NUCLEOLAR MATERIAL (NU) IN THE NUCLEOSOL. X 12180**

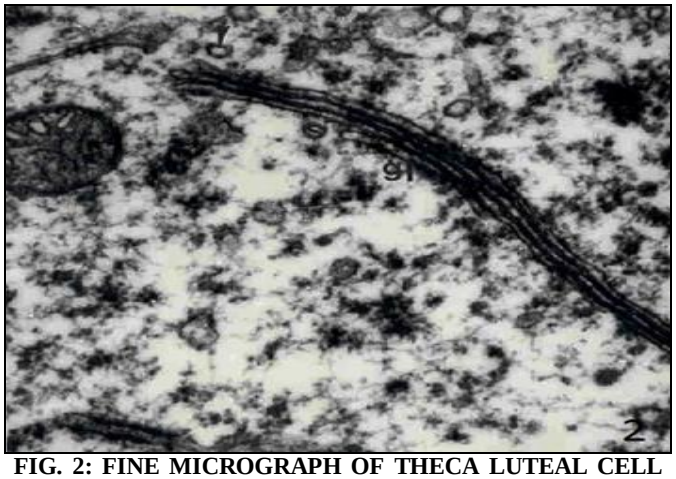


**FIG. 2: FINE MICROGRAPH OF THECA LUTEAL CELL FROM 11-15 DAYS OF CORPUS LUTEUM EXHIBITING NARROW AND ELONGATED STACKS OF GOLGI COMPLEXES (GL). NOTE THE TIPS OF THE GOLGI COMPLEXES REVEALING THE STAGES OF PINCHING OFF THE SECRETORY VESICLES (S). X 56700**

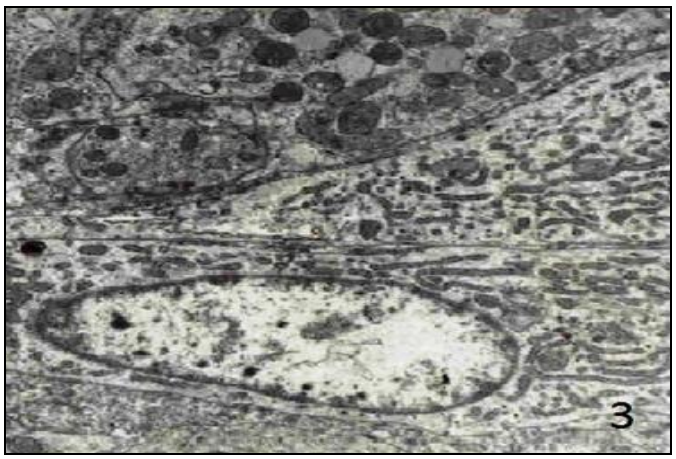


**FIG. 3: FINE STRUCTURE OF GRANULOSA LUTEAL CELL OF CORPUS LUTEUM OF REGRESSING CATEGORY (16-21 DAYS) OF CORPUS LUTEUM SHOWING A VERY FEW OVAL SHAPED MITOCHONDRIA. X 12180**

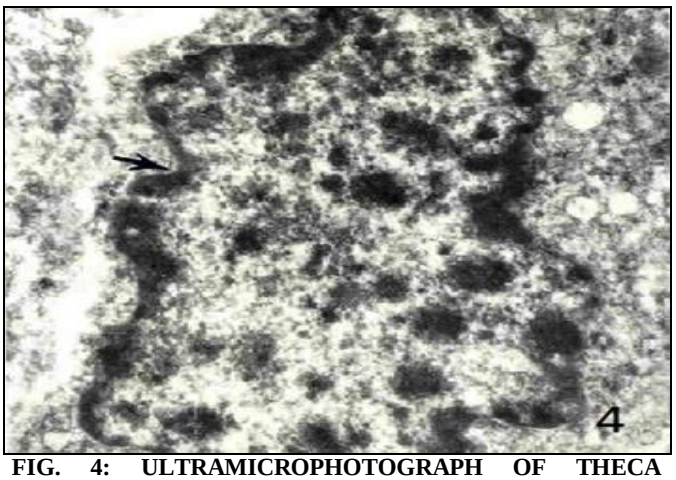


**FIG. 4: ULTRAMICROPHOTOGRAPH OF THECA LUTEAL CELL FROM 18-21 DAYS OF CORPUS LUTEUM REVEALING INDENTATION OF THE NUCLEAR MEMBRANE AND PYCNOTIC NUCLEUS. X 24570**

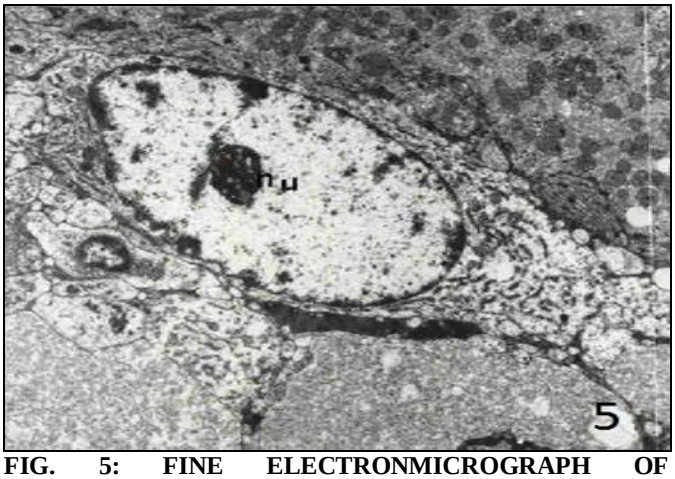


**FIG. 5: FINE ELECTRONMICROGRAPH OF GRANULOSA LUTEAL CELL OF CORPUS LUTEUM OF PREGNANCY SHOWING CONDENSED NUCLEOLUS (NU), ROUGH ENDOPLASMIC RETICULUM (RER), SMOOTH ENDOPLASMIC RETICULUM (SER) AND MITOCHONDRIA. X 8190**

In large granulosa luteal cell of the regressing category (16-21 days), a very few oval-shaped mitochondria, degenerated stacks of the swollen rough endoplasmic reticulum were observed **Fig. 3** whereas in small theca luteal cell of the regressing category (16-21 days), the patches of chromatin material along the periphery of the uneven nuclear membrane and also within the nucleoplasm were observed. Increased number of lipid droplets was its prominent feature **Fig. 4**. The corpus luteum of pregnancy was characterized by condensed nucleolus, rough endoplasmic reticulum, smooth endoplasmic reticulum, and mitochondria **Fig. 5**.

During the luteal phase and early pregnancy, theca luteal cells revealed minimum to a less frequent number of lipid droplets. It is because the cholesterol acts as a precursor for steroidogenesis [19]. During regression, an increase in the number of lipid droplets is because of poor mobilization of lipids and a decline in progesterone synthesis [15, 20-22]. In regressing steroidogenic cells, an increase in a number of lysosomes is positively correlated to the appearance of autophagocytotic bodies with the declining level of progesterone as documented in most of the bovine species [23-25]. The number of mitochondria decreased from normal to regressing corpora lutea. In corpus luteum of pregnancy, the mitochondria were maximum which indicates increased steroid biosynthesis [26-28].

**CONCLUSION:** The steroidogenic cells of regressing corpus luteum of goat revealed apoptotic specific degenerating features. The increased in the number of lipid droplets correspond to the decreased level of progesterone whereas during pregnancy maximum number of mitochondria depict maximum synthesis of progesterone.

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**CONFLICTS OF INTEREST:** The author declares that there is no conflict of interest.

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