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HISTOENZYMIC STUDY OF THE SUPEROVULATED OVARIES OF ACYCLIC ASSAM LOCAL GOATS(*CAPRA HIRCUS*)

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ABSTRACT

A study on histoenzymic activity superovulated ovaries of confirmed acyclic Assam Local goats of Assam treated with pregnant mare serum gonadotropin (PMSG), human chorionic gonadotropin(HCG) after priming with .6- -Methyl-17-Acetoxy progesterone (MAP) for 11 days. After confirmation of superovulation by laparotomy both right and left ovaries were collewcted, frozen and sectioned with cryostat microtome. The sections were treated for histoenzymic localization of various phosphatases, dehydrogenases, and cytochrome oxidase. In response to to the superovulatry treatment combination the steroid secreting cells of the superovulated acyclic ovaries mostly theca interna, granulose cells of follicles exhibited typical histoenzymic reactivity and significance of localization had been discussed.

Key words: acyclic Asom Local goat, histoenzymic, superovulation

INTRODUCTION

Multiple ovulation and embryo transfer(MOET) have most been widely used for faster augmentation of livestock production where response to different agents causing superovulation is well understood. Assam Local goats. Assam Local goat has a trend of seasonal acyclicity. Available literature revealed success of oestrus induction (Goswami, 1992) and superovulation

of these type of goats(Bonia, 1992). But information on histoenzymic reactivity and their significance of localization in Assam Local goat is meager. So, the present investigation has been undertaken to ascertain the histoenzymic reactivity in superovulated acyc;lic goat ovary in response to superovulatory treatment combinations of progesterone priming plus gonadotropins.

MATERIALS AND METGODS

Three confirmed healthy anoestrous nanny goats were considered for the present observation. They were primed with oral feeding of 5mg of 6- -Methyl-17-Acetoxy progesterone(MAP) for 11 days and on the day of MAP withdrawal 750IU of PMSG(Folligon) were injected intra muscularly and then a vasectomied buck was used to detect oestrus at 4 hours interval. At oestrus an injection of 750IU of HCG(Chorulon) was given intramuscularly. On the 6^{th} day after confirmed onset oestrus both the right and left superovulated ovaries were collected by mid-ventral incision of the treated goats and exploration. Subsequently, the ovaries were subjected cryostat sections of 10µ thickness maintained at -20^{o}C. These section were later treated with different histoenzymic methods of Chayan *et al.*(1973) and Pearse(1980) and incubated localization of of enzymes and demonstration of phosphatases viz. alkaline phosphatase(AK-pase), adenocine triphosphatase(ATP-ae) and glucose-6-phosphatase(G-6-pase); dehydrogenases viz. succinate (SDH), malate (MDH), lactase(LDH), glucose-6-phosphate dehydrogenate (G-6-PD), Δ^5-3-β-hydroxisteroid(Δ^5-3-β-HSD) and 17-β-hydroxisteroid (17-β-HSD) and cytochrome oxidase(cyo-ase). The stained histochemical sections were fixed in 15% normal saline solution, examined under microscope for gradation of histoenzymic reaction as below and reactivity described respectively.

\+ = Weak

++ = Moderate

+++ = Strong

++++ = Intense

\- = Negative

RESULTSAND DISCUSSION

The result of the present investigation (Table 2) revealed that the different types of phosphatases, dehydrogenates and oxidases were found to be concentrated mainly in theca-interna and granulose cells of folliclers and lutein cells of corpus luteum of superovulated

ovaries. It was revealed that G-6-PD reactivity in theca interna of growing follicles was comparativel higher (strong to intense) than the thec cells of atretic follicles (moderate to strong) of superovulated ovaies uniformly while granulos cells of growing follicles exhibited moderate to strong activities. Administration of MAP, allyestrenol and PMSG without giving HCG, it was decreased tend in theca and granulose cells mature (strong) and atretic (Moderate to strong) of unovulated ovaries. This histoenzymic reactivity different enzymes in different steroid secreting cells of goat ovary as had been identified were also seen in normal cyclic ovaries (Bhattaccharya, 1982). However, intensity of these enzymes was found to be varied as shown in Table 2 where relatively stronger activity was noticed in the lutein cells of corpus luteum followed by theca-interna and granulose cells. The nature of 17-β-HSD reaction with the distribution of positive formazan granules in all steroid secreting cells of the concerned ovaries was almost similar Δ^5-3-β-HSD with little variation in the intensity of the particular enzymic activities and the stronger histoenzymic reactivity in these cells might be due to the direct involvement of hydoxysteroid dehydrogenates such as Δ^5-3-β-HSD, 17-β-HSD and other enzymes such as G-6-PD, MDH so on meant for the triggering effect on the hexose monophosphate shunt or pentose phosphate pathway for supplying necessary co-factors for steroid synthesis in the ovary as reported by earlier authors (Baillic *et al.*, 1966; Pupkins *et al.*, 1966; Saidapur and Greenwald, 1978 and Bhattacharya, 1982). However, lower intensity of hostoenzymic reactivity in membrane granulose cells probably indicated that these cells were not directly involved in steroid synthesis. Bhattacharya and Saigal(1990) had shown the samer in goat ovary which had been confirmed in the present experiment.

It is to be stated that dehydrogenate activity in the lutein cells of corpus luteum of superovulated acyclic ovary of the present study was repreented by formation of formazan granules whch did not show the same perinuclear distribution as shown in the corpus luteum spurium or verum (Bhattacharya, 1992) where the formazan granules were irregularly distributed presumably due to altered metabolic activity.

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Table 1. Treatment schedule for superovulation of acyclic Assam Local goats

Treated groups	Number of goats	Treatment combination	Dose	Mode of administration
I	2	MAP	5mg	Orally for 13 days
		PMSG	750IU	i/mly, Single injection 24 hours after last MAP feeding
		HCG	1000IU	i/mly, Single injection during oestrus just before or after breeding
II	2	Allylestrenol	5mg	Orally for 13 days
		PMSG	750IU	i/mly, Single injection 24 hours after last MAP feeding
		HCG	1000IU	i/mly, Single injection during oestrus just before or after breeding
III		MAP	5mg	Orally for 13 days
		PMSG	750IU	i/mly, Single injection 24 hours after last MAP feeding
		HCG	750IU	i/mly, Single injection during oestrus just before or after breeding
IV		MAP	5mg	Orally for 13 days
		PMSG	750IU	i/mly, Single injection 24 hours after last MAP feeding
		HCG	-	-

Table 2. Histoenzymic activities of glucose-6-phosphate- dehydrogenases (G-6-PD) on the ovary of superovulated acyclic goats in response different treatment combination

Treatment Groups	Ovaries	Graaffian follicle		Atretic follicle		CL
		TIC	GC	TIC	GC	LC
I	2 pairs	++++	++	+++	+	++++
II	2 pairs	+++ to ++++	++	++ to +++	+ to ++	+++ to ++++
III	2 pairs	++++	++	-	-	++++
IV	2 pairs	+++ to ++++	+++	++	++	++++

Table 3. Histoenzymic activities of malate dehydrogenase (MDH) on the ovary of superovulated acyclic goats in response different treatment combination

Treatment Groups	Ovaries	Graaffian follicle		Atretic follicle		CL
		TIC	GC	TIC	GC	LC
I	2 pairs	++++	+++	-	-	++++
II	2 pairs	++++	+++ to ++++	++	++	++++
III	2 pairs	+++ to ++++	++	++	++	+++ to ++++
IV	2 pairs	+++ to ++++	+++	+++ to ++++	+++	+++ to ++++

Table 4. Histoenzymic activities of Δ^5-3-β-hydroxisteroid(Δ^5-3-β-HSD) on the ovary of superovulated acyclic goats in response different treatment combination

Treatment Groups	Ovaries	Graaffian follicle		Atretic follicle		CL
		TIC	GC	TIC	GC	LC
I	2 pairs	+++ to ++++	+++	++	++	++++
II	2 pairs	++++	++ to +++	++	+	+++
III	2 pairs	+++	++	+	+	+++ to ++++
IV	2 pairs	++ to +++	+++	++ to +++	++	No CL