SUBINVOLUTION OF PLACENTAL SITES – ITS DIAGNOSIS AND TREATMENT IN A LABRADOR DOG

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A 2½ year old female Labrador was presented with a history of vaginal bleeding for the last three days. The animal delivered nine live pups 50 days back, normal puerperal discharge subsided and metrorrhagia started from day 48 postpartum onwards. Blood loss increased day by day and the animal became anaemic. Ultrasonography revealed slightly distended uterus and vaginal smear showed binucleate trophoblast like cells. Ovariohysterectomy was performed and treated the animal using plasma volume extenders and other supportive medications, which lead to an uneventful recovery. Impression smears from placental sites revealed binucleate, heavily vacuolated trophoblast like cells. Histopathology showed dilated endometrial glands, surrounded by collagen. Trophoblast like cells were seen beneath the collagen layer, myometrium and around the blood vessels. Diagnostic and therapeutic measures adopted in a case of SIPS were described.
Keywords: Subinvolution, Placenta, Trophoblast like cells, Dog.

Puerperium is the period after parturition during which the reproductive organs revert back to the normal state both anatomically and physiologically. Normal lochial discharge which changes from greenish to brownish to mucoid lasts upto three weeks postpartum. Several postpartum conditions lead to persistent serosanguinous vaginal discharge beyond the normal duration like trauma of genital tract, metritis, endometrial hyperplasia, tumours of genital tract or urinary bladder, cystitis, coagulopathy, subinvolution of placental sites (SIPS) and brucellosis. SIPS is the exudation of serosanguinous, non-inflammatory vulvar discharge beyond the normal postpartum lochial period ie, more than three weeks postpartum. SIPS occurs when the uterine involution process is delayed. The amount of discharge ranges from a few drops each day that subsides without treatment to acute, life-threatening metrorrhagia that requires immediate blood transfusion and/ or surgery.

Case history and Observations

A 2½ year old female Labrador bitch, weighing 26 Kg, was presented to the clinics of the Department of Veterinary Gynaecology and Obstetrics, Veterinary College, Bengaluru with a history of vaginal bleeding for the last three days. This primiparous animal delivered nine live pups 50 days back and had the normal puerperal vaginal discharge for nearly 15 days, then it subsided. The pups were weaned by 40 to 45 days. The bitch showed a few drops of serosanguinous vaginal discharge on day 48 postpartum, which increased in quantity day by day and was presented to the clinic for treatment.

On clinical examination, the animal was found to be active with normal physiological parameters. Mucous membranes were slightly pale. There was spotting of fresh unclotted blood from the vagina. Per-vaginal examination couldn't reveal any tumour or other lesions. Ultrasonography revealed slightly distended uterus with anechoic areas anterior to the urinary bladder. Free fluid accumulation in abdominal cavity suggestive of ascites was also observed. Smears were collected from vaginal tract and stained with Giemsa's stain, which revealed mostly RBCs with few vaginal epithelial cells and binucleate trophoblast like cells.

Haematology revealed a total RBC count of 3.55 millions/µl, WBC count of 3000/µl, PCV 32 %, Haemoglobin 10.4 g/dl,

platelet count of 2,12,000/µl and serum creatinine and SGPT levels of 1.3 mg/dl and 58U, respectively.

Based on history, clinical findings, ultrasonography, haematology and cytological studies, the case was tentatively diagnosed as SIPS. Since the animal was anaemic and there was increased blood loss from vagina and the owner wanted to sterilize the animal, it was decided to perform ovariohysterectomy.

Treatment and Discussion

Ovariohysterectomy was performed as per standard surgical procedures under general anaesthesia. Animal was treated parenterally with antibiotics (Cefazoline @ 20 mg/Kg. body weight), analgesics (Tramadol @ 1 mg/Kg. body weight), plasma volume extenders (Hetastarch 6 % solution @ 10 ml/ Kg. body weight) and intravenous fluids (Ringer lactate @ 10 ml/ Kg. body weight) for the next three days. Oral antibiotic therapy and alternate day wound dressing was followed for next one week. The sutures were removed on the 10th postoperative day and the animal had an uneventful recovery.

On gross examination, the uterine horns were not fully involuted, distended and all placental attachment sites could be appreciated from the serosal surface. Two placental sites on left and five placental sites on right horns were hyperaemic, ellipsoidal, thin walled and more distended. Both ovaries contained regressing corpora lutea (Fig.-1). Uterine horns were partially filled with clotted blood. Placental sites were clearly visible, haemorrhagic with 10 to 12 greyish varying sized (2 to 4 mm) nodules spread randomly over each site (Fig.-2). Impression smears were collected from these placental sites, stained with Giemsa's stain, which revealed mostly binucleate, heavily vacuolated trophoblast like cells (Fig.-3). Representative tissue samples were collected from uterine horns, preserved in 10% buffered formol saline and processed for histopathological studies.

Fig.-1 Canine uterus showing haemorrhagic, with greyish nodules

Fig.-2 Canine uterus showing thickened endometrium ellipsoidal swellings on serosal surface

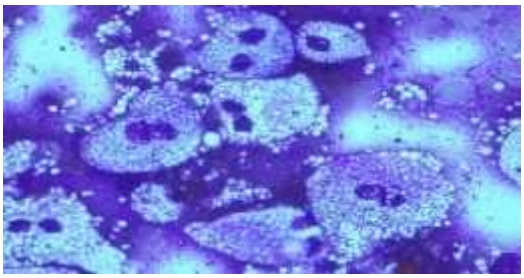

Fig.-3 Binucleated heavily vacuolated trophoblast like cells (400X)

The cause of subinvolution is not fully known, but this process may be a manifestation of an abnormal interaction between fetal-derived trophoblast and maternal tissue as also mentioned by Weydert and Benda, 2006. In vitro studies using

canine trophoblast cells showed that interleukin–8 (IL-8) increased cell migration by 35% and tissue inhibitor of metalloproteinase –2 (TIMP-2) decreased cell invasion by 57% as also recorded by Gullaba, 2013. Bitches showing SIPS are normal except for metrorrhagia for several weeks postpartum. The blood loss may range from a few drops per day that subsides without any treatment to life threatening metrorrhagia that requires blood transfusion and surgery. In the present case also, the blood loss increased day by day and the animal became moderately anaemic and developed ascites. Therefore ovariohysterectomy was preferred before the condition of the animal further deteriorated. The animal was stabilized with plasma volume extenders and fluids postoperatively.

Diagnosis can be made from the trans-cervical vaginal endoscopy to identify trophoblast persistence as also mentioned by Schlafer, 2012, trans-abdominal palpation of uterine swellings, which can be confirmed by radiography or ultrasonography. Detailed anatomical and histological studies of postpartum uterus indicated that normal involution of uterus in bitch can last upto three months simillarly reported by Orfanou *et al.,* 2008

Since spontaneous remission occurs, most of the cases may not require any treatment. Ovariohysterectomy may be indicated in severe blood loss or ulceration of endometrium/ myometrium leading to perforation and subsequent peritonitis Blood transfusion may be recommended in severely anemic cases. Hysterolaparotomy with curettage of selected placental sites and Ergonovine @ 0.2 mg/15 Kg. body weight as single dose intramuscularly Oral administration of low doses of progestagen for two weeks (Megestrol acetate, 0.1 mg/Kg. body weight once daily for the first week, then 0.05 mg/Kg. body weight once daily for the second week) was found effective in treating SIPS, with neither side effects nor subsequent reduced fertility as also mentioned by Voorhorst *et al.,* 2013. This indicated that supplementation of progesterone will help in the endometrial repair process. Spontaneous full recovery of a SIPS case in an American Staffordshire terrier as was reported by Sontas *et al.*, 2011. It was concluded that occurrence of SIPS does not affect the subsequent oestrus, fertility, gestation and parturition as also recorded by Voorhorst *et al.*, 2013 and the condition does not necessarily develop following subsequent pregnancies. Administration of a dose of oxytocin in postpartum bitches was suggested to prevent the occurrence of SIPS.

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