CLINICAL EVALUATION OF DIFFERENT TECHNIQUES OF OVARIOHYSTERECTOMY IN BITCHES

THESIS

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CERTIFICATE

Certified that **Dr. Anil Kumar Singh**, a candidate of M.V.Sc. (Final) examination of 2006 in **Veterinary Surgery & Radiology** has worked under my supervision during the session and that the accompanying thesis entitled, **"CLINICAL EVALUATION OF DIFFERENT TECHNIQUES OF OVARIOHYSTERECTOMY IN BITCHES"** which he is submitting is his genuine work.

B. Singh

(Bharat Singh)

Guide

DEDICATED

TO

MY PARENTS

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Writing of this manuscript signals the completion of another milestone in my academic journey. The almighty is the first, before whom I bow my head with great reverence because without his endless blessings no task can be accomplished.

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Anil Kumar Singh

(Dr. ANIL KUMAR SINGH)

ABBREVIATIONS

@	: At the rate of
b. Wt.	: Body weight
BWG	: British wire gaze
B.P.	: Bard parker
Cm	: Centimeter
Cu mm	: Cubic millimeter
DLC	: Differential leucocyte count
DMRT	:Duncan's Multiple Range Test
et al.	: et alii/alia
^{0} F	: Degree Fahrenheit
EDTA	: Etylenediamine tetra acetic acid
Fig	: Figure
G	: Gram
>	: Greater than
Hb	: Haemoglobin
HR	: Heart rate
hr	: Hour
i.e.	: id est. (that is)
i.m.	: Intra muscular
i.v.	: Intravenous
inj	: Injection
I.U	: International unit
kg	: Kilogram
mg	: Milligram
min	: Minutes
ml	: Milli liter
<	: Less than
No	: Number
nm	: Neno meter
OD	: Optical density
OH	: Ovariohysterectomy
PCV	: Packed cell volume
PGA	: Poly glycolic acid
%	: Percentage
/	: Per
RR	: Respiration rate
RT	: Rectal temperature
SE	: Standard error
TLC	: Total leucocyte count
TEC	: Total erythrocyte count
Viz.,	: Videlicet (that is)
WBC	: White blood cell
Wt	: Weight

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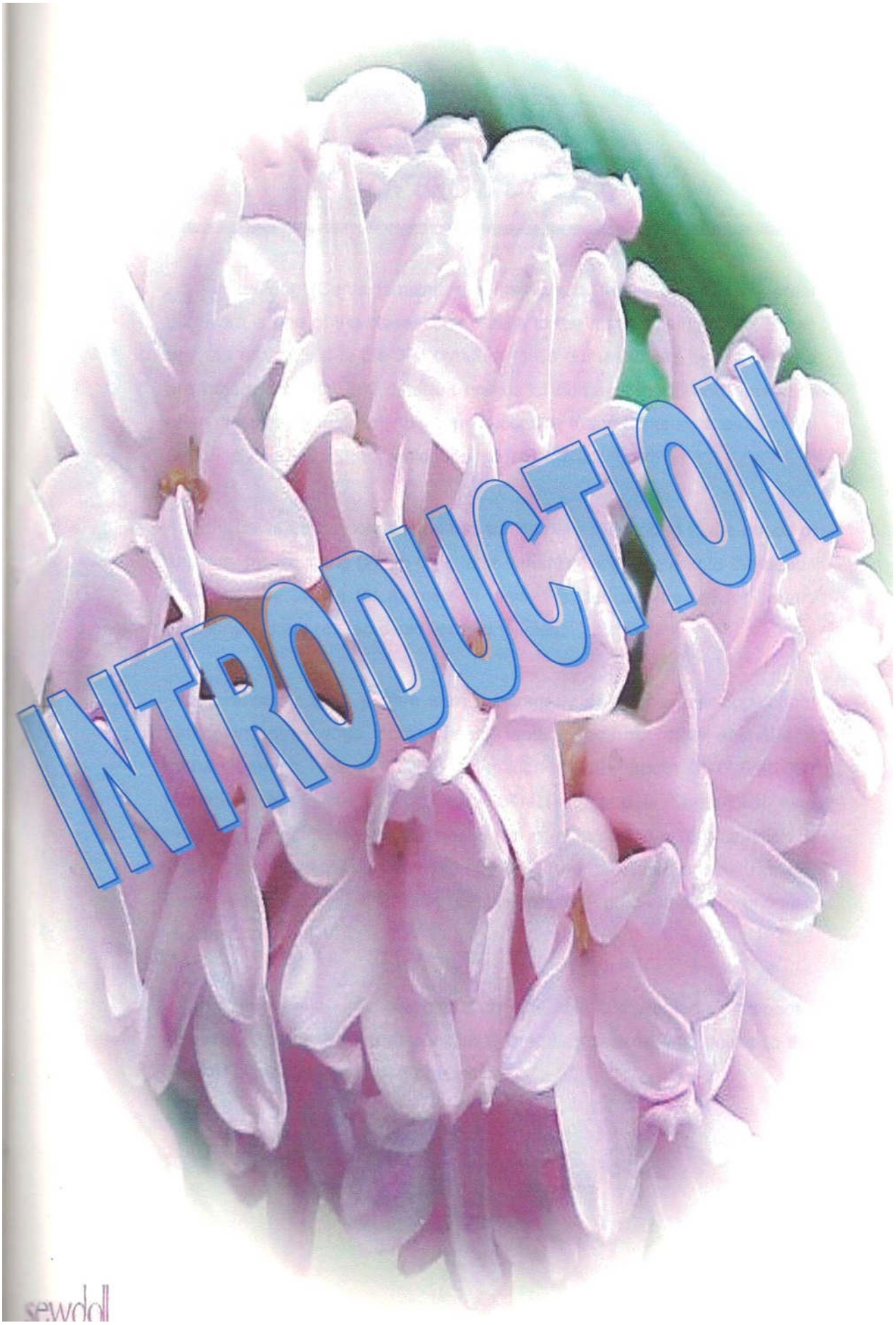

INTRODUCTION

CHAPTER -1

INTRODUCTION

The dog (Canis familiaris), a member of family canidae of order carnivora is one of the closest companion of the human beings since stone age, as probably it is the first creature to be domesticated by man. There are now more than 300 different breeds of domestic dogs and these are often classified into different groups according to the tasks such as hunting, guarding and herding, for which they were originally bred.

The veterinary profession has traditionally responded to the needs of animals, by providing health care for a variety of species, and to the needs of society by caring for animals in ways that reduce the risk of people acquiring animal-related diseases and injuries.

However India suffers from the tragedy of canine over-population, particularly stray dogs, posing health hazards both for other animal and human beings. As per WHO estimation (1999), about 35,000-40,000 human deaths occur annually due to rabies alone in South-East Asia region. High incidence of rabies occurred in India due to high population of stray dogs (about 25 million, most of them are unprotected against rabies). Every year about 3-4 million people receive post-exposure antirabic vaccination.

The prevalence of dog bite is considerable and problems of zoonotic diseases, aggressive behaviour during breeding season, etc. points to the need of controlling stray dog population.

So, keeping in view, the sterilization, i.e. spaying and neutering on a national scale in the form of Animal Birth Control (ABC) programme as directed by Animal Welfare Board, Chennai has been initiated by various Municipal Corporations, Canine Welfare Societies and various NGOs, for the same purposes.

Neutering is the surgical procedure whereby the reproductive organs are removed rendering the dog incapable of producing offsprings. In the female (bitch), the technique is referred to as ovariohysterectomy (OH) in which both the ovaries and the uterus are removed.

Ovariohysterectomy (OH) is more effective method to control reproduction in dogs, in comparison to castration, as we know that every female dog delivers 6-8 puppies in a litter and these unneutered animals reproduce logarithmically more unwanted litters to be recycled every year and hence, canine population increase in a fast manner.

Ovariohysterectomy/spaying in the bitch is a common procedure for birth control, to prevent oestrus and pseudopregnancy (Janssens, Janssens, 1991). Spay animals no longer feel the need to roam to look for a mate. The result is that they stay home and have less chance of being involved in traumatic accidents. They also have a much lower incidence of contracting contagious diseases, and get into fewer fights. Spay surgery also eliminates the heat cycle and associated mood swings and undesirable behaviors, messy spotting and the attraction of all available males to the yard.

Ovariohysterectomy not only the preventing uncontrolled breeding of dogs but also it is the treatment of choice for most uterine diseases like pyometra, uterine torsion and uterine neoplasia, and may also be indicated

for diabetic and epileptic animals to prevent hormonal changes that alter the effectiveness of medications. Ovariohysterectomy also decreases the incidence of mammary cancer (the rate goes down to almost zero if the spaying is done before the first heat cycle) in bitches.

Now days, Pet owners putting much pressure on the veterinarians to come up with a humane and effective technique for sterilization/OH in order to prevent breeding and cure the uterine disorders, this stressed the need of clinical evaluation and specializations of effective techniques for ovariohysterectomy to overcome the problems encountered in the techniques used traditionally.

Keeping in view, the above problems, different techniques of OH were undertaken in bitches and various clinical parameters were studied with the following objectives:

To evaluate the mid-ventral and right flank approach for various surgical disorders.

To find out the better approach of OH based on the different physiological, hematological and biochemical parameters.


REVIEW OF LITERATURE

CHAPTER-2

REVIEW OF LITERATURE

Gonadectomy is one of the most ancient surgical procedures performed upon domestic animals. As reported by Smithcors (1957), a treatise on the diseases of dogs, written in 284 B.C., indicated that the most common surgical practice for animals, possibly dating to prehistory, was castration. Despite the common practice and long history of surgical sterilization, little was written on the subject until the 20th century.

Age and timing for Ovariohysterectomy

According to Smithcors (1957) the concept of early neutering is not new. In the early 1900s veterinarians advocated spaying in bitches between 3-6 months of age or even prior to weaning.

Joshua (1965) had highlighted the effects of spaying on puppies before puberty and after puberty. Author had opined that spaying should be delayed until after completion of one, or preferably more heat cycle, to minimize the disadvantageous effect of spaying, as prepubertal neutering would result in persistence of an infantile vulva and perivulvarular dermatitis. The best time for spaying was in anoestrous and it was undesirable in proestrus and estrus.

According to Smith (1974) though ovariohysterectomy could be done at almost any age and at any phase of the reproductive cycle, it was best performed either before puberty or during anoestrous. Ovariohysterectomy, at 4 to 6 months of age was generally considered best. He stated that some surgeons preferred to wait until a bitch passed through one heat period before spaying, as they believed that it would allow her to develop more female characteristics. He also reported that the surgery was most hazardous during estrus and pregnancy and in old,

obese females. The most favorable time to spay a mature bitch was 3-4 months after estrus. After whelping, the operation should be done as soon as the puppies have been weaned and mammary gland activity has ceased. This was about 6-8 week following parturition.

Salmeri *et al.* (1991) were of the opinion that elective gonadectomy provided a permanent means of birth control; the efficacy of sterilization programmes could be greatly enhanced if all animals were neutered early.

Aronsohn and Faggella (1993) neutered Ninety-six kittens (48 males and 48 females) between the ages of 6 and 14 weeks. There were no important anesthetic complications, or complications during or after surgery. Pediatric neutering of kittens is a low-risk procedure when specific guidelines are followed. It is recommended that a complete preanesthetic evaluation be performed, a quiet and warm preoperative and postoperative environment be provided, handling of kittens be minimized, bleeding during surgery be meticulously controlled, fragile pediatric tissues be handled gently, kittens be offered food shortly after standing to prevent hypoglycemia, and dextrose be administered per.os. or intravenous if recovery is prolonged.

Stubbs and Bloomberg (1995) recommended early age neutering of dogs and cats as a safe and effective means of pet population control. The surgical techniques are similar to those already familiar to the veterinary practitioner and pose minimal risk to the animal patient. Advantages include a shorter operative time, better intra-abdominal visualization, and rapid animal patient recovery. Prepubertal gonadectomy does not seem to adversely affect skeletal, physical, or behavioral development in the dog and cat.

Bloomberg (1996) stated that the technical advantages of early age neutering included decreased operative time, improved visibility of intra abdominal structures, and rapid recovery from anaesthesia. He stated that most veterinarians, during training were instructed that the optimal age for neutering cats was 5-8 months, but a search of literature had revealed that early in this century cats, dogs and other domestic farm animals were neutered at 4 weeks to 6 months of age. Now there was evidence that neutering cats and dogs less than 4 months of age was a safe and effective procedure.

Fingland (1997) stated that minimum scientific evidence existed to support the widely accepted practice of delaying elective sterilization until an animal was 5-8 months old and an early prepubertal gonadectomy, i.e. at 8-12 weeks of age, had been supported because of efficacy of sterilization programmes.

Howe *et al.* (2001) determined long-term results and complications of gonadectomy performed at an early age (prepubertal) or at the traditional age in 269 dogs, and concluded that with the exception of infectious diseases, prepubertal gonadectomy might be safely performed in dogs without concern for increased incidence of physical or behavioral problems during at least a 4-years period after gonadectomy.

Olson *et al.* (2001) stated that prepubertal gonadectomy, often referred to as early-age neutering, and has increased in popularity in the United States. Early-age neutering is one technique that was used to combat pet overpopulation, a problem whereby millions of unwanted healthy dogs and cats are euthanised each year. Author reported that early-age neutering does not stunt growth in dogs or cats (a once-held belief), but may alter metabolic rates in cats. The anaesthetic and surgical procedures are apparently safe for young puppies and kittens; morbidity is lower and recovery is faster than in adult animals. To date, adverse

side effects are apparently no greater in animals neutered at early ages (7 weeks) than in those neutered at the conventional age (7 months).

Spain *et al.* (2002) conducted a survey in practicing veterinarians in small-animal or mixed-animal practice in New York state, USA about their beliefs and practices regarding the age at which dogs and cats should be neutered and their attitudes toward early neutering (at 4 months of age or younger) in 2000. The majority of veterinarians routinely recommended neutering for all client animals (70.6%) and supported the routine neutering of shelter animals before adoption (90.3%). More veterinarians in this study reported at least one perceived benefit (91.3%) for early neutering than reported at least one perceived risk (84.4%). Veterinarians with experience of early neutering were less likely to believe that the procedure was associated with one or more risks.

Kustritz (2002) observed that, pediatric animals may have an enhanced response to relatively low doses of anaesthetic agents. Animals should be fasted no more than 3 to 4 hours before surgery to prevent hypoglycemia, and hypothermia should be avoided. Heart and respiratory rates must be monitored carefully throughout anaesthesia. Pediatric gonadectomy surgeries are quick with minimal bleeding. Anesthetic recovery is rapid. No significant short-term or long-term effects have been reported. Prepuberal gonadectomy is most useful for humane organizations and conscientious breeders wishing to preclude reproduction of pet dogs and cats while placing animals at an enough young age to optimize socialization and training.

Rocken (2002) studied the pros and cons of neutering cats from the age of 6 weeks in Sweden and found that the early spay or neuter surgery is quick, with minimal bleeding and quick recovery. There is less time required for the surgery and less stress to the animal. No common, age-related, serious intraoperative or short-term postoperative complications

have been reported. Spaying and neutering can be performed under injectable anaesthesia on kittens over 12 weeks of age.

Spain *et al.* (2004) evaluated the long-term risks and benefits of early-age gonadectomy, compared with traditional-age gonadectomy, among dogs adopted from a large animal shelter and found that early-age gonadectomy was associated with increased rate of cystitis and further decreasing the age at gonadectomy was associated with increased rate of urinary incontinence. It was further concluded that in female dogs increased urinary incontinence suggests that delaying gonadectomy until at least 3 months of age might be beneficial.

INDIACATIONS FOR OVARIOHYSTERECTOMY

Ovariohysterectomy has been indicated as a preventive measure or treatment for a number of conditions listed below-

1. To reduce pet overpopulation and numbers of unwanted litter

Salmeri *et al.* (1991) stated that one of the most common reasons for neutering young dogs is to prevent unwanted pregnancies, and concluded that elective gonadectomy provides a permanent means of birth control, through surgical sterilization might not be an ultimate solution to the pet overpopulation.

Stockner (1991) describe the economics and impact of surgical and nonsurgical methods of spaying. He opined that neutering of adult animals by veterinarians is the most common form of pet population control.

Johnston (1991) had used surgical neutering as a means of pet population control and stated that the benefits of the surgery on population control and individual animal health should be weighed against

surgical/anaesthetic risks, and the desirable or undesirable sequelae to the procedure.

Jochle (1991) reported that surgical interventions (spaying;neutering), always an option for pet population control and also reported that in western Europe, 90 % of all efforts for pet population control in the bitch involves progestins and 10% involve spaying.

Rowan (1991) stresses the need to pay to sterilization as apart of the solution to pet overpopulation. In the city of Los Angeles for example, a local spay/neuter programme has been widely credited with reducing the number of animals

Avanzino (1991) stated about various agencies which actively participates in the spay/neuter programme and stressed that veterinarians should get increasingly involved in putting an end to the pet overpopulation problem

Bloomberg (1996) compared surgical and nonsurgical means for controlling pet population, specially that of cats and concluded that surgical sterilization of cats whether pre or post pubertal was the most reliable and commonly used method of rendering cats incapable of reproducing and it also reduced the incidence of androgen and oestrogen dependent medical disorders and other reproductive diseases or conditions related to the reproductive organs.

Remfry (1996) also used ovariohysterectomy to control feral cat population in the United Kingdom and concluded that neutering feral cats and returning them to the site of capture was widely accepted as an effective method of population control, more humane and cost effective than any other currently available alternative.

2. For prevention of ovarian and uterine cancer/diseases.

Dow (1957) suggested that pyometra was a common disorder of the canine uterus and was most common in bitches more than 6 years old or in those given oestrogen injection for mismating. Spaying was a solution to prevent this condition.

Dow (1958) suggested that the commonest form of endometritis in the bitch was the condition loosely described as pyometra, and the survey indicated that the cystic hyperplasia-pyometra complex was in some way related to the functional activity of the ovaries. This was supported by the fact that the clinical disease could be cured by ovariectomy.

Frye (1983) had removed a canine ovarian teratoma from a four and half year old, multiparous German Shepherd bitch in estrus which was presented for ovariohysterectomy.

Christiansen and Schmidt (1983) observed that the number of the follicles and corpora lutea indicated that pyometra may occur at any state of the cycle, but most frequently in metoestrus and oestrus. The uterine discharge was either sterile or infected with gram-negative or gram-positive bacteria, most frequently E. coli. The condition is caused by an intense reaction to physiological concentrations of progesterone and oestrogen, eventually followed by secondary invasion of microorganisms normally occurring in the vagina. The most effective treatment is still ovariohysterectomy, because the use of prostaglandins cannot be recommended until further investigations have been carried out.

Thacher and Bradley (1983) diagnosed vulvar or vaginal neoplasia in 99 bitches. The commonest clinical sign was vulvar discharge or the sudden appearance of a protruding mass from the vulva. Most of the dogs were old (average age, 10.8 years) and sexually intact, consisting of 72 benign, 17 malignant, and 10 transmissible venereal tumours. Except for the transmissible venereal tumours, most tumours were of smooth muscle

or fibrous tissue origin (leiomyoma, fibroma, and leiomyosarcoma). In the dogs with benign tumours, surgical excision of the tumour combined with ovariohysterectomy was effective in preventing recurrence of disease.

Sorribas (1987) performed ovariohysterectomy in an 11 year old cat, who was presented in a weak condition with a mucopurulent vulvar discharge. Celiotomy revealed a 400 g tumour within the uterus which on histopathological examination diagnosed as a submucous uterine fibroma.

Karczewski *et al.* (1987) treated surgically 206 bitches of pyometra in 3 years, of which 150 were over 7 years old, 48 between 4 and 7 years and 8 between 1 and 3 years. In 23% ovariohysterectomy was performed shortly after the sudden onset of pyometra; the remaining 77% underwent surgery after 2 or more weeks. Fatal outcome of the operation was recorded in 24 bitches, of which 41.6% died within 24 hours. Of 182 bitches which survived the operation, 160 had an uneventful recovery. Postoperative behavioural changes viz., were observed in 61.5% and excessive obesity in 50.5%.

Nelson and Feldman (1987) found that Ovariohysterectomy was the treatment of choice in pyometra in older bitches and queens. The incidence of pyometra appears to be increasing, especially in younger animals, as a result of increasing use of oestrogen and progesterone for mismating and for certain medical disorders. The clinical signs of pyometra and abnormalities on physical examination are dependent on the patency of the cervix and how quickly the client recognizes the problem.

Johnston (1991) stated that vaginal prolapse is an estrogen-mediated disorder of the young, sexually intact large-breed bitches at the time of proestrus or estrus, or at the end of diestrus and it can be prevented by ovariohysterectomy or by any treatment to prevent reproductive cycling.

Fingland (1997) stated that ovariohysterectomy was a treatment for most uterine diseases as diffuse cystic endometrial hyperplasia, uterine rupture and uterine neoplasia.

3. As a solution to be public health implications due to the stray animals (Dogs)

In a study of behavioural alteration in 2 groups of stray cats having been neutered, Neville and Remfry (1984), found a general fall in aggression and rise in affectionate interactions and also a fall in the undesirable actions as noisy courting, urine marking etc. So spaying could be used to reduce aggression amongst stray animals as suggested by the author.

Beaver (1991) reviewed the public health implications due to the large surplus stray animal population and found considerable prevalence of dog bites, problems of zoonotic diseases, garbage scattering, faecal deposits and urine volume as the chief consequences of the stray dog and cat overpopulation, faced by the citizens. He opined that public health implications of surplus animal problem were important and suggested use of surgical castration and ovariohysterectomy to combat the same.

4. To prevent occurrence of mammary tumors.

Cotchin (1959) reported that mammary neoplasms are not known to develop in bitches that are spayed before sexual maturity, although they are occasionally found in bitches spayed when adult.

Brodey (1970) opined that mammary neoplasia was the most common tumor of the sexually intact bitch, comprising 42% of tumors from all sites and 82% of reproductive tumors in the bitch.

Johnston (1985) stated that ovariohysterectomy before 1st estrus reduced the incidence of mammary neoplasia to 0.5%, after 1st estrus

risk factor of mammary neoplasia was 8% and after 2 or more oestrous the risk rise to 26% as compared to sexually intact bitches.

Center and Randolph (1986) treated Feline mammary hyperplasia recurring continuously in three successive gestations, in a 10-month-old, female domestic short-haired cat by ovariohysterectomy.

Morris *et al.* (1988) reported 58 bitches out of 90 bitches having benign tumours and, of these 15 (26%) developed a new mammary tumour within 2 years of the surgical removal of the primary tumours, irrespective of whether the bitch was spayed or not. The other 32 bitches had malignant tumours which were grouped into 'invasive' and 'well defined' carcinomas. 63% of the spayed bitches and 57% of the entire bithes, with invasive carcinoma were dead within 2 years of surgery as a result of their mammary tumours. For those with well defined carcinomas the tumour-related death rates were 18% and 33%, for the spayed and entire bitches respectively and they suggest that ovariohysterectomy when mammary tumours are removed does not have a significant effect on the recurrence of mammary tumours.

Schneider (1990) stated that ovariohysterectomy in bitches prior to first estrus reduces the likelihood of mammary tumor development to a greater extent than would spay after first estrus.

Concannon and Meyer (1991) stated that ovariohysterectomy has the additional advantage of substantially reducing the risk of mammary neoplasia if performed before the first or second estrous cycle.

Johnston (1991) reported that sexually intact bitches had 3-7 times greater risk of developing mammary cancer compared with neutered females, and risk is lowest for bitches spayed at a young age. Once mammary neoplasia had been diagnosed (usually in bitches more than 6 year of age), and if the tumor was an adenocarcinoma (about 24% of all canine mammary tumors) rich in oestrogen and progesterone

receptors (about 40% of adenocarcinomas), ovariectomy might be beneficial in preventing progression of the mammary disease.

Sorenmo *et al.* (2000) reported significant decrease in the risk of developing mammary gland tumors in dogs by ovariohysterectomy at an early age. However, previous studies have not found a benefit to ovariohysterectomy concurrent with tumor removal in dogs with established mammary gland tumors, suggesting that the progression of these tumors is independent of continued estrogen stimulation. The purpose of this study was to evaluate the effect of spaying and of the timing of spaying on survival in dogs with mammary gland carcinoma. Spay status and spay age, tumor characteristics, treatment, survival, and cause of death of 137 dogs with mammary gland carcinoma were analyzed. The dogs were classified into 3 groups, according to spay status and spay time. (1) Intact dogs, (2) dogs spayed less than 2 years before tumor surgery (SPAY 1) and (3) dogs spayed more than 2 years before their tumor surgery (SPAY 2). Dogs in the SPAY 1 group lived significantly longer than dogs in SPAY 2 and intact dogs (median survival of 755 days, versus 301 and 286 days, respectively, P = .02 and .03). After adjusting for differences between the spay groups with regard to age, histological differentiation, and vascular invasion, SPAY 1 dogs survived 45% longer compared to dogs that were either intact or in the SPAY 2 group (RR = .55; 95% CI .32-.93; P = .03). This study reveals ovariohysterectomy to be an effective adjunct to tumor removal in dogs with mammary gland carcinoma and that the timing of ovariohysterectomy is important in influencing survival.

TECHNIQUES

Preparation of Site

Gibson *et al.* (1997) compared a one-step iodophor skin preparation solution with chlorhexidine gluconate application as a pre-operative skin

preparation method in 100 animals undergoing elective ovariohysterectomy. Pre-operative and intra-operative skin cultures demonstrated no difference in antiseptic efficacy. No animal in the study demonstrated signs of systemic infection, and no adverse local effects from either antiseptic were seen and recommended the iodophor solution (DuraPrep) as a safe and effective pre-operative skin preparation agent in small animal patients undergoing clean surgical procedures.

Hansson (2005) noted in Sweden, the most common way to castrate a bitch is by midline approach. He compared flank and midline in order to evaluate incision by the flank as an alternative technique for ovariohysterectomy in the bitch. The study was performed on stray dogs in the central part of Thailand under field conditions. 32 bitches were included in the study. The flank and midline approach, alternating with every other bitch were used. The study showed that the time used for surgery was equivalent for the two different techniques. The midline approach though, had a larger variation in time. The amount of suture material used was almost the same for both techniques. Dogs that were castrated by the midline approach had more complications with respect to bleedings from the ligatures during surgery. However, tissue bleeding when passing the abdominal wall was found to be less in this group. Post surgery complications as inflammation and infection were similar for the two techniques. The results of this study show that the flank approach can be a proper alternative to the midline approach.

MIDLINE APPROACH

Wilson and Balasubramanian (1967) stated that the most widely accepted site of incision for spaying in standard text book is a median incision which is made on the midline through the linea alba, as in this muscle fibers are not incised, no vessels or nerves are cut and the scar left is invisible. But abdominal wound dehiscence characterized by evisceration and prolapse on the 4^{th} or 6^{th} post operative day has been

found to be the main hazard in this method. Further, they have opined that often, considerable difficulty is encountered in large, obese patients through this approach in the exposition of ovaries, because in the nongravid uterus, the suspensory ligaments are short.

Smith (1974) performed spaying through a midline abdominal incision beginning over the umbilicus and extending caudally for 2-5 cm, using 3 forceps tie for ligation and severing of ovaries and uterine body.

The author reported hemorrhage, infection, wound dehiscence, recurrent oestrous, pseudocyesis and post surgical problems associated with anaesthesia and shock as complications of ovariohysterectomy in bitches.

Dorn (1975) stated that the usual surgical approach for ovariohysterectomy, through ventral midline requires animal to be restrained in dorsal recumbency and anaesthesia has to be maintained till the completion of surgery. Besides, these the other demerits of mid-ventral approach are the increased possibility of wound dehiscence, due to weight of abdominal viscera, pressing on the incision, improper suturing of surgical site or introduction of infection during surgical procedure.

Lande (1975) discussed the disadvantages of the customary long incision (from symphysis pubis to umbilicus) for ovariectomy in the bitch. To avoid these a new method was developed, in which an initial 3-4 cm long incision is made just above the symphysis pubis, a finger inserted cranially to lift the abdominal wall away from the organs, then a second incision made, a little more than 4 cm below the umbilicus, but leaving at least 3-4 cm of the abdominal wall intact. Suturing and severing of the cervix was performed through the caudal incision, and then the ovariohysterectomy was completed through the cranial incision.

Rubin and Maplesden (1978) used a small skin incision on ventral midline, posterior to the umbilicus, approximately one forth of the distance

between the umbilicus and symphysis pubis for ovariohysterectomy in bitches.

Bojrab (1981) discussed midline approach as easier, less traumatic and is associated with less post-operative pain than the flank approach, which involves incising three muscle layers. Also, any intra-operative haemorrhage was more easily controlled.

Yanoff *et al.* (1982) reported ventral midline incision for ovariohysterectomy in raccoons, midway between the umbilicus and the pubis. The ovaries were grasped and their suspensory ligament broken manually one by one and the ovarian and uterine stumps were transfix and ligated with polyglycolic acid 3-0 suture. The abdominal wall and subcutaneous tissue were closed with 3-0 Dexon and skin with multifilament nylon using simple interrupted pattern in all 3 layers.

Stone (1985) opined that the Spaying may be done through a midline incision from umbilicus approximately 5 cm caudally in dog and 1cm caudal to umbilicus, extending caudally for 3cm in cat. The ovaries and uterine body were ligated using 3 forceps tie, and severed. Delayed wound healing, suture abscess, infection and self-inflicted trauma to the wound were found as most common complications of mid-ventral approach to ovariohysterectomy.

Hoque (1991) has discussed the demerits of spaying through linea alba. According to author, longer incisions were needed to exteriorize uterus and ovaries through linea alba and it led to more manipulation of abdominal viscera, during tracing of uterus and ovary and frequent evisceration due to straining. Moreover, skin incision at linea alba took 3-4 more days for healing than flank incisions which might be due to decreased vascularity at the site. Wound dehiscence was a common complication in the cats spayed through linea alba and resuturing of skin wound had to be undertaken in such cases.

Aronsohn and Faggella (1993) documented surgical neutering of 6-14 week old kittens through a 3-4 cm ventral midline skin incision, 1 cm caudal to the umbilicus for ovariohysterectomy. Medium haemoclips were used for ligation of ovarian and uterine stumps. Ventral fascia was closed with 3-0 nylon in a continuous pattern and skin was left unsutured.

Flynn (1995) advocated suture less spaying operation through linea alba in young (2 month old) prepubertal animals. The operation was done with the animal in topsy-turvy position, through a ½-1 inch skin incision, 2 inches posterior to the umbilicus, by exteriorizing the uterine horns one by one and removing the ovaries by torsion using spaying forceps. The uterine stumps were replaced in peritoneal cavity and skin incision was left unsutured, but a piece of 2 inch adhesive tape was finally passed around the patient's body to support the incision and close the small skin wound. The technique consumed less time and the patient could be discharged 23-48 hours after surgery.

Fingland (1997) made use of a midline abdominal incision extending from the umbilicus to a point half way between the umbilicus and the brim of pubis, for ovariohysterectomy in dog. He first located the left uterine horn and exteriorized the ovary. The ovarian pedicle was triple clamped and ligated using a circumferential and transfixation suture. The right ovarian pedicle and uterine stump were also ligated and severed in a similar fashion. He prefered use of absorbable suture (e.g. chromic catgut) for all ligatures. Abdominal incision was closed with either absorbable suture in an interrupted pattern or with a non-absorbable suture in simple continuous pattern.

Sohst *et al.* (2002) conducted a study on female 2015 cats, which were presented for neutering at the Animal Hospital. The surgery performed along the linea alba in cats fixed on a lying or sloping operation table has 98.8 per cent chance of healing. During surgery pregnancy was diagnosed in almost a quarter of all patients. This is due to the high

number of feral cats. Neutering is generally carried out by ovariectomy or partial ovariohysterectomy. Only in cases of pathologically changed uterus or pregnancy in progress is the complete uterus removed. Wound healing problems in the postoperative phase occurred in more than 6 per cent of the queens, which can mainly be related to the suture material and technique. Suturing the operation wound in layers is recommended. To avoid the removal of stitches, especially in feral cats, an intracutaneous suture should be made.

Sgorbini *et al.* (2002) attempted ovariectomy in 70 cases of cats using a specific "castration hook" that is introduced through a very small median laparotomic incision. In 47 subjects, surgery was successfully performed, while in 16 others, some difficulties arose in uterine clasping. Only in the 7 remaining females was a broader abdominal incision required to carry out ovariectomy or ovariohysterectomy. In uterine clasping, two different Desmarres' retractors showed greater efficiency and safety than the castration hook. Young, thin, pubertal females were the best candidates for this intervention.

Sylvestre *et al.* (2002) performed a ovariohysterectomy through a ventral midline 2.5 and 5 cm long, skin incision, using standard procedure. The ligatory suture material used for ovarian and uterine pedicles was 2-0 or 3-0 poliglecaprone 25. The surgical site was closed. The linea alba was sutured in a simple interrupted pattern using 2-0 or 3-0 poliglecaprone 25, the subcutaneous layer was closed with a simple continuous pattern using 3-0 poliglecaprone 25. Knots were tied with 4 single throws for the interrupted pattern and 5 throws for the continuous pattern

FLANK APPROACH

O'conner (1950) has suggested the use of vertical 3/8 inches long incision in level with the anterior border of the ilium and about the middle of flank with the patient in ventral position for ovariohysterectomy in cat.

Ormrod (1966) has advocated the use of an oblique flank incision for ovariohysterectomy in cats and has opined that the skin incision at linea alba takes 3-4 days more for healing, which may be explained by the fact that there is reduced vascularity at linea alba.

Wilson and Balasubramanian (1967) performed spaying in dogs through right flank with the animal restrained in left lateral recumbency and under general anaesthesia, making use of a 7-8 cm vertical or oblique skin incision in mid-flank region below the transverse processes of lumbar vertebrae in right paralumbar fossa. Muscle separation technique was followed for entering the abdomen, with the right ovary lying just beneath the incision. For closure of the abdominal wound, the muscles were sutured in 3 layers using medium chromic catgut '0' with simple continuous suture and skin was sutured with cotton, interrupted mattress sutures.

Krzaczynski (1974) used flank approach to ovariohysterectomy in cat, which was performed through a vertical skin incision over the cranial border of tensor fasciae lata muscle over left flank. Spaying was done as per the standard procedure, but the rent in abdominal musculature was left unsutured and skin was sutured with interrupted pattern. Author reported that with flank approach evisceration never occurred, it was less time consuming, there was less anaesthetic requirement and so it was more economic. But the disadvantages were that if a stump was lost accidentally, its recovery was difficult and site was located improperly, reaching either ovary or bifurcation of uterine horn would be difficult and the approach was contraindicated in advanced pregnancy.

Dorn (1975) has reported the widespread use of flank approach of ovariohysterectomy in United Kingdom and other areas of the world. He had performed spaying in dogs through right flank, making use of muscle separation technique for entering the abdominal cavity and avoids hemorrhage which might result from cutting of muscles. He further pointed

that the approach was easier from the right side, because in dog, the right ovary is located farther cranially than the left ovary and approach on this side permitted better access to ovary. Approach from the left side was more difficult because the omentum covered the viscera and the spleen lied in the surgical field. The advantages of right flank approach were the speed of procedure and low incidence of wound dehiscence. But the demerits included increased possibility of haemorrhage, limited exposure of contralateral ovary, resultant surgical scar and the hazard of dropping the left uterine horn back into abdominal cavity. The approach was not recommended for dogs with a short, tight suspensory ligament and with ovarian or uterine pathology.

Hoque (1991) performed ovariohysterectomy in cats through left flank, under tranquilization with triflupromazine hydrochloride and local analgesia with 2% procaine hydrochloride, by giving a 2.5 cm long horizontal incision below and parallel to mid lumbar region. He compared ovariohysterectomy via horizontal flank incision with vertical and oblique flank incisions and operations through linea alba and found horizontal flank incision superior over other techniques due to easy and complete localization and exteriorization of uterine horns and ovaries, due to synchrony in length and position of uterine horns and no intra and postoperative complications.

Janssens and Janssens (1991) used a bilateral surgical flank approach for ovariectomy in 72 bitches of 27 breeds. They pointed out that the main technical advantages of this approach were better ovarian exposure, possible deeper placement of ligatures and small incisions. The disadvantages were the need to turn the animal, to redrape it and the difficulty in finding the ovarian bursa in some animals and difficulty in removing the uterus when necessary. Postsurgical complications were minimum and consisted mainly of seroma formation.

Remfry (1996) has recommended neutering of feral cats in United Kingdom via flank approach, with the center of incision lying perpendicularly ventral to the tuber coxae,and the line of incision was bisect the angle between the last rib and the tips of the lumbar vertebral processes, performed under anaesthetic combination of ketamine and xylazine with the little risk of evisceration, compared with that for midline incision.

MISCELLANEOUS TECHNIQUES

Wood *et al.* (1984) conducted a study using stainless steel, monofilament nylon, coated polyamide, polypropylene, and silk in linea alba closure following routine ovariohysterectomy in 12 dogs. Groups of three dogs were killed at 30, 60, 90, and 120 days after surgery. Tissue surrounding the incision site was collected, and histological sections of the suture-induced inflammatory reactions were evaluated. Silk caused the most severe inflammatory reaction irrespective of the time at which sutures were evaluated. Stainless steel and nylon caused the least inflammation. Granuloma formation around fragments of polypropylene increased the grade of the tissue reaction so that it exceeded that of nylon. Tissue reaction to coated polyamide varied greatly among dogs. On the basis of this study, nylon and stainless steel would appear to be the sutures of choice. However, the advantage of minimal tissue reaction must be balanced against other qualities when selecting the appropriate suture.

Okamoto *et al.* (1998) performed laparoscopy assisted ovariohysterectomy in 4 dogs and 3 cats, after producing pneumoperitoneum and inserting 2 trocars with cannulae at both lateral sides of abdominal wall and 1 cannula in the cranial side of pubis. Ovarian vessels, suspensory ligament and mesovarium were resected through lateral trocars. The ovaries and uterus were extracted through the caudal

trocar and uterine body was extracted after ligation. No complications occurred after surgery.

Barbosa *et al.* (1999) used titanium vascular staples (Ligaclip LT-200) to ligate the ovarian artery and vein (ovarian AV complex) and median uterine artery, during elective ovariectomy in 20 healthy cats of varied age group. The staples remained in place as revealed by radiographs taken 14 and 28 days postoperatively and by sacrificing 3 cats 90 days after surgery, they concluded that the staples were safe, effective, easy and quick to use and remained in place after surgery.

Valocky *et al.* (1999) advocated the use of laparoscopy for sterilization of 8 German shepherd bitches between 2-7 years age, in 3 ways, ie by creating mechanical occlusion of oviducts using an endostapler technique (n=3), forming oviduct occlusion by electrocoagulation with the aid of an electrocoagulater (n=3) and performing ovariectomy using modified endosuture techniques (n=2), and found these methods relatively effective and advantageous.

Dharmaccelan *et al.* (2000) compared 3 different techniques of laparoscopic sterilization viz; resection and removal of the ovary after clip application, electrocautery of the ovary and electrocautery and removal of the ovary after clip application in 18 healthy female dogs. They opined that laparoscopic sterilization was more effective and safe method than conventional surgery, because of its benefits as absence of large abdominal incision, reduction in post operative pain, early wound healing and rapid normalization of general well being. The ovaries and associated structures were better visualized by laparoscopy and no intra or postoperative complications occurred. They found resection and removal of ovary after clip application, superior over the other 2 techniques and were of the view that it could be better adopted for female dog sterilization.

Sylvestre *et al.* (2002) conducted a study to compare postoperative wound healing in canine ovariohysterectomy following the use of an absorbable monofilament poliglecaprone 25 suture in 2 different skin closure techniques, the buried continuous subcuticular (BCS) suture pattern and the simple interrupted (SI) suture pattern. These 2 skin closure techniques were evaluated against a non-absorbable polypropylene monofilament suture in an SI pattern. Wounds were assessed by using a semi quantitative scoring system at 18 to 24 hours and 10 to 14 days, postoperatively. Results indicated that the BCS closure using poliglecaprone 25 demonstrated a higher rate of tissue reactivity initially (18-24 hours postoperatively), as compared with the SI closure using either suture material. By 10 to 14 days postoperatively, poliglecaprone 25 used in a BCS closure was associated with significantly lower wound scores than with the same material used in an SI closure. It was concluded that the BCS closure may affect a better cosmetic appearance to the skin closure in a canine ovariohysterectomy at the time of the recheck appointment. Furthermore, by obviating the need for suture removal, use of the BCS pattern may eliminate the requirement for this return appointment.

POSTOPERATIVE ANALGESIA

Slingsby and Waterman (2002) conducted a study in eighty female cats, presented for ovariohysterectomy by randomly allocating to one of two treatment groups in this assessor-blinded trial. After pre-anaesthetic assessment, the cats were premedicated with acepromazine (0.1 mg/kg). Anaesthesia was induced with thiopentone and maintained with halothane in oxygen. Forty cats received carprofen (4 mg/kg subcutaneously) and 40 received meloxicam (0.3 mg/kg subcutaneously) after anaesthetic induction. Following routine flank ovariohysterectomy the cats were assessed using visual analogue scale scores for pain and sedation over a 20-hour study period. Blood samples were taken before sedation and at

20 hours for serum biochemistry (urea, creatinine, alanine aminotransferase and aspartate aminotransferase). There were no significant differences between the groups for pain and sedation scores. Serum biochemistry values were similar between the groups, with some differences within groups between the pre-sedation and 20-hour values. One cat in the carprofen group and two cats in the meloxicam group required rescue analgesia with intramuscular morphine (0.2 mg/kg).

Burrow *et al.* (2006) twenty entire female cats were randomly assigned to two groups of 10; for ovariohysterectomy by a midline approach and flank approach. Cats were assessed for signs of pain and scores were assigned pre- and postoperatively. There was a tendency for the cats neutered by a flank approach to be in more pain postoperatively ($P<0.05$). The final pain score for cats in either group was equal to or lower than their baseline score.

COMPLICATIONS OF OVARIOHYSTERECTOMY

The complications and sequelae of ovariohysterectomy can be listed as-

1. Hemorrhage

Joshua (1965) described the various operative and postoperative complications of ovariohysterectomy viz; hemorrhage granuloma and ligature sinuses, hydronephrosis, pyometra in residual uterine horns, recurrent heat, pseudopregnancies and adhesion formations in abdominal viscera.

Pearson (1973) suggested that hemorrhage after ovariohysterectomy could be classified into two types: primary and secondary. Primary hemorrhage is a result of a ligature not properly placed or not tightly secured, or due to excessive traction on the uterine body which could tear the uterine vessels, among other factors. This type of hemorrhage leads to blood accumulation in the peritoneal cavity.

Secondary hemorrhage could be a result of ruptured ovarian or uterine vessels or infection around the ligature due to the use of contaminated material, which would lead to vaginal bleeding a few days after surgery.

2. Recurrent estrus and remnant ovarian syndrome

Pearson (1973) opined recurrent estrus in ovariohysterectomized bitches as a result of left over functional residual tissue, usually found on 2nd exploratory laparotomy and more frequent on right than left side and could be effectively treated by excision of the ovarian remnant during metoestrous.

Okkens-*et al.* (1981) reported single or multiple complications following ovariohysterectomy in 109 dogs, which showed gynaecological symptoms: discharge from the vulva (28), attractiveness to male dogs (37), periods of heat (28) and pseudopregnancy (14). In addition to a general clinical examination, the bitches were given a selective gynaecological examination usually consisting of clinical, cytological and bacteriological studies and of these nearly half (47) having remnants of ovarian tissue which were removed during laparotomy.

Wallace (1991) the ovarian remnant syndrome is a complication of ovariohysterectomy resulting in the presence of functional ovarian tissue in the abdomen. The usual symptom is a return to estrus in a previously ovariohysterectomized bitch or queen. The available methods of diagnosis are vaginal cytologic evaluation during estrus, resting hormone assays, hormone challenge testing, and exploratory laparotomy. Surgical removal of the ovarian remnant is the preferred treatment.

Perkins and Frazer (1995) described ovarian remnant syndrome (ORS) in the dogs and cats and it appears to be a less frequent complication of routine ovariohysterectomy in the female dog and queen than in women. This has been attributed to the rare occurrence of pelvic inflammatory disease and of the associated adhesions in small animals,

allowing the surgeon to recognize and remove the ovaries more easily in routine ovariohysterectomy. Animals with ORS present with a range of symptoms associated with endogenous hormone production by the remnant of ovarian tissue present in the abdomen. These include vulval swelling, proestral bleeding, estrous behavior and occasional mammary gland enlargement and milk production. The recommended treatment for ORS is surgical removal of remaining ovarian tissue. Histological examination of ovarian remnant tissue usually demonstrates the presence of corpora lutea or simple cystic structures. In a series of 72 females referred to the University of Bristol for complications arising after ovariohysterectomy, 12 (17%) were referred for continuing signs of estrus. No diagnostic tests were performed on these cases and functional ovarian tissue was found in all cases during exploratory laparotomy. The time interval from ovariohysterectomy to reported signs of estrus varied from 17 d to 9 yr. Hormonal challenge testing using GnRH was utilized in all eleven feline cases to demonstrate an elevation in serum progesterone concentration attributed to ovarian tissue. Signs of recurrent estrus resolved in all cases after surgical removal of ovarian tissue.

Sangster (2005) reported Ovarian remnant syndrome in a 5-year-old German shepherd bitch presented with a 3-week history of vaginal bleeding, diagnosed on vaginoscopy, vaginal cytology, serum progesterone levels, ultrasonography, and exploratory laparotomy. The condition resolved following surgical excision of the remaining ovarian tissue.

3. Urinary incontinence and ligature of ureter

Brothwick (1972) and Turner (1972) reported hydronephrosis in a spayed bitch, due to accidental ligation of a ureter during ovariohysterectomy.

Ruckstuhl (1978) found intermittent urinary incontinence in 16 (13%) of 120 bitches after ovariectomy and in 7 (10%) of 69 bitches after

ovariohysterectomy. Of 1147 uncastrated bitches examined in 1973, only three had urinary incontinence. Of the 189 spayed bitches, 98 were more than 15 kg in weight and 91 were less than 15 kg; the respective incidence of urinary incontinence was 17 and 6%, respectively, which was significantly different. Most of the cases occurred in the 24 months after the operation. Incontinence was intermittent in all animals. Long-term treatment with oestrogens or progestational hormones was generally successful.

Sehic *et al.* (1981) found a seldom occurring bilateral hydronephrosis caused by ureteral obstruction following abdominal surgery (caesarean section and ovariohysterectomy) and diagnosed radiologically in a bitch. The renal ducts were detached from the connective tissue adhesions, and the hydronephrotic kidneys were drained. Clinical observations including radiography and laboratory examinations were carried out during the course of the disease until complete renal sufficiency occurred two years later.

Thrusfield (1985) observed hormonal urinary incontinence in 34 of 79 (4%) bitches, submitted to ovariohysterectomy and 7 of 2,434 (0.3%) of sexually intact bitches.

Arbeiter (1986) found that nocturnal urinary incontinence in spayed bitches was sometimes a functional disorder, for some cases respond to treatment with an oestrogen and/or ephedrine, and sometimes the result of formation of scar tissue in the lumbosacral area. A new treatment was paracervical infiltration of 20 ml Xyloneural (containing lidocaine).

MacCoy *et al.* (1988) reported a complication of an ovariohysterectomy due to formation of an ureterovaginal fistula secondary to simultaneous ligation of the left ureter and vagina. Ureteral transplantation and ligation of the fistula resulted in complete resolution of urinary incontinence.

Arnold *et al.* (1989) performed a follow up study in 412 spayed bitches, in order to determine the incidence of urinary incontinence. The period between the operation being performed and the survey being made varied between 3 and 10 years. 83 animals (20.1%) were incontinent independent of the surgical procedure (ovariectomy versus ovariohysterectomy). The onset of incontinence varied between immediately to 12 years with an average period of 2.9 years after surgery. There appears to be a strong connection between body weight and the incidence of incontinence. Bitches with a body weight of less than 20 kg (9.3%) were incontinent. Whereas bitches with a body weight of more than 20 kg the incidence was 30.9%. The breed based incidence of incontinence recorded was 65% in Boxers, 10.6% in German Shepherds and 11.1% in Dachshunds with an average incidence rate of 20.1%.

Ewers and Holt (1992) reported development of right hydroureter, hydronephrosis and vesicovaginal fistula following ovariohysterectomy done at the time of caesarean section in a 2 year old Chihuahua bitch, and opined that urinary incontinence was one of the most common urological complications and could occur as a consequence of a vesicovaginal fistula resulting from an accidental inclusion of the caudal bladder neck in a cervical stump ligature which resulted in complete ligation of the right ureter.

Arnold (1997) reported acquired urinary incontinence occurs in 20% of spayed bitches and there exists a strong correlation between body weight and the risk of urinary incontinence. Bitches with a body weight of more than 20 kg have a risk of 30% while smaller dogs have a risk of 10%. A particular breed disposition exists in Boxers, in which 65% are affected. Other breeds with a more than average disposition for urinary incontinence are Dobermans and Giant Schnauzers. Urinary incontinence due to spaying manifests itself mainly while the dogs are sleeping. The cause is a urethral sphincter incompetence which can be verified by a urethral

pressure profile. Further it is reported that the urethral closure pressure is significantly lower in incontinent bitches (4.6 +/- 2.3 cm H2O) than normal continent bitches (18.6 +/- 10.5 cm H2O). In addition, the urethral closure pressure dropped significantly within 12 months after surgery. Histological examination revealed that the functional urethral closure cannot be explained by the extent of discernible structures of the urethral wall as seen by light microscopy.

Stocklin *et al.* (2001) enquired owners of 206 bitches that had been spayed before their first oestrus, were questioned for 3 years on the occurrence of urinary incontinence as a result of spaying. At the time of the enquiry the average age of the bitches was 6.5 years, and the average age at the time of surgery was 7.1 months. Urinary incontinence after spaying occurred in 9.7% of bitches. This incidence is approximately half that of spaying after the first oestrus. Urinary incontinence affected 12.5% of bitches that were of body weight (> 20 kg) and 5.1% of bitches that were of body weight (< 20 kg). The surgical procedure (ovariectomy versus ovariohysterectomy) had no influence on the incidence, or on the period between spaying and the occurrence of urinary incontinence. Urinary incontinence occurred on an average at 2 years and 10 months after surgery and occurred each day, while the animals were awake or during sleep. However, compared with late spaying the clinical signs of urinary incontinence were more distinct after early spaying.

Moormann *et al.* (2003) reported violent abdominal pain and urine incontinence after ovariohysterectomy in a bitches, the surgical correction of urinary bladder portion by a surgical technique of a bladder neck suspension by using a facial flap led to correction of the problem.

4. Increase in Body Weight

LeRoux (1977) compared activity level and body weight fluctuations in spayed bitches with those of intact females and opined that spayed

bitches did not gain weight on a diet of a fixed amount of commercial dog food.

Johnston (1991) opined that ovariohysterectomized bitches may eat more and gain more weight than sexually intact bitches if allowed free access to food, but do not necessarily gain weight postoperatively if food intake is regulated.

Scott *et al.* (2002) investigated the body condition by measuring body weight (BW), body condition score (BCS), and falciform fat pad of feral cats after neutering. The lateral abdominal radiographs were taken at the time of neutering of 105 adult feral cats for measurement of falciform fat pad depth and area. At that time they also assessed BW and BCS. A subsample of 14 cats, at the time of surgery, revaluated that the cats were lean but not emaciated (BW 3.1±0.9 kg; BCS 4±1; based on a 1 to 9 scale ranging from 1 [emaciated] to 9 [grossly obese]). Falciform fat pad depth and area averaged 7.1 mm and 197.4 mm square, respectively, indicating a small amount of fat. 1 year after neutering, revaluation of these cats indicated increased 260%+90% in falciform fat pad depth, 420%±390% in fat pad area, 40%±4% in BW, and 1 level in BCS ranking (1 to 9 scale; all differences $P < .001$). Similar to confined socialized cats, feral cats gained significant weight and body fat after neutering.

Kanchuk *et al.* (2002) conducted a study with the aim to investigate the mechanism underlying the weight gain associated with neutering in cats. He found that both male and female cats significantly increase body weight after neutering. Body weight is regulated through control of energy intake and energy expenditure. Although it has been demonstrated that food intake increased after neutering, it was not clear whether an increase in food intake was the primary cause of body weight gain in neutered cats.

Hoenig and Ferguson (2002) conducted a study to determine whether changes in concentrations of hormones involved in glucose and

fatty acid homeostasis were responsible for the increased probability that neutered cats will develop obesity and diabetes mellitus. Results of glucose tolerance tests and concentrations of hormones and nonesterified fatty acids (NEFA) were examined before and 4, 8 and 16 weeks after neutering. They concluded that changes in NEFA suppression, caloric intake, and leptin concentrations might be indicators of, and possible risk factors for, the development of obesity in cats after neutering.

5. Intestinal and peritoneal adhesions

Gaag *et al.* (1981) reported upon 109 dogs submitted with complications following ovariohysterectomy, 20 were found to show enterological problems. Presenting symptoms in these animals consisted in vomiting, diarrhoea and emaciation. A large number of animals also showed body temperatures above 39 degrees Celcious. An irregular, often painful mass was palpable on examination of the mesogastrium in 17 cases. Plain radiography revealed the features of ileus in 10 dogs, a mass of soft tissues in the abdominal cavity in 6 and the pattern of peritonitis in 5 cases (2 animals were classifiable with 2 groups). One dog did not show any changes. Laparotomy was performed in all 20 dogs. In view of the extent of the lesions, euthanasia was performed in 10 animals. Partial enterectomy was performed in the other 10 dogs. The findings at laparotomy and subsequent post-mortem examination, were summarized as, there were adhesions between the ovarian-stump granulomas and an intestinal loop in 5 animals, while 12 animals showed adhesions between the ovarian-stump granuloma and the intestines which also adhered to each other; in 5 of these bitches, the intestinal loops were also attached to the wall of the abdomen, and also to the cervix stump in 2 cases. 3 dogs only showed adhesions between intestinal loops and the cervix stump was involved in the adhesions in one dog.

Coolman *et al.* (1999) Partial extramural obstruction of the descending colon was diagnosed in two dogs and a cat as a complication

of elective ovariohysterectomy. In each case, the obstruction was caused by fibrous tissue that encircled or crossed the descending colon, severely restricting the organ's normal mobility and luminal diameter. Clinical signs secondary to obstipation were observed in two cases, five weeks and 27 months after elective ovariohysterectomy. In one dog without clinical signs, the adhesion was an incidental finding during a laparotomy performed nine years after the ovariohysterectomy. The fibrous adhesions were removed surgically in all three cases without additional complications.

6. Ligature sinuses and fistulations

Joshua (1965) has reported the various surgical complications of ovariohysterectomy of which ligature granulomas which lead to fistulations or adhesions to adjacent viscera as ureter or intestine are a common sequelae.

Lubberink *et al.* (1981) reported inflammatory lesions caudal to the costal arch in 22 dogs over a period of examination for thirty months, which were showing following ovariohysterectomy. 17 patients showed fistulae, 4 showed painful swellings and 1 patient showed a fistula on one and a swelling on another side.

7. Cervical and ovarian stump granuloma

Gadelha *et al.* (2004) reported a case of seven-year-old bitch with a history of strangury since two months. Clinical signs had developed within two days of elective ovariohysterectomy. Ultrasonographic and radiographic examinations suggested stump pyometra or cervical granuloma and vesicovaginal fistula, which was treated surgically by doing laparotomy and further medically.

8. Miscellaneous

Mann (1971) described the potential complications of prepubertal spaying than after the 1st oestrous cycle. Complications associated with ovariectomy include perivulvar dermatitis and bilateral military dermatoses, however perivulvar dermatitis was more likely to develop and be of greater severity following prepubertal spaying.

Dorn and Swist (1977) evaluated 73 bitches undergoing ovariohysterectomies during a six-month period in 1973 for surgical complications. Twenty-three of 73 (31.5%) had complications such as delayed healing, hemorrhage during surgery, problems with anaesthetic management, infection, tracheobronchitis or coughing, haematuria, and self-inflicted trauma.

Johnston (1991) reported that the incidence of complications after ovariohysterectomy varies with the experience of the surgeon, types of suture materials and preoperative patient risk factors such as age, concurrent reproductive diseases (pyometra, for example), and non reproductive diseases including congestive heart failure.

Ettinger *et al.* (1995) reported that spayed and neutered dogs have also been known to develop hormone-responsive alopecia (hair loss).

Millis *et al.* (1992) hemostatic profiles were evaluated in 15 healthy dogs immediately before and 24hr after celiotomy for routine ovariohysterectomy. Prothrombin time, activated partial thromboplastin time, fibrinogen, fibrin degradation products, antithrombin III activity, platelet count, and haemogram were measured. There were no significant changes in prothrombin time, activated partial thromboplastin time, fibrin degradation products, antithrombin III activity or platelet count. Fibrinogen concentration was significantly higher following surgery. Postoperative leukocyte

differential counts were typical of stress leukograms, and were characterized by leukocytosis, neutrophilia, lymphopenia and eosinopenia. Mild decreases in packed cell volume, red blood cell count and haemoglobin concentration were consistent with minor blood loss during surgery or fluid retention and haemodilution postoperatively. It was concluded that celiotomy and routine ovariohysterectomy in healthy dogs did not alter haemostatic profiles 24hr after surgery. Abnormal postoperative haemostatic profiles should not be attributed to surgery alone; other causes of abnormal haemostatic profiles should be investigated.

Karmakar *et al.* (2002) studied the changes in total erythrocyte count (TEC), total leukocyte count (TLC), differential leukocyte count (DLC), haemoglobin percentage (Hb %) and packed cell volume (PCV) of twelve Spitz bitches (6-9 years old and 10.5±2 kg body weight) with pyometra before and after ovariohysterectomy. Six apparently healthy, non-seasonal and non-pregnant adult mongrel bitches (12±2 kg body weight) were used as control. Blood values were determined in all dogs prior and 15 days post-operation. A marked decrease in TEC, significant increase in TLC, neutrophils (shift to the left), eosinophils, lymphocytes and decrease in Hb% and increase in PCV were observed in bitches with pyometra prior to ovariohysterectomy. TEC gradually increased (4.02±0.19 to 5.38±0.09 x 106/cmm), TLC remarkably decreased (30.82±0.36 to 17.87±0.44 x 103/cmm) and Hb% gradually fell (16.27±0.53 to 13.28±0.06) in the experimental animals 15 days post-operation. Pyometra gives rise to major fluctuations in peripheral blood pictures. Although such changes in blood values were not sufficiently diagnostic, these help to confirm clinical diagnosis along with other characteristic clinical signs.

CHAPTER-3

SURGICAL ANATOMY

Surgical anatomy related to the ovariohysterectomy is mainly based on Miller (1964).

(1) RIGHT FLANK APPROACH

Site of Incision:- A horizontal, mid lateral rectus abdominal skin incision was made at right lower flank started from a point about 4-6 c.m. lateral to the umbilicus and parallel to midline, and about 1.5-2 c.m. above the caudal abdominal mammary teat, extending about 3-4 c.m. caudally.

At the site of flank surgical incision, following anatomically structures were encountered from outward within:

1. Skin
2. Subcutaneous Fascia
3. Cutaeneous Trunci Muscle
4. Deep Fascia
5. Abdomoinal Muscles and their aponeurotic parts viz. Obliques abdominis externus, Obliques abdominis internus, Rectus abdominis and Transversus Abdominis
6. Parietal Peritoneum
7. Viscera

The skin, superficial fascia and Cutaeneous Trunci muscle are closely associated with each other and usually remain so upon incision.

1. **Skin:** - The skin is thin and loosely attached in this area. The hairs are scanty in this region.
2. **Subcutaneous fascia:** - Subcutaneous fascia (Superficial fascia) surrounds all the mammary parts of ventral abdomen and extends

laterally. Medially it blends with deep fascia at the linea alba. Superficial fascia contains the Cutaeneous Trunci muscle.

3. **Cutaeneous Trunci muscle:** - As a thin leaf it covers almost the entire dorsal, lateral and ventral walls of the thorax and abdomen. It begins caudally in the gluteal region, running forward and downward, covers the dorsal and lateral surfaces of the abdomen and thorax. It ends in the axilla and on the caudal border of the deep pectoral muscle. It lies in the superficial trunk fascia and do not attach the vertebral spines. The course of the fibers is slightly ventrocranial. It is principally a longitudinal muscle. The dorsal borders of the muscle on each side run parallel along the spines of lumber and thoracic vertebrae. Its ventral border crosses, in the fold of the flank, to the lateral and ventral abdominal wall.

4. **Deep Fascia:** - Deep fascia forms the abdominal tunic and it is bright yellowish white in colour. It is thin layer and closely attached to M. Obliques abdominis externus.

5. **Abdominal Muscles and their aponeurotic parts:** - From without inward the abdominal muscles are; Obliques abdominis externus, Obliques abdominis internus, Rectus abdominis and Tranversus abdominis. The abdominal muscles form much of the lateral and the entire ventral abdominal wall.

a. **Obliques abdominis externus- This** is an expensive sheet covering the ventral half of the lateral thoracic wall and the lateral part of the abdominal wall. The fibres of the External oblique muscle run caudoventrally, the caudal part being more horizontal than the cranial.

   Origin: - Muscle originates in two parts; the *pars costalis* arise as indistinct serrations in a caudally rising line from the middle parts of the 4^{th} or 5^{th} to 12^{th} rib, and the adjacent deep trunk fascia, which covers the external intercostal muscles. It is partly covered by the ventral edge of the M. Latissimus dorsi at its origin. The unserrated *pars lumbalis*

arise from the last rib and, in common with the *pars costalis* of the Obliques abdominis internus, from the principal lamina of the thoracolumbar fascia.

Insertion:- In the ventral abdominal wall, 6-8 c.m. from the midline in large dogs, It forms a wide aponeurosis, differentiated in to an abdominal and pelvic tendon separated by means of external inguinal ring. The abdoomainal tendon, is by far the largest part of the aponeurosis of the M. Obliques abdominis externus. It arises from *pars costalis* of the muscle. This flat tendon extends over the M. Rectus abdominis to the linea alba, where it unites with that of the opposite side. It extends caudally also to attach to the pectin ossis pubis. The pelvic tendon arises essentially from the *pars lumbalis* of M. Obliques abdominis externus. Ventrally the pelvic tendon is separated from abdominal tendon by sharp-edged saggital slit, the external inguinal ring. The lateral crus of the pelvic tendon meet the medial crus of the abdominal tendon to form, the cranial and caudal commissures of the superficial inguinal ring.

b. Obliques abdominis internus: - It is a flat muscle lying medial to M. Obliques abdominis externus in the lateral abdominal wall where it is almost completely covered by the external oblique muscle. Its fibers in general run cranioventraly and there by cross those of the external oblique muscle, approximately at right angle.

Origin: - Its fibers arise from the principal lamina of the thoracolumbar fascia caudal to the last rib, in common with the lumbar portion of M. Obliques abdominis externus. It originates mainly from the tuber coxae, M.Iliopsoas and the inguinal ligament.

Insertion: - The cranial part (*pars costalis*) proximally ending on the 13^{th} rib and on cartilage of 12^{th} rib. The middle portion (*pars abdominalis*) gives rise to a broad aponeurosis at the lateral border of M.rectus abdominis, it extends from the bend of the 12^{th} costal cartilage to the iliopectineal eminence. It forms the outer rectus sheath with

aponeurotic part of muscle obliques externus and ends on the linea alba. The caudal part (*pars inguinalis*) of the muscle which comes from the tuber coxae, by means of a short aponeurosis, and forms the inguinal ligament, sends in the inguinal region caudoventrally towards the linea alba.

C. Rectus abdominis: - It is a long, rather wide, flat muscle which extends, one on each side of thye linea Alba on the thoracic and abdomibal walls between the external and internal leaves of the rectus sheath, and runs from the first costal casrtilage to the pectin ossis pubis. Cranially, in large dogs, it is 7-8 c.m. broad, caudally it gradually narrows to 3.5-4 c.m. The fibers of the muscle course longitudinally and have tendinous inscriptions that divide the muscle in to a series of segment.

Origin: - It arises by a broad, flat tendon from the sternum and the first costal cartilage and rib. It also has a fleshy origin by means of a special serration from the sternal portion of the 9^{th} costal cartilage.

Insertion: - At its insertion each muscle unites with the tendon of origin of the M. pectenius and the pre pubic tendon. Medially these muscles of both side united by linea alba.

D. Tranversus Abdominis: - It is the deepest abdomoinal muscle and, like the oblique muscles, it is developed in to an extensive leaf which reaches a thickness of 2-4mm. in large dogs. It lies in the laterasl and ventral abdominal wall on the internal surface of the M. Obliques abdominis internus and adjacent costal cartilages.

Origin: - It arises from the 8^{th} costal cartilage to the last lumber transverse processes and the tuber coxae. The lumba part (*pars lumbalis*) arises by broad, short tendons from the transverse processe of all the lumbar vertebrae and the deepest division of thoracolumbar fascia. The costal part (*Pars Costalis*) arises muscularly on the medial sides of the 13^{th} and 12^{th} ribs and the 11^{th} to 8^{th} costal cartilages in such a way that its line of origin crosses that of diaphragm.

Insertion:-The end aponeurosis forms most of the inner leaf of the sheath of the rectus abdominis .it units inseparably at the linea Alba with the external leaf. In the costal region, it units only with part of the end aponeurosis of the M.Obliques abdominis internus.

E. **Parietal Peritoneum:** Parietal peritoneum lies just beneath the transverses abdominis muscle and it forms a layer of abdominal wall interiorly.

(2) MID - VENTRAL APPROACH

Site of Incision: - A 4-5 cm long, ventral midline abdominal skin incision was made over linea alba extending form umbilicus to a point half way between the umbilicus and brim of the pubis.

At the site of mid ventral surgical incision, following anatomically structures were encountered from outward within:

1. Skin
2. Subcutaneous Fascia
3. Deep fascia
4. Linea alba
5. Parietal Peritoneum
6. Viscera

Linea alba: - **It** is a midventral strip of collagenous tissue, which extends from the xiphoid process to the symphysis pubis. It serves for the main insertion of the abdominal transverse and external and internal oblique muscles. The medial borders of the right and left rectus muscle lies closely against its lateral borders. The linea alba contains a scar, the umbilicus (annulus umblicalis), a remnant of the umbilical ring and cord.

(3) FEMALE GENITAL ORGANS

The female genital organs consist of ovaries, uterine tubes, uterus, vagina and vulva.

Broad ligament: - The ovaries, oviduct and uterus are attached to the dorsolateral wall of the abdominal cavity and to the lateral walls of the pelvic cavity by paired double fold of peritoneum called the right and left broad ligaments. It contains the round ligament of them uterus in its free border. Cranially broad ligament attached by means of the suspensory ligament of the ovary to the junction of the middle and distal third of last rib. Morphologically broad ligament is divided in to three regions.

1) Mesovarium: - It is that part of the broad ligament which attaches the ovary to the dorsolateral region of abdominal wall. It contains the utero-ovarian vessels.

2) Mesosalpinx: - It is another double fold of peritoneum, extends laterally from the dorsal peritoneal layer of the mesovarium. It curves around the dorsal end ventrolateral borders of the ovary to attach to the medial surface of the broad ligament just dorsal to ovary. It encloses the ovary with in a small peritoneal cavity, the ovarian bursa. The entire uterine tube lies between the peritoneal layers of the mesosalpinx.

3) Mesometrium: - It begins at the cranial edge of the uterine horn, where it is continuous with the mesovarium, and extends caudally to a point where the peritoneum of the broad ligament reflects on to the bladder and the colon. It leaves the uterine horn, the body of uterus, the cervix and the cranial part of the vagina to attach along the abdominal and pelvic walls. It also contains cranial and caudal uterine arteries and veins.

The Ovaries: - The ovaries are paired oval organs, located in abdominal cavity caudal to the kidneys. In a large dog an ovary averages 1.5 cm.in length and 0.7 cm. in width.

Position and relation: - The left ovary is located approximately 12 cm. caudal to the middle of the 13th rib and 1-3 cm. caudal to the corresponding kidney. Typically it lies between the abdominal wall and the left colon, opposite to 3rd -4th lumbar vertebrae. The right ovary is located approximately 10 cm. caudal to the rib of the right side, between descending duodenum and the lateral abdominal wall, opposite to 3rd lumbar vertebrae. In addition to the mesovarium the ovary has two other ligamentus attachments. The suspensory ligament lies between the two layers of peritoneum and form cranial portion of the free border of the broad ligament. It is continued caudally by the proper ligament of the ovary, this in turn attaches to the cranial end of the uterine horn.

Vessels and nerves: - The ovaries are supplied with blood through ovarian artery, which arise from aorta. Usually the right ovarian artery arises slightly cranial to the left. In addition to supplying the ovary, it also supplies branches to kidney capsule and uterine tubes and uterus. Caudally the ovarian artery anastomoses with the uterine artery (branch of urogenital artery). The right and left ovarian veins have different terminations. The right vein drains into caudal venacava, where as left enters the left renal vein. The ovarian veins also anastomose with uterine veins. The nerve supply to the ovaries is via the sympathetic innervations reaches by way of renal and aortic plexuses.

The Uterine tubes (oviduct):-The uterine tubes are small and average 4-7 cm. in length. Each uterine tube is located between the peritoneal layers of the mesosalpinx and connects the peritoneal cavity with the uterine cavity. The ovarian extremity of the tube, the infundibulum, is open into the ovarian bursa through an opening called abdominal ostium. From the abdominal ostium, the uterine tube at first runs

craniolaterally between the ventral layers of the mesosalpinx, then it swings onto dorsal aspect of the suspensory ligament and runs in a tortuous manner caudomesially toward the ovary. At middle of ovary it curves caudolaterally toward the apex of the uterine horn, where it terminates into uterine horn via uterine osteum.

Vessels and nerves: - Uterine tubes are supplied by ovarian arteries and uterine arteries. The veins are satellites of the arteries. The nerve supplies derived from aortic and renal plexuses.

The Uterus: - The uterus consists of a neck (cervix uteri), a body (corpus uteri) and two horns (Cornua uteri). It is a tubular, Y-shaped organ which communicated with the oviducts cranially and the vagina caudally.The uterine horns averages 10 -14 cm. long and 0.5-1 cm. in diameter.The cervix (Cervix Uteri) average 1.5-2 c.m. long. A small caudal portion of the cervix may protrude 0.5-1 c.m. in to the vagina. The diameter of this intra vaginal cervix is approximately 0.8 c.m. The canal of cervix is directed caudoventrally from uterus to vagina. The cervix lies diagonally across the uterovaginal junction. The internal orifice of the cervical canal is facing directly dorsally; where as the external orifice is directed towards the vaginal floor.

Position and relation: - Dorsally the uterus is in contact with the descending colon, the psoas muscles, the transverse abdominal muscle, and the ureters.Ventrally it contacts the urinary bladder, the greater omentum, the jejunum, the ileum, and the descending duodenum. The recto uterine space, a potential peritoneal cavity between the rectum and the uterus is continuous with the Para rectal fossa. The vesicouterine space, between the urinary bladder and the uterus (with its attached broad ligament), is separated from the paravesical fossa by double peritoneal folds, the lateral ligaments of the bladder.

Vessels and nerves: - The uterus is supplied with arterial blood via the ovarian and uterine arteries. The uterine and ovarian vein follows a

course similar to that of the arteries, except at their terminations. The right ovarian vein empties into the caudal venacava at the level of the right ovary, where as the left enters the left renal vein. The uterus receives sympathetic and visceral afferent fibers through the hypogastric plexus and parasympathetic and visceral afferent fibers via the pelvic nerves.

The Vagina: - The vagina is a musculomembranous, highly dilatable canal extending from the uterus to the vulva. Cranially the vagina is limited by the fornix and the intravaginal cervix. Caudally the vagina ends just cranial to the urethral opening.

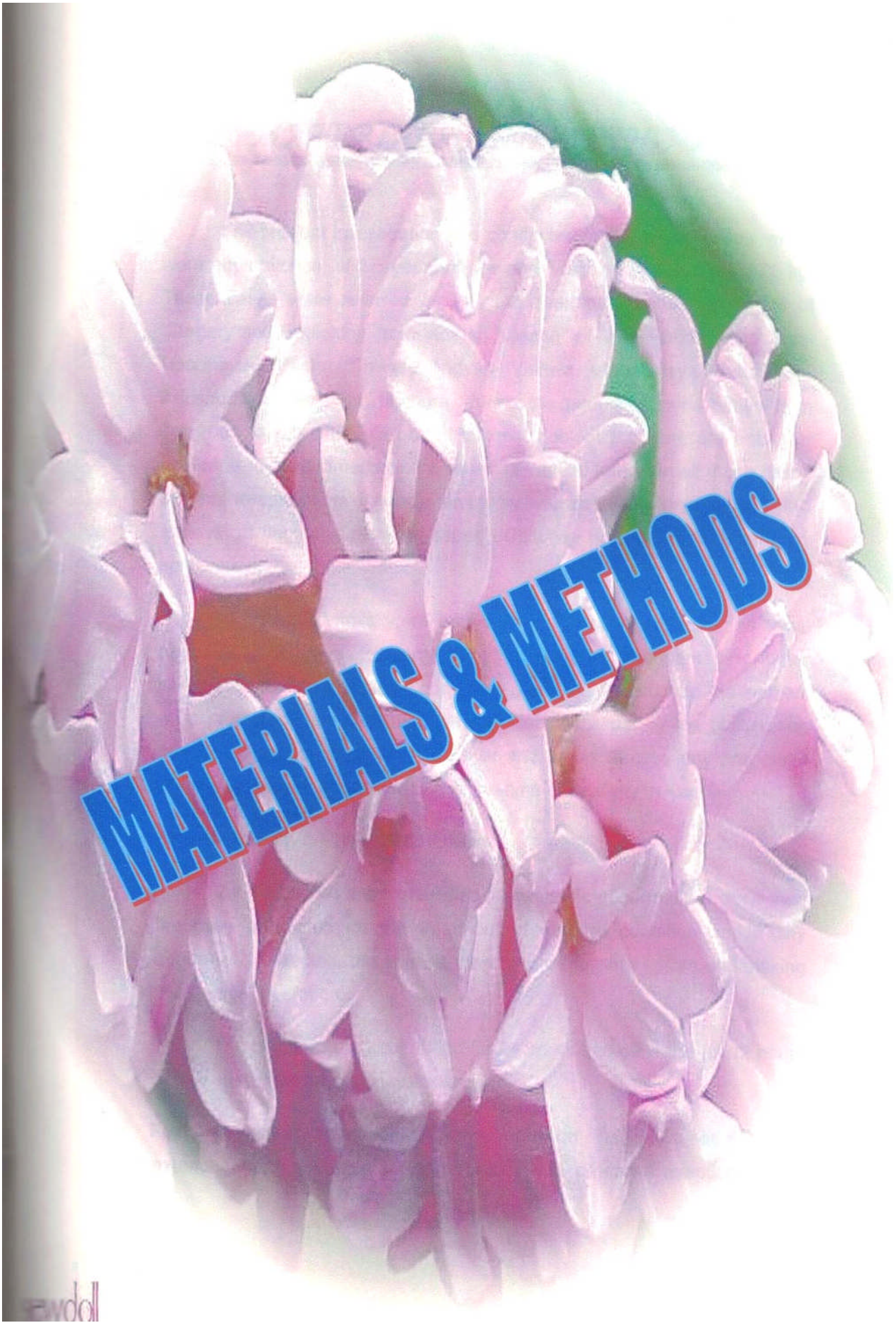
Position and relation: - The cranial portion of vagina is covered dorsally by peritoneum which reflects onto the colon, forming the recto uterine pouch ventrally, this portion of the vagina has a peritoneal covering which reflects onto the bladder, forming the vesicouterine pouch. Laterally, the dorsal and ventral peritoneal coverings of the vagina fuse and become part of the broad ligaments. The caudal half of the vagina is retroperitoneal, being connected dorsally to the rectum and ventrally to the urethra by means of loose connective tissue. Laterally, the caudal part of vagina is related to the vaginal blood vessels and nerves and to the ureters. The right and left ureters, with there peritoneal coverings, cross the lateral surface of uterovaginal junction.

Vessels and nerves: - Arterial blood is supplied to the vagina via the vaginal artery, a branch of pudendal artery. In addition to its vaginal distribution, the artery also supplies branches to the urethra and vestibule. The vaginal veins are satellites of the arteries and drain in to the internal pudendal veins. The vagina is innervated by sympathetic and parasympathetic nerves from pelvic plexus and by sensory afferent fibers via the pudendal nerve.

The Vulva: - The vulva or external genitalia consist of three parts; vestibule, clitoris and labia. The vestibule is the space connecting the

vagina with the external genital opening. The urethral tubercle is a ridge like projection on the ventral floor of the vestibule, near the vaginovestibular junction. It contains the external urethral orifice. In a non pregnant mature large dog the external vulvar opening is 3 c.m. long. The distance from the ventral commissure of the vulva to the urethral opening is 5 c.m. and the diameter of the vaginovestibular junction is 1.5- 2 c.m. The clitoris, the homologue of the male penis, is composed of paired roots, a body and a glans.The labia or lips, from the external boundary of the vulva. The labia fuse above and below the vulvar cleft to form dorsal and ventral vulvar commissure.

Vessels and nerves: - The external genitalia and urethra of the female are supplied with blood through the vaginal and the external and internal pudendal arteries and veins. Autonomic innervation to the external genitalia and urethra in the female is through the hypogastric and pelvic nerves.


MATERIALS & METHODS

CHAPTER-4

MATERIALS AND METHODS

The present investigation was conducted on 16 clinical cases, which were adult bitches, of 2-6 years of age and weighing between 14 to 25 kg. These cases were referred for surgical treatment to the department of 'Surgery and Radiology' from 'Kothari Hospital' of U. P. Pandit Deen Dayal Upadhayay Pashu Chikitsa Vigyan Vishwa Vidhayalaya Evam Go-Anusandhan Sansthan, Mathura, during the period of investigation from Jan.2005 to July 2006. Out of 16 bitches (Table 1.), 8 were presented for neutering, 5 for pyometra and 3 cases of vaginal tumor. In all these cases surgical treatment by ovariohysterectomy was decided along with surgical removal of vaginal tumors (in cases of bitches having vaginal tumor) by episiotomy.

All these animals were admitted and kept in separate individual cages for 12 days to carry out the surgical treatment and other investigations. Strict hygienic conditions were maintained and the animals were protected from inclement weather conditions during the observation period. All bitches were provided milk and bread twice a day and clean whole some water ad libidum during the time of captivity in Kennel.

PRE-OPERATIVE PREPARATIONS:

All the bitches were examined for systemic abnormalities other then the case presented for the treatment of surgical condition and if any abnormality detected, the animals were treated for that before undertaking the surgical operation.

The bitches were kept off-fed for 24 hours and off water for 12 hours before submitted for ovariohysterectomy.

For mid-ventral approach- A large area on the mid ventral region extending from prepubic to xiphisternum was prepared aseptically. The

Table-1. Layout of clinical cases for ovariohysterectomy (OH).

Groups of Animals	sub groups (sutures used)	animal No.	Body wt (kg)	Age (year)	Case presented for
Group A (Right flank approach with subcuticular suture)	(a) Vicryl # 2-0	A1	16	2.5	Neutering
		A2	22	5	Pyometera
	(b) Chromic gut # 2-0	A3	23	2	Neutering
		A4	14	3	Vaginal tumors
Group B (Right flank approach with external skin suture)	(a) Vicryl # 2-0	A5	22	6	Pyometera
		A6	18	3.5	Neutering
	(b) Chromic gut # 2-0	A7	23	4.5	Pyometera
		A8	18	4	Neutering
Group C (Mid-ventral approach with subcuticular suture)	(a) Vicryl # 2-0	A9	15	2	Neutering
		A10	26	3.5	Vaginal tumors
	(b) Chromic gut # 2-0	A11	19	4	Pyometera
		A12	16	2	Neutering
Group D (Mid-ventral approach with external skin suture)	(a) Vicryl # 2-0	A13	15	3	Vaginal tumors
		A14	23	2.5	Neutering
	(b) Chromic gut # 2-0	A15	25	5.5	Pyometera
		A16	14	3	Neutering

hairs were removed from the area by clipping and shaving. The site was scrubbed with cetrimide (3%) lotion and then washed with clean water; the procedure was repeated 4 times. Finally the area was painted with 5% povidone iodine using sterilized gauze piece.

For right flank approach- A large area on the right flank region extending from fold of stifle to caudal border of the last rib was prepared aseptically as mentioned above for the mid-ventral approach.

DESIGN OF EXPERIMENT:

The lay out of experimental design for all 16 clinical cases subjected to elective ovariohysterectomy was basically to evaluate the surgical techniques of laparotomy for performing ovariohysterectomy. These 16 bitches were randomly categeorized into 4 groups, for 4 replications in each group and these groups were further subdivided into 2 subgroups based on suture material used and consisting of 2 animals in each subgroup, for surgical maneuvers viz; mid-ventral and right flank laparotomy and ovariohysterectomy

Group A: Ovariohysterectomy was done through right flank, and the closure of the abdominal wound after operation was carried out in layers. The transverse muscle with its attached peritoneum was sutured first, applying simple continuous sutures. The internal oblique and external oblique muscles were sutured together with simple continuous sutures. Finally the subcuticular suture was applied for skin apposition. Vicryl (P.G.A. 910) # 2-0 was utilized in A1 and A2, while Chromic catgut # 2-0 in A3 and A4 animals as suture material in all the layers as well as for ovarian and uterine stump ligation.

Group B: In this group the ovariohysterectomy was done through right flank, and the closure of the abdominal wound after operation was carried out as in group 'A' using Vicryl (P.G.A. 910) # 2-0 in A5, A6 animals and Chromic catgut # 2-0 in A7, A8 animals. However, finally skin apposition was done with interrupted horizontal mattress suture using Nylon.

Group C: Ovariohysterectomy was done through midventral approach and the closure of the abdominal wound after operation was carried out in layers. The peritoneum was sutured first, applying simple continuous suture. The linea alba was also sutured with simple continuous sutures. Finally the subcuticular suture was applied in simple continuous pattern for skin apposition. Vicryl (P.G.A. 910) # 2-0 was utilized in A9 and A10, while Chromic catgut # 2-0 in A11 and A12 animals as suture materials for all the purposes.

Group D: Ovariohysterectomy was done through midventral approach and the closure of the abdominal wound after operation was carried out in layers, the peritoneum was sutured first, applying simple continuous sutures, as in group 'C' animals using Vicryl (P.G.A. 910) # 2-0 in A13, A14 animals and Chromic catgut # 2-0 in A15, A16 animals, however finally skin apposition was done with interrupted horizontal mattress suture using Nylon.

ANAESTHESIA AND RESTRAINT

Keeping in mind the purpose of present study, the ovariohysterectomy (OH) of these animals was done under general anaesthesia. The bitches were premedicated with atropine sulphate (0.044mg/kg b.wt.) and xylazine hydrochloride (0.5mg/kg b.wt.) administered intramuscularly. After 10 minutes a combination of xylazine hydrochloride and ketamine hydrochloride (1:3) was administered intravenously till effect, followed by maintenance of general anaesthesia with the intravenous incremental dosages of same combination as per the need.

The animals were restrained either in dorsal recumbency for mid-ventral approach or in left lateral recumbency for right flank approach.

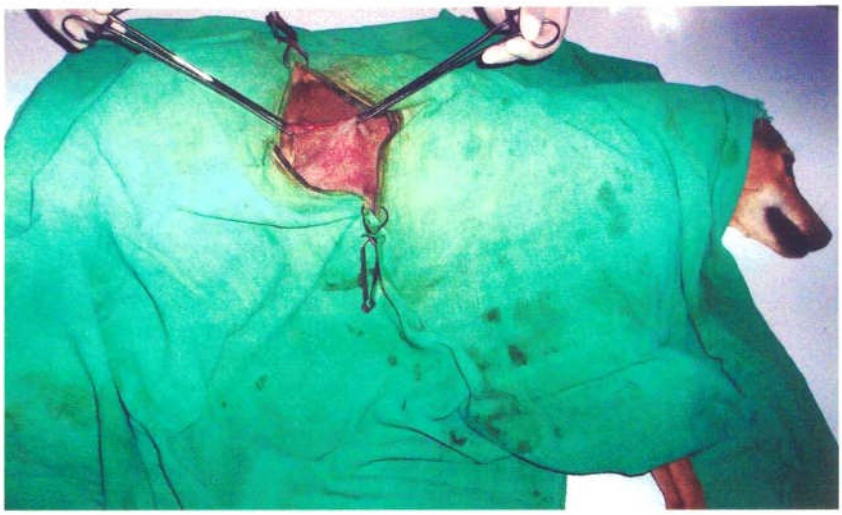


Photo-1. Right flank approach for ovariohysterectomy (incision site).

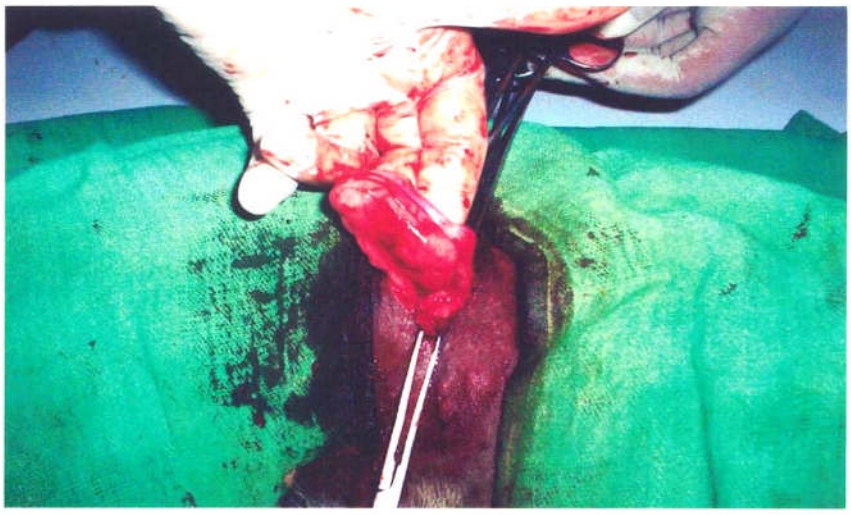


Photo-2. Exteriorization of right ovarian stump.

TECHNIQUE OF OVARO-HYSTERECTOMY

(A) OPERATIVE PROCEDURE ADOPTED IN GROUP "A" and "B":

(1) **Site of incision-** For performing ovariohysterectomy through right flank : A horizontal, mid lateral rectus abdominal skin incision was made at right lower flank started from a point about 4-6 c.m. lateral to the umbilicus and parallel to midline, and about 1.5-2 c.m. above the caudal abdominal mammary teat, extending about 3-4 c.m. caudally.

(2) **Position of Animal-**The animals were positioned in left lateral recumbency on horizontal operation table and ventral aspect facing towards the surgeon. The fore and hind limbs were held in extended position in order to expose maximum area of flank.

(3) **Technique of operation-**

(a) **Laparotomy -**The incision was made at the proposed site of incision by using B.P. blade # 23 and skin was cut. The underlying structures i.e. subcutaneous tissue, abdominal muscles (external obliquous; internal obliquous and transversus abdominis muscles) and perital peritoneum were incised layer by layer using B.P. blade # 11, while holding the structures with the help of Allis tissue forceps. As soon as the peritoneum was incised, abdominal cavity was exposed.

(b) **Ovariohysterectomy-** As soon as the abdominal cavity was exposed, the right ovary and cranial end of right uterine horn was visualized through the laparotomy wound with the help of right hand index finger, above the descending duodenum. The ovarian arterio-venous plexus was located and exteriorized after loosening its suspensory ligament and a 'window' was made in mesovarium immediately caudal to the arterio-venous plexus with the help of mosquito hemostat. The ovarian arterio-venous plexus was double clamped with straight hemostatic forceps; the third clamp was placed over the proper ovarian ligament between the ovary and uterine horn. A circumferential suture was

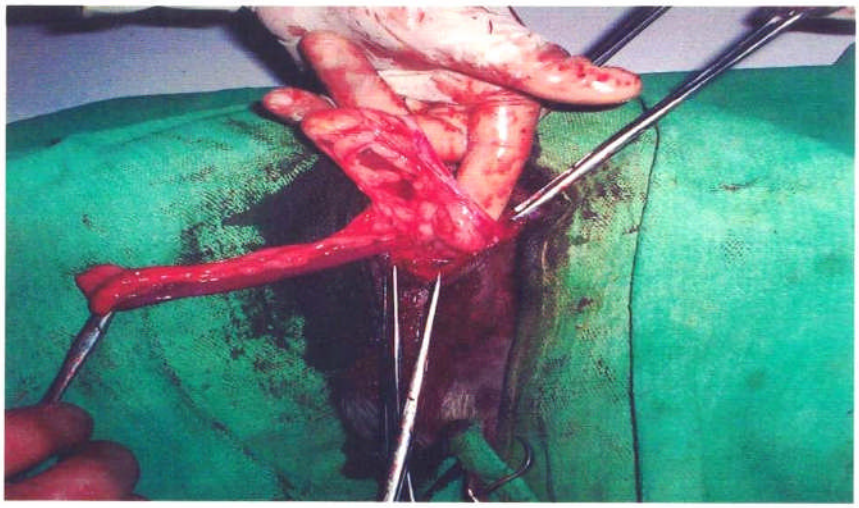
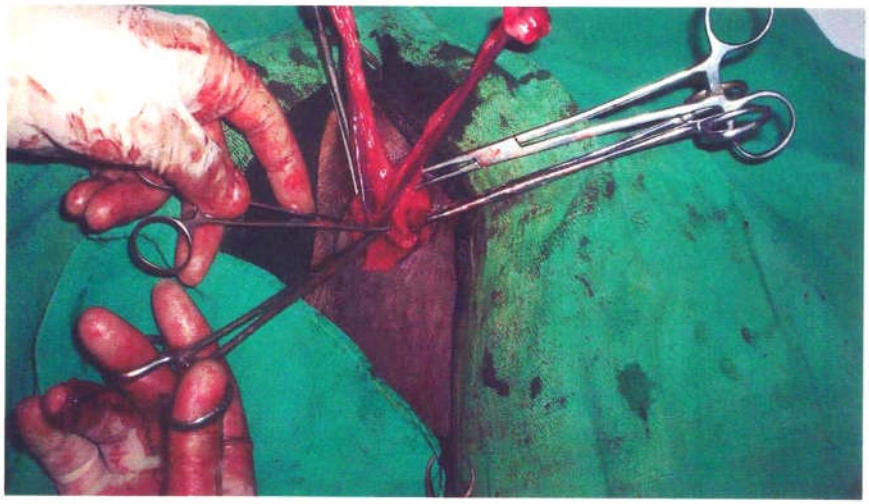


Photo-3. Exteriorization of left ovarian stump.



loosely placed around the ovarian pedicle just behind the proximal clamp; the clamp was removed as the circumferential ligature was tightened so that the circumferential ligature lies in the groove of crushed tissue created by the clamp. Then the pedicle was severed between the middle clamp and the ovary. The clamp was removed from the pedicle and inspected for bleeding before repositing it into the abdominal cavity. The left uterine horn was located by following the right uterine horn distally to the bifurcation and then the left horn was followed up to the left ovary. The ligation procedure was repeated on the left ovarian pedicle as in right ovarian pedicle. The broad ligament was grasped and detached from uterine horns, taking care to avoid uterine artery and vein. When the uterine horns are free, the uterine body was exteriorized and three clamps were placed immediately proximal to the cervix by taking care to avoid cutting rather than crushing of uterine body. A loose transfixation circumferential ligature was placed between distal clamp and cervix, while the uterine artery and vein were ligated individually. The ligature was tightened after removal of distal clamp and the uterine body was severed between middle clamp and the proximal clamp. The uterine stump was checked for bleeding after loosening the remaining clamp. The cut end of uterine stump was sutured using Parker-kerr suture over the remaining clamp and then clamp was removed together with tightening the suture. After completion of the suture the uterine stump replaced into the abdomen.

(4) **Suturing technique-**The suturing of laparotomy wound was done as has been described earlier for the groups 'A' and 'B'. The povidone iodine solution was painted over the suture line and the adjoining skin.

(5) **Sealing of the incision-** The cutaneous wound was dressed aseptically by using 5% povidone iodine solution and protective dressing was done with sterilized napkin pad with the help of adhesive tape.

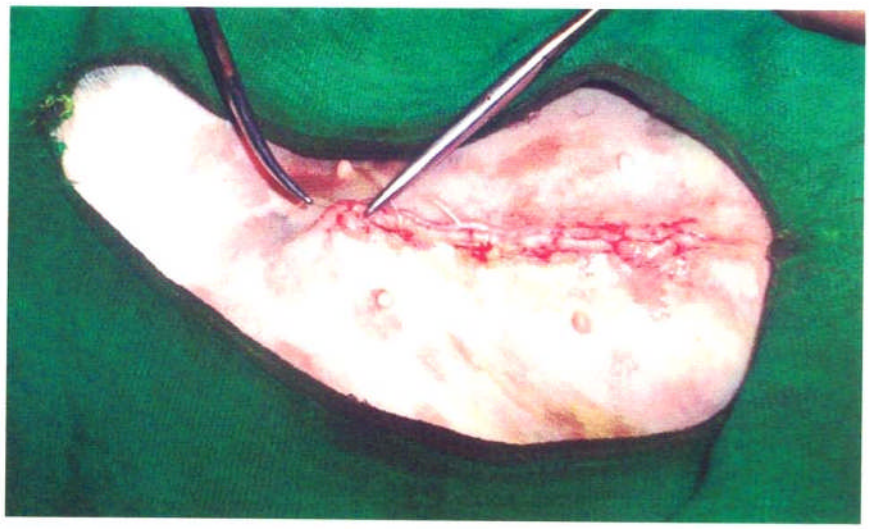


Photo-5. Mid-ventral approach for ovariohysterectomy (completed).

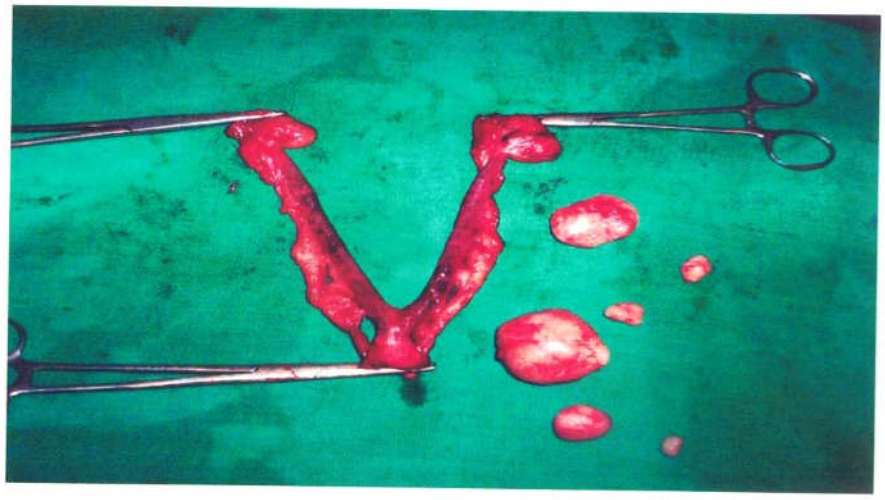


Photo-6. Removed ovaries, uterus and vaginal tumors.

(B) OPERATIVE PROCEDURE ADOPTED IN GROUP "C" and "D":

(1) Site of incision- For performing ovariohysterectomy through linea alba: A 4-5 cm long, ventral midline abdominal skin incision was made over linea alba extending form umbilicus to a point half way between the umbilicus and brim of the pubis.

(2) Position of the animal- The bitches were positioned lying on dorsal recumbency over horizontal operation table with neck extending forward, and the fore and hind limbs were tied on either side of operation table with slight flexion.

(3) Technique of operation

(a) Laparotomy - The incision was made at the proposed site of incision by using B.P. blade # 23 and skin was cut. The underlying structures i.e. subcutaneous tissue, linea alba, peritoneum and falciform ligament were incised by using B.P. blade # 11, while holding the structures with the help of Allis tissue forceps. As soon as the peritoneum was incised, abdominal cavity was exposed.

(b) Ovariohysterectomy- As soon as the abdominal cavity was exposed, the right uterine horn was located and grasped by passing an index finger through the laparotomy wound, under the urinary bladder and over the descending colon for exteriorization, separation, ligation and cutting and removal of the ovaries, along with uterine horns and uterine body the same procedure was adopted as in group 'A' and 'B'.

(4) Suturing technique-The suturing of laparotomy wound was done as has been described earlier for the groups 'C' and 'D'. The betadine solution was painted over the suture line and the adjoining skin.

(5) Sealing of the wound- The cutaneous wound was dressed aseptically as described for group 'A' and 'B'

(1) CLINICAL OBSERVATIONS:

All the bitches were examined for any physiological abnormalities, other then the case presented for the treatment of surgical condition and if any abnormality detected, the animals were treated for the same before undertaking the surgical operation. The temperature, pulse rate and respiration rates were recorded before operation, day of operation and after operation on 1st (24hrs), 2nd, 3rd 5th, 7th and 10th days of operation in all the bitches. The average of morning and evening were calculated and presented as reading of the day for temperature, pulse and respiration rates. The temperature, heart and respiration rates were taken preoperatively as follows:

- Heart rate was noted by stethoscope placed on left side of chest.
- Respiration was recorded by closely observing the ribs and abdominal movements. Observations were recorded in per minutes.
- Temperature was recorded carefully by inserting the thermometer into rectum at a constant & proper depth. Temperature was recorded in ^{0}F.

(2) Observations during operation:

(A) Duration of operation- The time taken from surgical skin incision to completion of skin closure is recorded in minutes as duration of operation. It depends upon technique of exteriorization of ovary and uterine horn and time taken in ligation procedures and closure of laparotomy wound due to suture techniques adopted in different groups of animals.

(B) Amount of hemorrhage during entire procedure- Evaluation of hemorrhage during operative procedure was done by using Napkin pads. These Napkin pads were sterilized in autoclave, and were used for soaking blood in entire procedure. Weight of each napkin pads was

recorded and one napkin at one time was used for soaking the blood and collected in air tight polythene bag. After completion of procedure the used napkins were weighed. The weight of napkins used subtracted from the weight of soaked napkins to calculate the amount of hemorrhage in grams.

Amount of hemorrhage (gm) = weight of soaked napkins – weight of these napkins before use

(3) Observations After operation: The postoperative parameters were recorded individually at 1st (24 hrs), 2nd, 3rd, 5th, 7th and 10th day postoperatively as follows:

(A) Status of wound healing- Status of wound healing was observed on the basis of following parameters

(a) Oedema/swelling of the suture line/ incision site

Oedema of suture line was graded from '0' to '4'as follows:

1- **Normal as pre operative**- No oedema.- **(0)**

2- **Slight swelling-** with no tension on the suture line- **(1)**

3- **Slight to Mild swelling-** With slight tension on the suture line-**(2)**

4- **Moderate swelling-** Oedema occurs with moderate tension on suture line and surrounding area- **(3)**

5- **Sever swelling**- Severe swelling with severe tension on suture line and embedding of sutures in the oedematus tissue **-(4)**

(b) Pain on palpation at the site of incision

Pain at the site of incision was observed by touching and pressing the site and the degree of reactions was shown by the animal and graded from '0' to '4' as mentioned below:

1- **Normal as pre operative-**No reaction-**(0)**

2- **Slight pain-** Animal allow to touch and permit pressure on the suture line without any noticeable body reaction-**(1)**

3- **Slight to Mild pain-** Animal allow touching the suture line with gentle pressure and reacts slightly turning the head towards incision site. **-(2)**

4- **Moderate pain-** On touching the suture line animal reacts quickly by crying-**(3)**

5- **Severe pain-** On trying to touch the suture line animal reacts violently and forcefully and did not allow palpation of site of suture line-**(4)**

(c) Warmth of the incision line

Warmth of the incision site was compared with normal body surface by taking temperature of normal body surface and of the incision site with the help of touch strip thermometer and their difference was graded and recorded in ^{0}F as below:

The pre operative skin warmth taken as 0 ^{0}F

Normal warmth	-Difference	≤ 1 ^{0}F
Mild warmth	- Difference	≥ 1-2 ^{0}F
Moderate warmth	- Difference	≤ 2-3 ^{0}F
Severe warmth	- Difference	≤ 3-4 ^{0}F

(B) Post-operative complications- The observations were taken for any other postoperative complications at different time intervals with status of wound healing for each individual as below:

1. Postoperative hemorrhage.
2. Exudation of cutaneous wound was observed while by observing the moistness and gently pressing the sides of the incision line for presence of any exudates while dressing the wound.

3. Suture biting.
4. Suture dehiscence.
5. Evisceration.
6. Peritonitis and death.

(4) POSTOPERATIVE FOLLOW-UP:

All the clinical animals were administered inj. Megapen (Ampicillin and Cloxacellin) @ 10-12 mg/kg. intravenously for 5 days twice daily and inj. Melonex (Meloxicam) @ 0.2-0.4 mg/kg. intramuscularly for 3 days twice daily followed by antiseptic dressing of suture line with povidone iodine solution (5%) and wound was covered with sterilized napkin pad with the help of adhesive tape.

Dressing of suture line was repeated on the days of postoperative observation periods viz. 1st (24hrs), 2nd, 3rd, 5th, 7th, and 10th day and local observations of the site of incision line were recorded. The external skin sutures were removed on 10th postoperative day and betadine solution was applied.

(5) GENERAL BODY CONDITION:

The animals were evaluated for their general body conditions on the basis of appetite, feed and water intake, urination and defecation during the observational periods of present study.

(6) COST OF EXPENDITURE INCURRED:

The total cost of operation was computed on the bases of cost of following items

(1.) Cost of anaesthesia: The total quantity of anaesthetic agents used according to body wt and duration of operation required for completion of operation was recorded for estimating the total cost of operation..

(2.) Cost of suture material: The length of the absorbable suture and non absorbable suture material used was recorded in centimeters in each animal as its cost is to be calculated for estimating the total cost of operation. The suture material, particularly absorbable was shared by more than one animal, hence was divided by number of animals in order to draw the cost of suture material used in one animal.

(3.) Cost of site preparation: The cost of site preparation was also recoded based on the antiseptics utilized.

(4.) Cost of medicines used post operatively was recorded based on need based use of analgesics, antibiotics and dressing material.

(5.) Cost of man power technical hours used was not considered.

(7) HAEMATOLOGICAL EXAMINATION:

Collection of blood sample: The blood samples were collected aseptically on preoperative and postoperative i.e. on 1^{st} (24hrs), 2^{nd}, 3^{rd}, 5^{th}, 7^{th} and 10^{th} day after operation, from the cephalic/recurrent tarsal vein by a 5 ml sterile disposable syringe and #22 BWG hypodermic needle, out of 2 ml, 0.5 ml blood was taken in clean, dry and sterile test tube containing EDTA at the rate of 1mg/ml of blood and remaining 1.5 ml blood was taken in clean, dry and sterile test tube for taking the serum for various biochemical examination.

Blood smear was prepared from a drop of fresh blood by the routine method and stained with Leishman's stain for differential leukocyte count (DLC). The results were expressed in percentage (%) for each leucocytes type.

Total leucocyte count (TLC) in thousand/cu. mm. and differential leucocyte count (DLC) were performed as per the method described by Benjamin (1986). Packed cell volume (PCV) in percentage (%) and the haemoglobin (Hb) in g/100ml, were performed by Wintrobe's method and Sohli's method.

(8) BIOCHEMICAL EXAMINATION:

Biochemical tests for serum glucose, serum creatinine, acid phosphatase, alkaline phosphatase were performed, on pre operative and postoperative i.e. on 1st (24hrs), 2nd, 3rd, 5th, 7th and 10th day after operation, with the serum using span diagnostic semi automated kit*.

i) Serum glucose

The serum glucose was estimated by using span diagnostic semi automatic kit for glucose, employing End-point-O-toluidine method, using spectrophotometer. The O.D was measured at 505 nm in spectrophotometer. The results were expressed in mg/100 ml of blood.

ii) Serum creatinine

Serum creatinine was estimated by using span diagnostic semi automatic kit for creatinine, employing alkaline picrate method.

Creatinine in a protein free solution reacts with alkaline picrate and produces a red coloured complex. The O.D was measured at 520 nm in spectrophotometer. The results were expressed in mg /100 ml of blood.

(9) STATISTICAL METHODS:

The data obtained in the research work were statistically analysed and compared using standard formulae given for mean, standard error and analysis of variance as per the procedures explained by Snedecor and Cochran (1967). Multiple comparisons of the means were done by using ANOVA and Duncan's new multiple range test (DMRT).

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* Span Diagnostics Ltd. Plot No. 336, 338, 340, Road No.3, G.I.D.C., SACHIN 394230 (Surat)


RESULTS

CHAPTER-5

RESULTS

(1) Clinical Observations:

(a) Heart rate:

The observations on the heart rate for different groups of animals are presented in Table-2. A nonsignificant difference was observed in heart rate in between different groups of bitches at different time intervals and within the group in different groups at different time intervals. However a nonsignificant decrease in heart rate at day of operation and increase in heart rate at 1st (24hrs) day after operation in all groups of animals was observed.

(b) Respiration rate:

The observations on the respiration rate for different groups of animals are presented in Table-3. A nonsignificant difference was observed in respiration rate in between different groups of bitches at different time intervals and within the group in different groups at different time intervals. However a nonsignificant decrease in respiration rate at day of operation in all groups of animal and nonsignificant increase in respiration rate upto maximum at 1st (24hrs) day after operation in group A and B while in group C and D it increased up to 2nd postoperative day.

(c) Rectal temperature:

The observations on the rectal temperature (^{0}F) for different groups of animals are presented in Table-4. No significant differences in the values of rectal temperatures were recorded in between different groups of animals at different postoperative periods within the groups. Only minor fluctuations in the body temperature were recorded.

Table 2.

Mean values ± SE of Heart rate (per minute) in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group A (subcuticular suture)	group B (external skin suture)
preoperative	89.00 ± 6.09	84.25 ± 5.60	85.50 ± 3.77	85.00 ± 6.81
day of operation	81.25 ± 6.18	79.50 ± 5.20	76.50 ± 3.09	77.25 ± 6.49
1st day (24hrs)	94.50 ± 5.26	92.50 ± 4.33	89.75 ± 3.66	87.50 ± 5.86
2nd day (48hrs)	91.75 ± 5.58	92.00 ± 4.84	86.50 ± 3.86	85.00 ± 5.43
3rd day	90.25 ± 6.04	89.50 ± 5.05	84.50 ± 3.52	83.75 ± 4.02
5th day	88.25 ± 6.27	83.25 ± 5.94	84.25 ± 3.90	83.75 ± 6.30
7th day	88.25 ± 6.56	83.75 ± 6.08	83.50 ± 3.77	84.25 ± 5.43
10th day	89.50 ± 6.35	84.75 ± 5.80	86.00 ± 3.18	83.25 ± 5.18

1. Values are mean ± SE of four replicates.

Table 3.

Mean values ± SE of Respiration rate (per minute) in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group A (subcuticular suture)	group B (external skin suture)
preoperative	25.25 ± 0.75	26.00 ± 0.40	27.00 ± 0.91	25.75 ± 0.75
day of operation	23.75 ± 0.47	24.50 ± 0.28	25.00 ± 0.57	24.75 ± 0.75
1st day (24hrs)	28.25 ± 1.10	28.25 ± 1.10	26.50 ± 0.64	26.75 ± 0.25
2nd day (48hrs)	27.25 ± 1.31	27.500 ± 0.64	27.43 ± 0.43	27.81 ± 0.49
3rd day	26.75 ± 1.10	26.50 ± 0.64	27.25 ± 0.75	26.75 ± 0.75
5th day	25.50 ± 0.86	26.25 ± 0.47	26.75 ± 0.85	26.25 ± 0.62
7th day	24.25 ± 0.94	25.75 ± 0.85	26.75 ± 0.75	26.25 ± 0.62
10th day	25.00 ± 0.70	26.75 ± 0.47	26.25 ± 0.62	26.25 ± 0.62

1. Values are mean ± SE of four replicates.

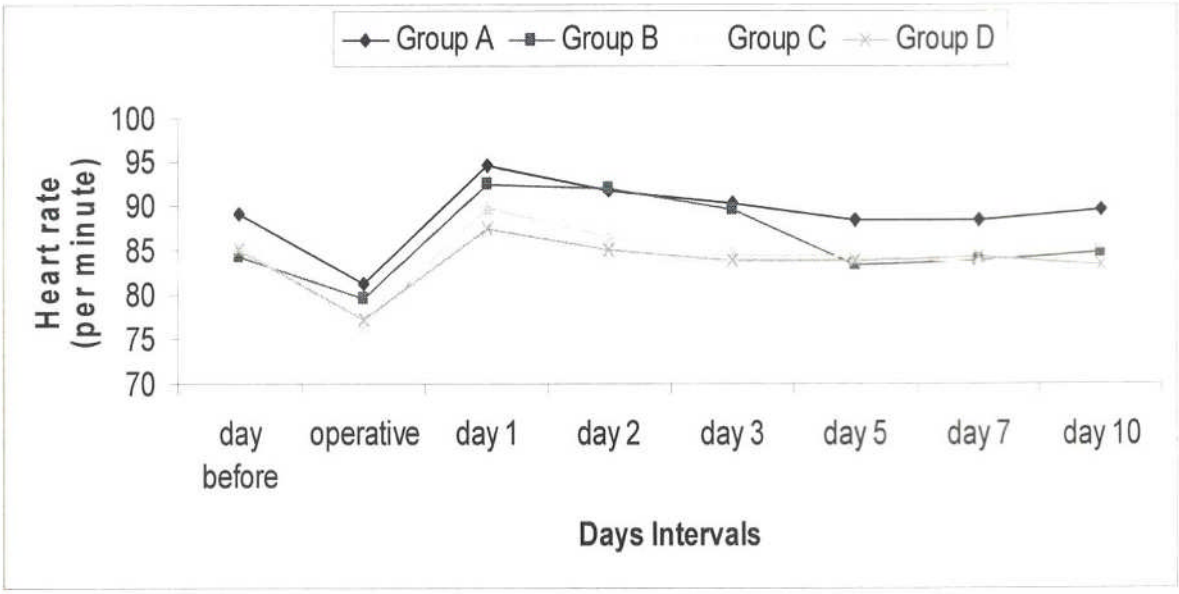


Fig.-1: Heart rate (per minute) in different groups of bitches at different intervals.

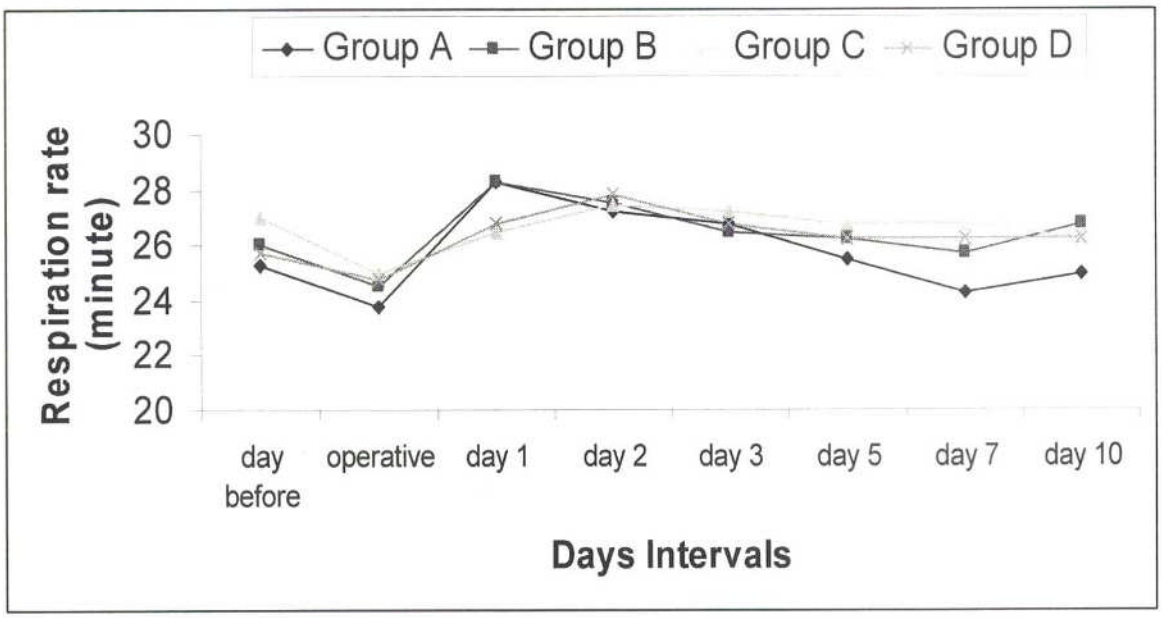


Fig.-2: Respiration rate (per minute) in different groups of bitches at different intervals.

Table 4.

Mean values ± SE of Rectal temperature (^{0}F) in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group A (subcuticular suture)	group B (external skin suture)
preoperative	102.35 ± 0.30	101.70 ± 0.47	102.10 ± 0.77	102.15 ± 0.83
operative	101.50 ± 0.47	101.95 ± 0.59	101.80 ± 0.43	101.85 ± 0.71
1st day (24hrs)	102.00 ± 0.74	102.00 ± 0.58	102.00 ± 0.58	101.95 ± 0.59
2nd day	101.95 ± 0.59	102.10 ± 0.57	102.00 ± 0.58	102.30 ± 0.34
3rd day	102.10 ± 0.57	101.50 ± 0.47	102.30 ± 0.38	101.80 ± 0.43
5th day	101.95 ± 0.59	102.10 ± 0.57	102.00 ± 0.58	102.25 ± 0.41
7th day	102.10 ± 0.57	102.20 ± 0.63	101.95 ± 0.59	102.35 ± 0.64
10th day	101.85 ± 0.71	102.20 ± 0.32	102.10 ± 0.57	102.20 ± 0.63

1. Values are mean ± SE of four replicates.

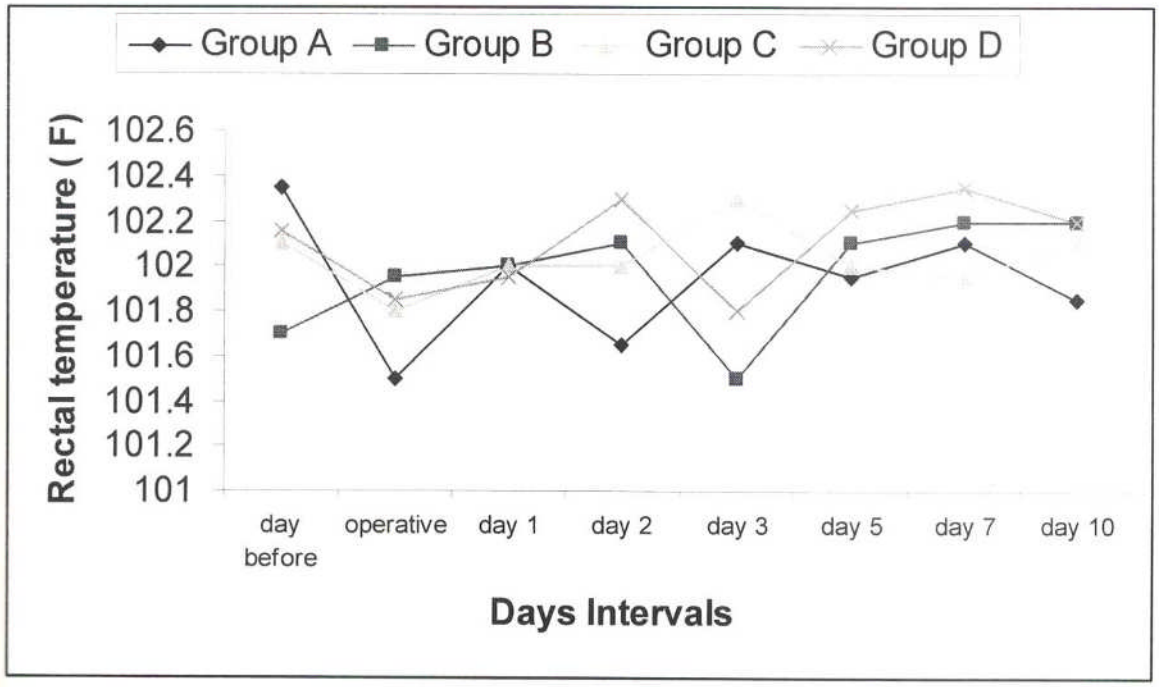


Fig.-3: Rectal temperature (^{0}F) in different groups of bitches at different intervals.

(2) Observation on various parameters during operation

(a) Duration of operation:

The time taken for surgery in different groups of bitches is presented in Table-5. A significant ($P< 0.05$) more time was taken for completion of operation in group C and D in comparison to A and B groups of animals. However, there was no significant difference between group A (31.25 ± 1.99) and B (32.25 ± 1.61) and group C (39.00 ± 2.14) and D (44.25 ± 2.17), respectively.

In present study the animals of group D which underwent ovariohysterectomy through linea alba (midline approach) with external skin sutures had highest mean time for completion of surgery followed by group C, B, and A in decreasing order.

(b) Amount of hemorrhage during operation:

The intra-operative hemorrhages during ovariohysterectomy in different groups of animals are presented in Table-5. There was a significant ($P< 0.01$) more amount of hemorrhage recorded in A and B groups of bitches in comparison to C and D groups of bitches. However, there was no significant difference between group A (50.00 ± 2.91) and B (51.25 ± 1.79) and group C (23.00 ± 2.19) and D (25.25 ± 1.65), respectively.

(3) Observation on various parameters after operation

(i) Status of wound healing:

(a) Inflammatory oedema at cutaneous incision site:

The observations with regard to the degree of inflammatory oedema at incision site for different groups of animals are presented in Table-6. A significant ($P< 0.05$) increase in cutaneous oedema at 1st day (24hrs) was recorded in all the groups of bitches in comparison to normal status of

Table 5.

Mean Values ± SE of Time of surgery and Intra-operative hemorrhage in different groups of bitches.

Parameters	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
Time of surgery (minute)	31.25_a ± 2.25	32.25_a ± 1.61	39.00_b ± 2.14	44.25_b ± 2.17
Intra-operative hemorrhage (gm)	50.00_b ± 2.19	51.25_b ± 1.79	23.00_a ± 2.19	25.25_a ± 1.65

1. Values are mean ± SE of four replicates.
2. Values sub scribed with same alphabets do not differ significantly ($P<0.05$) between groups at different time intervals.
3. Small letters are used to differentiate between groups at different time intervals.

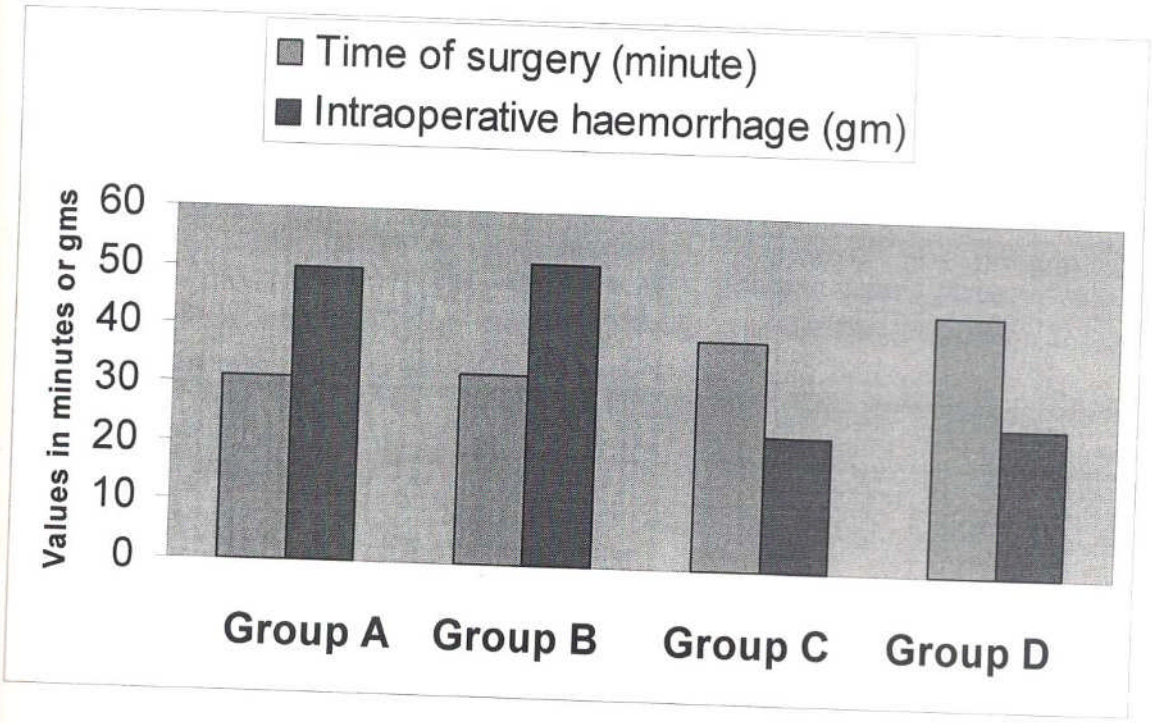


Fig.-4: Duration of operation and intra-operative hemorrhage in different groups of bitches.

Table 6.

Mean Values ± SE of Cutaneous oedema score in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
1st day (24hrs)	2.25_{c} ± 0.25	2.50_{cd} ± 0.28	2.25_{b} ± 0.25	2.00_{b} ± 0.00
2nd day (48hrs)	3.25_{d} ± 0.25	3.00_{d} ± 0.40	2.50_{b} ± 0.28	2.50_{b} ± 0.28
3rd day	3.00_{d} ± 0.00	2.75_{d} ± 0.47	3.25_{b} ± 0.47	3.00_{b} ± 0.40
5th day	1.75_{c} ± 0.25	1.50_{bc} ± 0.28	2.50_{b} ± 0.28	2.25_{b} ± 0.25
7th day	0.75_{b} ± 0.25	0.50_{ab} ± 0.28	1.25_{a} ± 0.25	1.25_{a} ± 0.25
10th day	0.25_{a} ± 0.00	0.25_{a} ± 0.25	0.50_{a} ± 0.28	0.50_{a} ± 0.28

1. Values are mean ± SE of four replicates.
2. Values sub scribed with same alphabets do not differ significantly ($P<0.05$), within groups at different time intervals.
3. Small letters are used to differentiate within groups at different time intervals.
4. The values of cutaneous oedema taken as
   Normal as preoperative (0), Slight swelling (1), Slight to Mild swelling (2), Moderate swelling (3), Severe swelling (4).

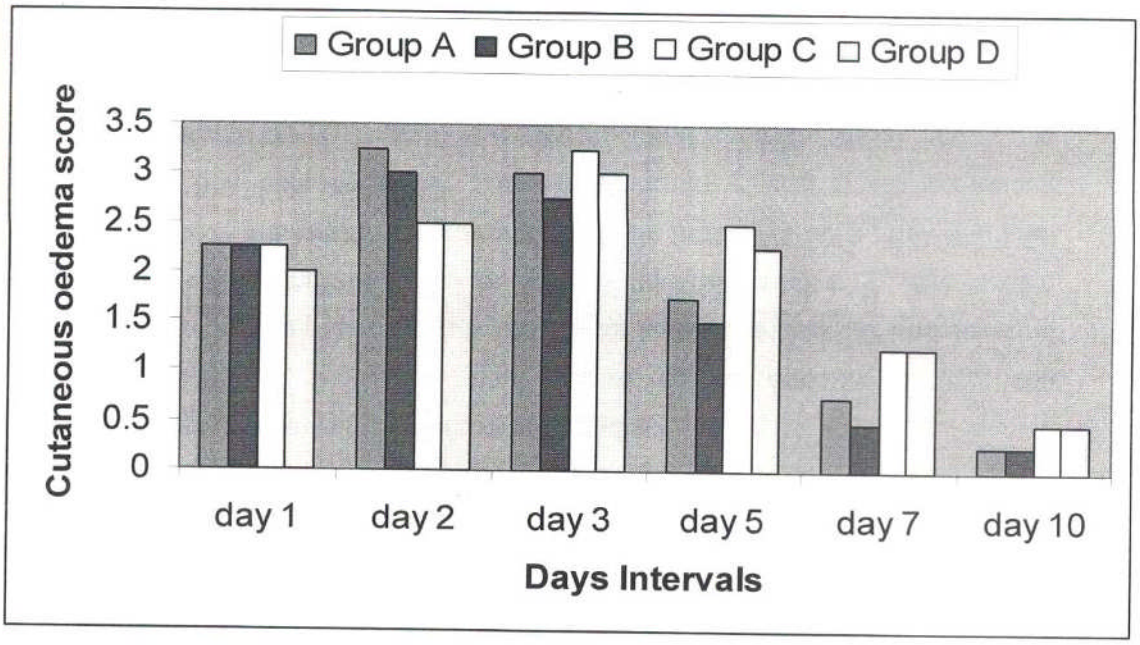


Fig.-5: Cutaneous oedema in different groups of bitches at different intervals.
0=Normal as preoperative ; 1=Slight swelling ; 2= Mild swelling ; 3=Moderate swelling ; 4=Severe swelling.

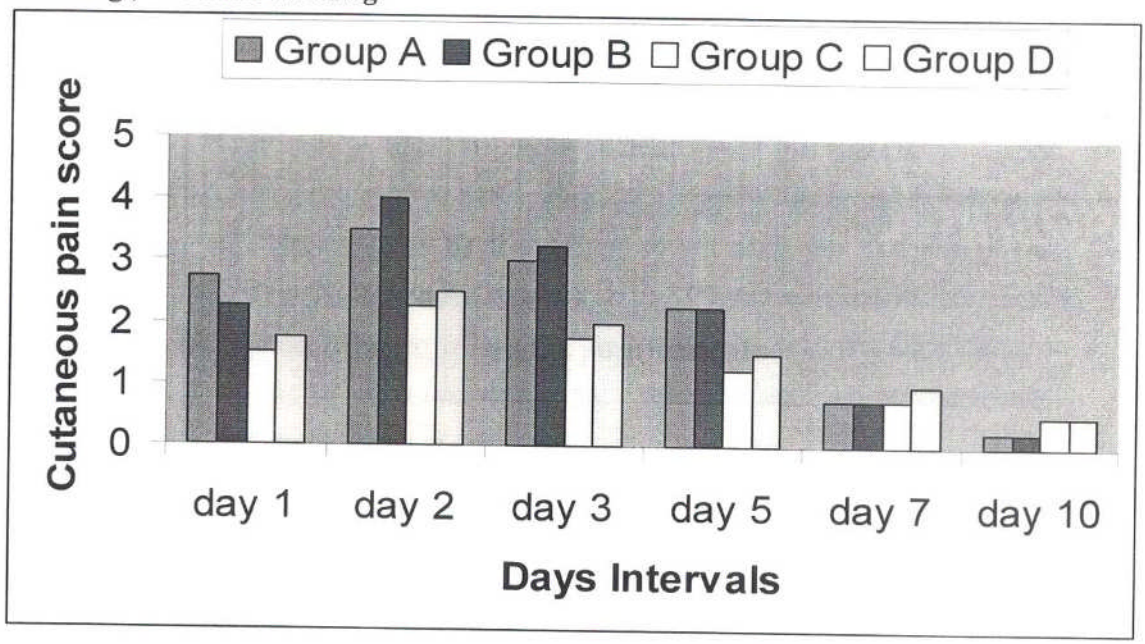


Fig.-6: Cutaneous pain in different groups of bitches at different intervals.
0=Normal as preoperative ; 1=Slight pain ; 2= Mild pain ; 3=Moderate pain ; 4=Severe pain.

skin with no significant difference in between groups. In the animals of group A and B, cutaneous oedema increased significantly to the maximum on 2nd (48hrs) and then decrease nonsignificantly from 5th to 10th day postoperatively, whereas in group C and D the cutaneous swelling increased non significantly to the maximum on 3rd day and then decreased nonsignificantly on 5th day and significantly on 7th day onward up to 10th day interval. The intensity of cutaneous oedema decreased to approaching normal or near normal as no oedema at 10th day postoperatively in all the groups of animals.

(b) Pain on palpation at the site of incision:

The observations with regard to the degree of cutaneous pain at incision site for different groups of animals are presented in Table-7. A significant ($P< 0.05$) increase in cutaneous pain at site of incision was recorded at 1st, 2nd and 3rd day postoperatively in group A and B in comparison to group C and D, respectively. While there were no significant differences between groups at 5th, 7th and 10th day intervals. A significant increase in cutaneous pain was recorded at 1st day (24hrs) after operation in comparison to normal in all the groups of bitches. However the intensity of pain increase significantly to its maximum on 2nd day after operation in all the groups, there after the intensity of pain gradually declined nonsignificantly up to 10th day postoperatively except at the period between 5th and 7th day, where it declined significantly in group A and B, while intensity of pain gradually declined nonsignificantly up to 10th day interval in group C and D.

(c) Warmth of cutaneous incision site:

The observations with regard to the degree of cutaneous warmth at incision site for different groups of animals are presented in Table-8. A significant ($P<0.05$) difference in site warmth was observed in all the groups at 2nd and 3rd day intervals while no significant difference in all the groups at 1st,5th,7th and 10th day postoperatively. The site warmth was

Table 7.

Mean values ± SE of cutaneous pain score in different groups of bitches at different intervals

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin sutbure)
1st day (24hrs)	2.75_{b}^{C} ± 0.25	2.50_{b}^{BC} ± 0.28	1.50_{b}^{A} ± 0.28	1.75_{b}^{AB} ± 0.25
2nd day (48hrs)	3.50_{c}^{B} ± 0.28	4.00_{c}^{B} ± 0.00	2.25_{c}^{A} ± 0.25	2.50_{c}^{A} ± 0.28
3rd day	3.00_{bc}^{BC} ± 0.40	3.25_{bc}^{C} ± 0.25	1.75_{bc}^{A} ± 0.25	2.00_{c}^{AB} ± 0.40
5th day	2.25_{b} ± 0.25	2.50_{b} ± 0.28	1.25_{b} ± 0.25	1.50_{bc} ± 0.25
7th day	0.75_{a} ± 0.25	0.75_{a} ± 0.47	0.75_{ab} ± 0.25	1.00_{ab} ± 0.28
10th day	0.25_{a} ± 0.25	0.25_{a} ± 0.25	0.50_{a} ± 0.28	0.50_{a} ± 0.28

1. Values are mean ± SE of four replicates.

2. Values sub / super scribed with same alphabets do not differ significantly ($P<0.05$)

3. Small letters are used to differentiate within groups at different time intervals.

4. Capital letters are used to differentiate between groups at different time intervals.

5. The values of cutaneous pain taken as
   Normal as preoperative (0), Slight pain (1), Slight to mild pain (2), Moderate pain (3), Sever pain (4).

Table 8.

Mean values ± SE of warmth of incision site (^{0}F) in different groups of bitches at different intervals

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
1st day (24hrs)	2.25_{c} ± 0.25	2.50_{c} ± 0.28	1.50_{c} ± 0.28	1.75_{b} ± 0.25
2nd day (48hrs)	3.25_{d}^{B} ± 0.25	3.50_{d}^{B} ± 0.28	2.25_{d}^{A} ± 0.25	2.75_{c}^{A} ± 0.25
3rd day	2.50_{cd}^{B} ± 0.25	3.00_{cd}^{B} ± 0.00	1.75_{cd}^{A} ± 0.25	2.00_{bc}^{A} ± 0.40
5th day	1.25_{b} ± 0.25	1.50_{b} ± 0.28	1.00_{bc} ± 0.40	1.50_{b} ± 0.28
7th day	0.00_{a} ± 0.00	0.25_{a} ± 0.25	0.50_{a} ± 0.28	0.50_{a} ± 0.28
10th day	0.00_{a} ± 0.00	0.00_{a} ± 0.00	0.50_{a} ± 0.28	0.18_{a} ± 0.10

1. Values are mean ± SE of four replicates.

2. Values sub / super scribed with same alphabets do not differ significantly ($P<0.05$)

3. Small letters are used to differentiate within groups at different time intervals.

4. Capital letters are used to differentiate between groups at different time intervals.

5. The values of warmth of incision site were taken as
   The preoperative skin warmth difference taken as 0 ^{0}F
   Normal warmth difference ≤ 1 ^{0}F
   Mild warmth difference ≥ 1-2 ^{0}F
   Moderate warmth difference ≥ 2-3 ^{0}F
   Severe warmth difference ≥ 3-4 ^{0}F

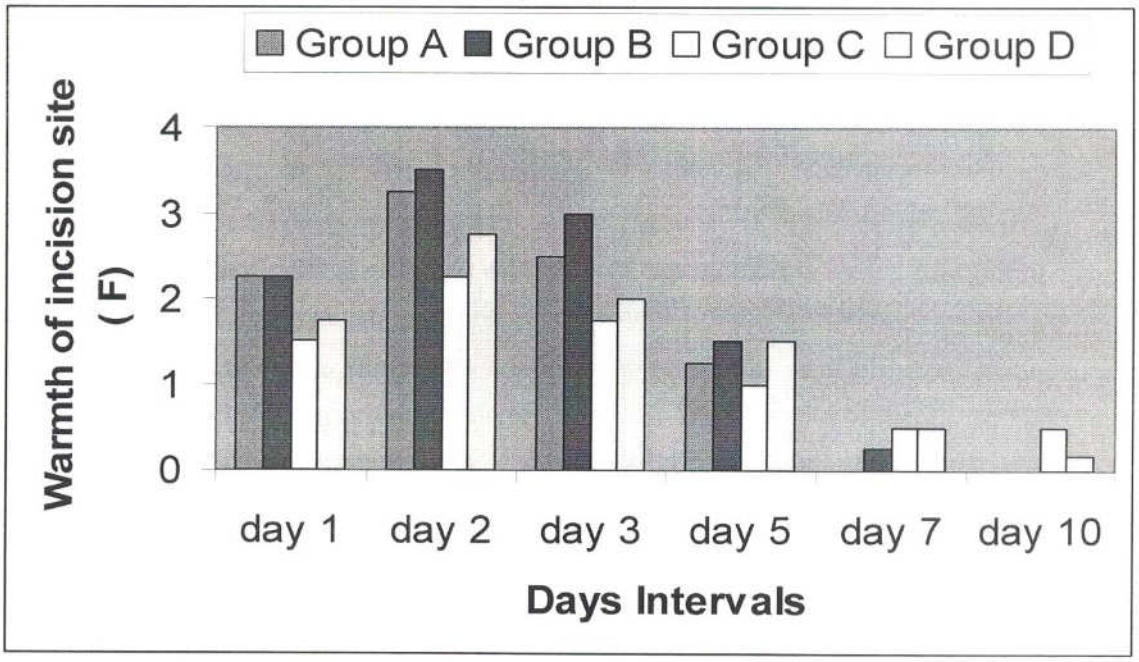


Fig.-7: Warmth of incision site (^{0}F) in different groups of bitches at different intervals.

The preoperative skin warmth difference taken as 0 ^{0}F

Normal warmth difference	**$\leq$ 1 ^{0}F**
Mild warmth difference	**$\geq$ 1-2 ^{0}F**
Moderate warmth difference	**$\geq$ 2-3 ^{0}F**
Severe warmth difference	**$\geq$ 3-4 ^{0}F**

significantly higher in groups A and B in comparison to groups C and D at 2nd and 3rd day postoperatively. A significant increase in site warmth up to maximum at 2nd day after that nonsignificant declined at 3rd day postoperative was observed in all groups of animals. However a significant decrease in site warmth was observed between 3rd day and 5th day intervals in group A and B while nonsignificant decrease in group C and D animals. There was a significant decrease in site warmth between 5th and 7th day postoperatively in all the groups, afterwards a gradual nonsignificant decrease in site warmth up to 10th day of operation was observed.

(ii) Postoperative complications:

The postoperative complications observed in different groups of animals are presented in Table-9. The animals of group D and group C had significantly ($P<0.05$) more postoperative complications with regard to suture biting, suture dehiscence in No.A16 and exudation in No.A15, animals of group D and exudation in No.A11 and A12 animals of group C was observed in comparison to group B in which suture biting was observed in No.A5 animal whereas group A animals were recovered uneventfully without any complication.

(4) Total cost incurred:

The cost of surgery included coat of anaesthetic, site preparation, suture material and postoperative medicines used are presented in Table-10, for different groups of animals. There was a significant ($P<0.05$) difference between all the groups of animals. However there was no significant difference between group A (251.08 ± 3.15) and B (235.18 ± 7.06), and group C (304.39 ± 17.65) and D (286.07 ± 13.53), respectively.

Table 9. Postoperative complications observed in different groups of animals.

Groups of animals	Postoperative complications							Total
	Animal number	Post-operative hemorrhage	Suture biting	Wound dehis-cence	Exudation from suture line	Evisce-ration	Peri-tonitis and death	
Group A	A1	-	-	-	-	-	-	0
	A2	-	-	-	-	-	-	
	A3	-	-	-	-	-	-	
	A4	-	-	-	-	-	-	
Group B	A5	-	+	-	-	-	-	1
	A6	-	-	-	-	-	-	
	A7	-	-	-	-	-	-	
	A8	-	-	-	-	-	-	
Group C	A9	-	-	-	-	-	-	2
	A10	-	-	-	-	-	-	
	A11	-	-	-	+	-	-	
	A12	-	-	-	+	-	-	
Group D	A13	-	-	-	-	-	-	4
	A14	-	-	-	-	-	-	
	A15	-	-	-	+	-	-	
	A16	-	-	+	-	+	-	

Table 10.

Mean ± SE of Total cost of surgery (Ovariohysterectomy) in different groups of bitches.

S. No.	Animal Groups	Cost of anaesthesia (Rs.)	Cost of site preparation (Rs.)	Cost of suture material (Rs.)	Cost of postoperative medicine (Rs.)	Total (Rs.)
1.	Group A	34.81_a ± 1.13	8.00 ± 0.00	60.86_a ± 9.79	132.50_a ± 2.50	236.17_a ± 7.44
2.	Group B	39.94_a ± 0.57	8.00 ± 0.00	57.75_a ± 9.42	144.50_b ± 6.07	250.19_a ± 5.41
3.	Group C	44.39_b ± 1.95	8.00 ± 0.00	75.88_b ± 12.75	152.50_b ± 7.50	280.82_b ± 5.15
4.	Group D	54.94_b ± 2.99	8.00 ± 0.00	65.97_b ± 10.78	172.5_c ± 11.08	301.41_c ± 7.6

1. Values are mean ± SE of four replicates.
2. Values sub scribed with same alphabets do not differ significantly ($P<0.05$) between groups at different time intervals.
3. Small letters are used to differentiate between groups at different time intervals.

(5) General body condition:

There were no abnormalities observed in general body condition with regard to appetite, feed and water intake, urination and defecation in all the groups of animals in the present study during observation period.

(6) Hematological observations:

(a) Hemoglobin (Hb):

The observations with regard to hemoglobin for different groups of animals are presented in Table-11. A significant ($P<0.05$) decline in values of hemoglobin concentration was observed in group A and B in comparision to group C and D at 1st day (24hrs.) and 2nd day postoperatively. There was also a significant decrease in hemoglobin within the groups was observed in group A and B at 1st and 2nd day postoperatively in comparison to preoperative values, whereas in group C and D the hemoglobin decline nonsignificantly at 1st and 2nd day postoperatively. Thereafter the hemoglobin values approached nearer to preoperative values on 7th or 10th day postoperatively in all the groups of animals.

(b) Packed cell volume (PCV):

The observations with regard to Packed cell volume (PCV) for different groups of animals are presented in Table-12. There was no significant difference observed in packed cell volume in between different groups of animals at any postoperative day. However a significant ($P<0.05$) decrease in PCV value in group A and B whereas a nonsignificant decrease in PCV value in group C and D animals was observed at 1st day (24hrs) and 2nd day after operation in comparison to preoperative values thereafter a gradual nonsignificant increase in PCV value was observed in all the groups of animals up to 10th day postoperatively, the values

Table 11.

Mean values ± SE of Hemoglobin (gm %) in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
preoperative	12.75_{b} ± 0.32	12.70_{c} ± 0.23	12.60 ± 0.29	12.75 ± 0.33
1st day	10.60_{a}^{A} ± 0.28	10.40_{a}^{A} ± 0.46	11.45^{B} ± 0.43	11.45^{B} ± 0.43
2nd day	11.20_{a}^{AB} ± 0.41	10.85_{a}^{A} ± 0.27	11.95^{B} ± 0.33	11.85^{B} ± 0.29
3rd day	12.20_{ab} ± 0.25	12.20_{ab} ± 0.11	12.45 ± 0.27	12.45 ± 0.27
5th day	12.50_{ab} ± 0.30	12.60_{b} ± 0.24	12.60 ± 0.29	12.60 ± 0.29
7th day	12.70_{b} ± 0.30	12.55_{ab} ± 0.26	12.70 ± 0.23	12.70 ± 0.35
10th day	12.80_{b} ± 0.29	12.85_{b} ± 0.32	12.60 ± 0.29	12.90 ± 0.31

1. Values are mean ± SE of four replicates.
2. Values sub / super scribed with same alphabets do not differ significantly ($P<0.05$)
3. Small letters are used to differentiate within groups at different time intervals.
4. Capital letters are used to differentiate between groups at different time intervals.

Table 12.

Mean values ± SE of Packed cell volume (%) in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
preoperative	42.81_b ± 056	41.67_b ± 0.87	40.25 ± 0.62	40.45 ± 0.66
1st day (24hrs)	38.60_a ± 0.64	37.90_a ± 0.73	38.35 ± 0.33	38.40 ± 0.32
2nd day	39.85_a ± 049	39.45_{ab} ± 0.65	39.15 ± 0.30	39.20 ± 0.35
3rd day	41.85_b ± 0.57	40.35_b ± 0.69	40.15 ± 0.73	40.20 ± 0.73
5th day	42.35_b ± 0.42	41.15_b ± 0.73	40.20 ± 0.70	40.25 ± 0.70
7th day	42.65_b ± 0.61	41.55_b ± 0.85	40.30 ± 0.71	40.30 ± 71
10th day	42.75_b ± 0.55	41.65_b ± 0.87	40.80 ± 0.63	40.80 ± 0.63

1. Values are mean ± SE of four replicates

2. Values sub scribed with same alphabets do not differ significantly ($P<0.05$) within groups at different time intervals.

3. Small letters are used to differentiate within groups at different time intervals.

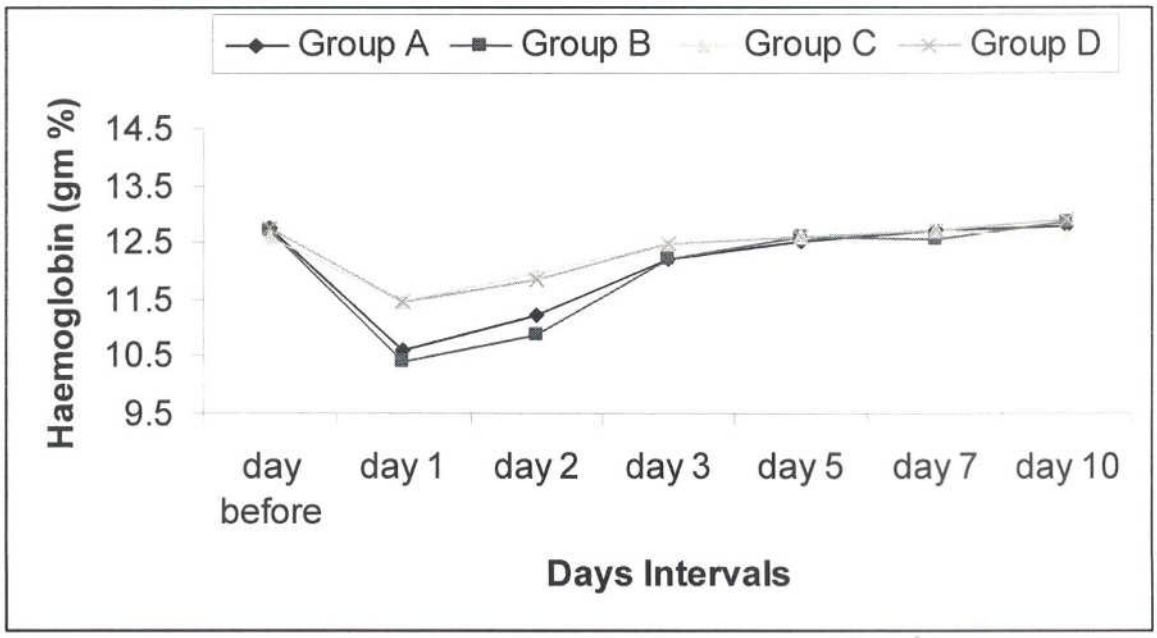


Fig.-8: Hemoglobin (gm %) in different groups of bitches at different intervals.

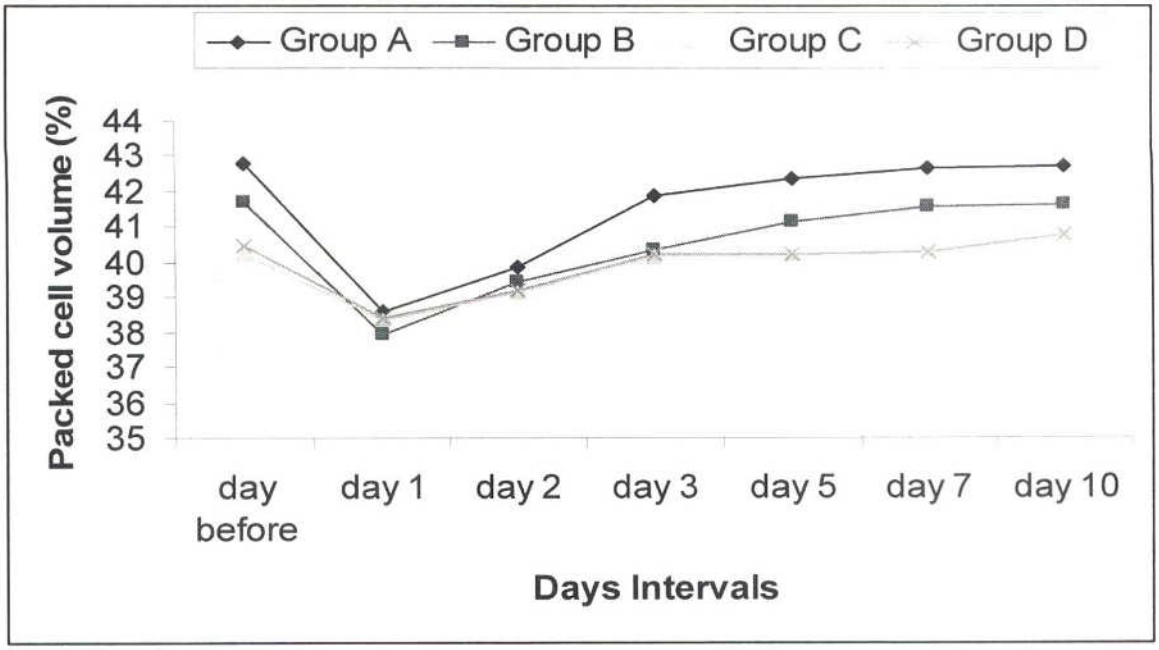


Fig.-9: Packed cell volume (%) in different groups of bitches at different intervals.

approached near to the preoperative value at 7th or 10th day after operation.

(c) Total leukocyte count (TLC):

The observations with regard to Total leukocyte count (TLC) for different groups of animals are presented in Table-13. There was no significant difference observed in TLC in between different groups of animals at any postoperative day. A significant ($P<0.05$) increase in TLC was observed within groups A and B at 2nd day postoperatively in comparison to normal preoperative values after that the TLC value shown nonsignificant gradual decrease up to 10th postoperative day. A nonsignificant increase in TLC up to 2nd day after operation and then a declined trend up to 10th postoperative day was also observed in animals of group C and D.

(d) Neutrophil:

The observations with regard to neutrophil for different groups of animals are presented in Table-14. There was no significant difference in the values of neutrophil recorded between different groups of animals at different postoperative days. A significant ($P<0.05$) increase in neutrophil up to maximum at 2nd day postoperatively was observed within group A and group B in comparison to normal preoperative values. A nonsignificant increase in neutrophil was also observed in animals of group C and D, reaching maximum at 2nd day postoperatively after that a gradual non significant decline in values of neutrophil was observed in all the groups of animals up to 10th day postoperatively where it approaches near preoperative values.

(e) Lymphocyte:

The observations with regard to lymphocyte for different groups of animals are presented in Table-15. There was no significant difference in the values of lymphocyte between different groups of animals at different

Table 13.

Mean values ± SE of Total leukocyte count (thousand / cu. mm) in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
preoperative	10.05_a ± 0.43	10.10_a ± 0.42	9.80 ± 0.29	10.00 ± 0.45
1st day (24hrs)	10.55_a ± 0.36	10.70_a ± 0.38	10.35 ± 0.34	10.80 ± 0.58
2nd day	12.55_b ± 0.35	12.35_b ± 0.32	11.25 ± 0.61	12.00 ± 0.58
3rd day	11.05_{ab} ± 0.29	11.25_{ab} ± 0.35	10.80 ± 0.29	11.00 ± 0.48
5th day	10.20_a ± 0.39	10.50_a ± 0.36	10.15 ± 0.41	10.45 ± 0.55
7th day	10.15_a ± 0.48	10.20_a ± 0.41	9.95 ± 0.33	10.10 ± 0.47
10th day	10.00_a ± 0.39	10.15_a ± 0.38	9.85 ± 0.33	10.05 ± 0.40

1. Values are mean ± SE of four replicates

2. Values sub scribed with same alphabets do not differ significantly ($P<0.05$) within groups at different time intervals.

3. Small letters are used to differentiate within groups at different time intervals.

Table 14.

Mean values ± SE of Neutrophil count (%) in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
preoperative	67.00_{a} ± 1.08	68.50_{ab} ± 1.19	66.75 ± 1.10	67.75 ± 1.10
1st day (24hrs)	71.50_{ab} ± 1.70	72.75_{bc} ± 1.60	70.25 ± 1.49	70.00 ± 1.47
2nd day	74.75_{b} ± 1.49	75.25_{c} ± 1.43	72.00 ± 1.47	72.75 ± 1.31
3rd day	71.50_{ab} ± 1.93	72.75_{bc} ± 1.70	69.25 ± 1.43	69.50 ± 1.19
5th day	69.75_{a} ± 1.79	70.00_{ab} ± 1.35	66.50 ± 1.25	67.75 ± 1.10
7th day	67.75_{a} ± 1.03	69.00_{ab} ± 1.35	67.75 ± 1.25	67.75 ±1.10
10th day	67.00_{a} ± 1.29	68.00_{a} ± 1.35	66.25 ± 1.25	67.00 ± 1.29

1. Values are mean ± SE of four replicates.
2. Values sub scribed with same alphabets do not differ significantly ($P<0.05$) within groups at different time intervals.
3. Small letters are used to differentiate within groups at different time intervals.

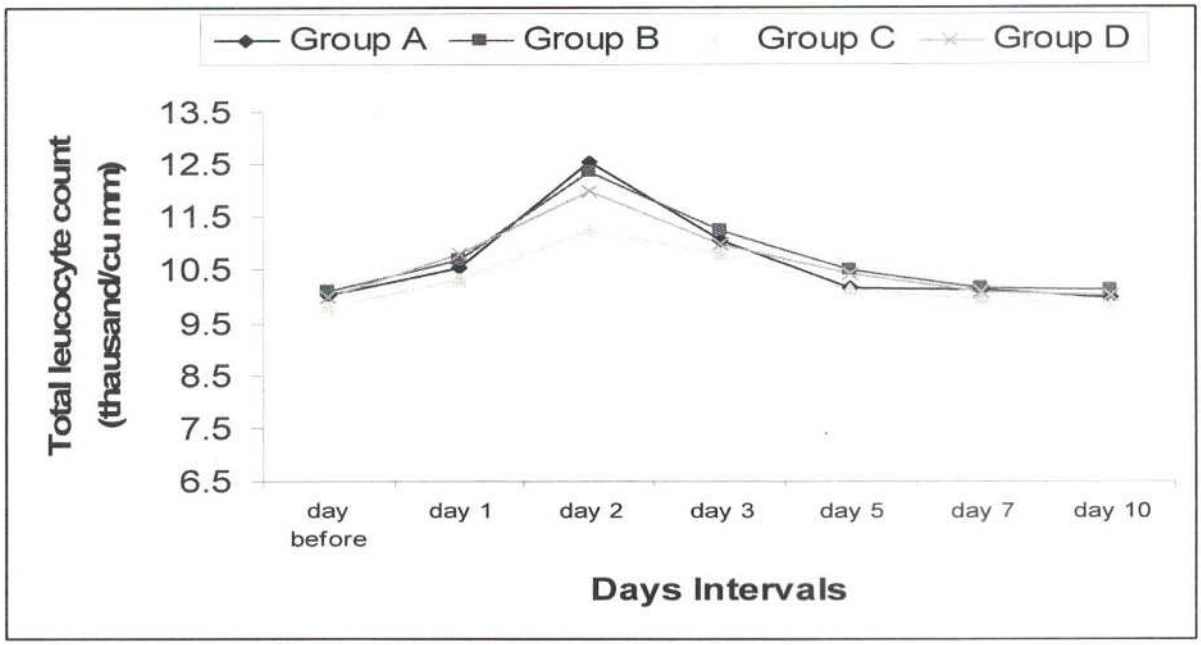


Fig.-10: Total leukocyte count (thousand / cu mm) in different groups of bitches at different intervals.

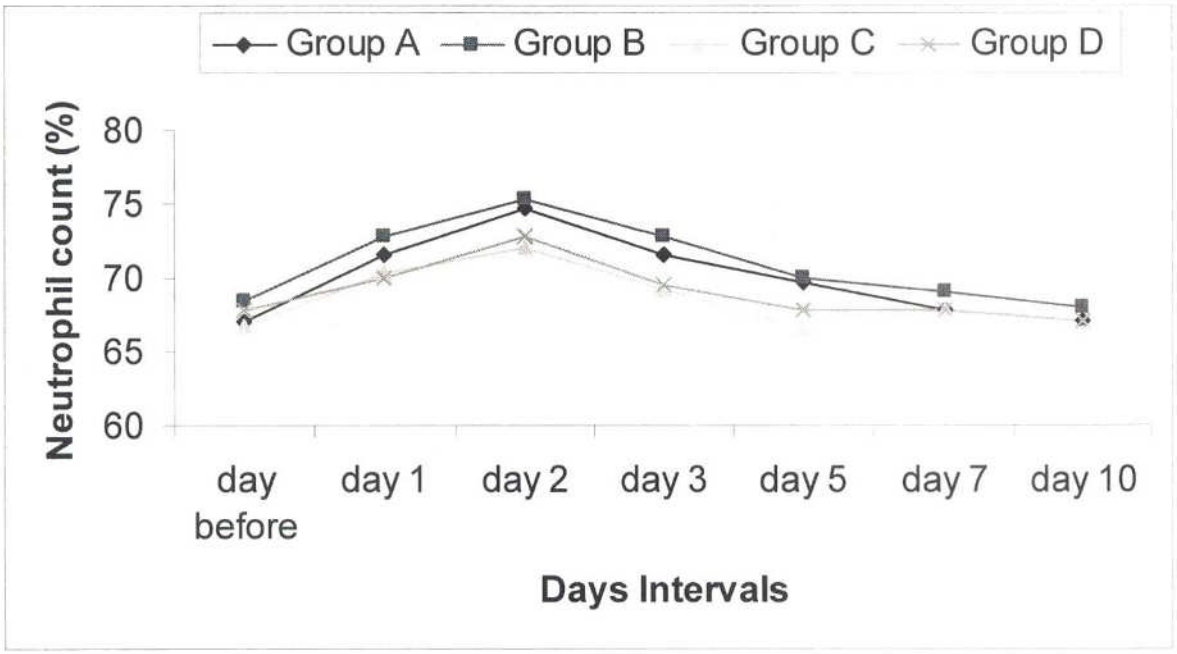


Fig.-11: Neutrophil count (%) in different groups of bitches at different intervals.

postoperative days. A significant ($P<0.05$) decrease in lymphocyte up to minimum at 2nd day postoperatively was observed within group A and group B in comparison to normal preoperative values. A nonsignificant decrease in lymphocyte was also observed in group C and D animals up to minimum at 2nd day postoperatively after that a gradual non significant increase in values of lymphocyte was observed in all the groups of animals up to 10th day postoperatively where it approaches near preoperative values.

(f) Monocyte:

The observations with regard to monocyte for different groups of animals are presented in Table-16. There was no significant difference in the values of monocyte between different groups of animals at different postoperative days. A significant ($P<0.05$) increase in monocyte up to maximum at 2nd day postoperatively was observed within group A and group B in comparison to normal preoperative values. A nonsignificant increase in monocyte was also observed in group C and D animals up to maximum at 2nd day postoperatively after that a gradual non significant decline in values of monocyte was observed in all the groups of animals up to 10th day postoperatively where it approaches near preoperative values.

(g) Eosinophil:

The observations with regard to eosinophil for different groups of animals are presented in Table-17. No significant differences in the values of eosinophil were recorded in between different groups of animals at different postoperative periods and within the groups. However a non significant decrease in number of eosinophil at 2nd day (48hrs) postoperatively after that gradual non significant increase in number of eosinophil up to 10th day postoperatively was observed in all groups of animals where it reached almost similar to preoperative values.

Table 15.

Mean values ± SE of Lymphocyte count (%) in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
preoperative	29.50_{c} ± 1.04	28.25_{c} ± 0.85	28.00 ± 1.08	29.00 ± 1.08
1st day (24hrs)	25.25_{a} ± 1.25	24.75_{a} ± 1.43	24.00 ± 0.91	25.50 ± 0.95
2nd day	25.00_{a} ± 1.29	23.75_{a} ± 1.25	23.75 ± 0.85	24.50 ± 0.95
3rd day	26.25_{ab} ± 1.25	25.50_{ab} ± 0.86	25.25 ± 0.86	26.75 ± 0.85
5th day	27.25_{b} ± 1.03	26.25_{b} ± 0.85	26.00 ± 1.08	27.25 ± 1.10
7th day	28.00_{bc} ± 1.08	27.50_{bc} ± 1.04	27.00 ± 1.08	28.25 ± 0.85
10th day	28.25_{bc} ± 1.25	28.00_{bc} ± 1.08	27.50 ± 1.04	28.50 ± 1.43

1. Values are mean ± SE of four replicates.
2. Values sub scribed with same alphabets do not differ significantly ($P<0.05$) within groups at different time intervals.
3. Small letters are used to differentiate within groups at different time intervals.

Table 16.

Mean values ± SE of Monocyte count (%) in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
preoperative	2.75_{ab} ± 0.25	2.25_{a} ± 0.25	2.75 ± 0.25	2.25 ± 0.25
1st day (24hrs)	3.50_{bc} ± 0.25	3.25_{c} ± 0.28	3.50 ± 0.28	3.50 ± 0.28
2nd day	3.75_{c} ± 0.25	3.50_{c} ± 0.28	3.50 ± 0.28	3.25 ± 0.25
3rd day	2.75_{ab} ± 0.25	2.75_{ab} ± 0.25	2.75 ± 0.47	2.75 ± 0.47
5th day	2.75_{ab} ± 0.25	2.75_{ab} ± 0.25	2.75 ± 0.25	2.75 ± 0.47
7th day	2.75_{ab} ± 0.25	2.75_{ab} ± 0.25	2.25 ± 0.25	2.50 ± 0.28
10th day	2.50_{a} ± 0.28	2.50_{a} ± 0.28	2.75 ± 0.47	2.25 ± 0.25

1. Values are mean ± SE of four replicates

2. Values sub scribed with same alphabets do not differ significantly ($P<0.05$) within groups at different time intervals.

3. Small letters are used to differentiate within groups at different time intervals.

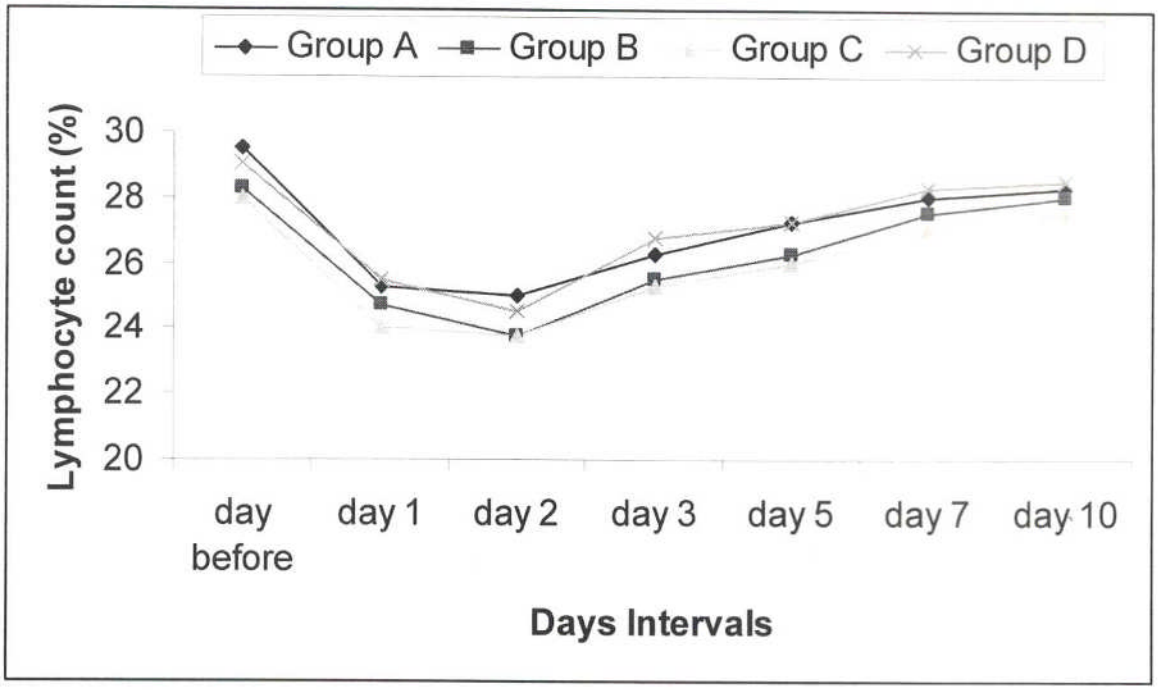


Fig.-12: Lymphocyte count (%) in different groups of bitches at different intervals.

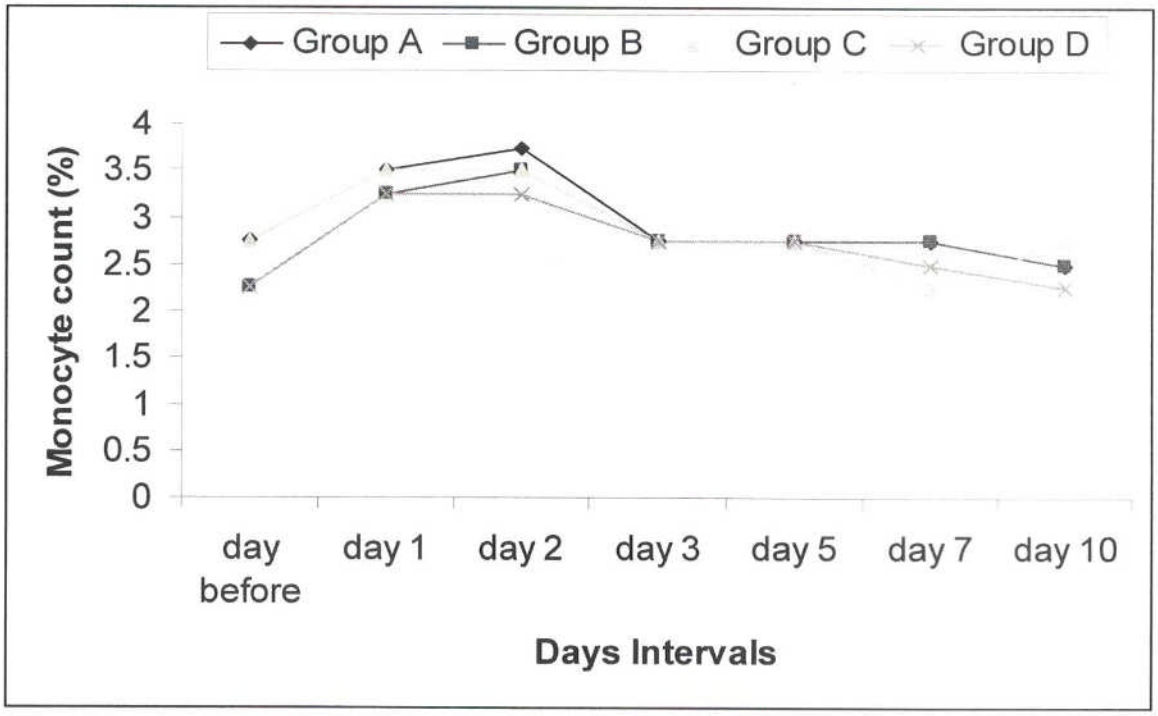


Fig.-13: Monocyte count (%) in different groups of bitches at different intervals.

Table 17.

Mean values ± SE of Eosinophil count (%) in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
preoperative	1.75 ± 0.41	2.00 ± 0.40	1.75 ± 0.41	1.75 ± 0.25
1st day (24hrs)	1.50 ± 0.28	1.50 ± 0.28	1.50 ± 0.25	1.50 ± 0.25
2nd day	1.25 ± 0.25	1.25 ± 0.25	1.00 ± 0.50	1.25 ± 0.25
3rd day	1.50 ± 0.28	1.50 ± 0.25	1.25 ± 0.25	1.50 ± 0.28
5th day	1.50 ± 0.25	1.50 ± 0.28	1.50 ± 0.25	1.50 ± 0.25
7th day	1.75 ± 0.25	1.75 ± 0.25	1.50 ± 0.25	1.75 ± 0.25
10th day	1.50 ± 0.28	1.75 ± 0.41	1.75 ± 0.25	1.75 ± 0.25

1. Values are mean ± SE of four replicates.

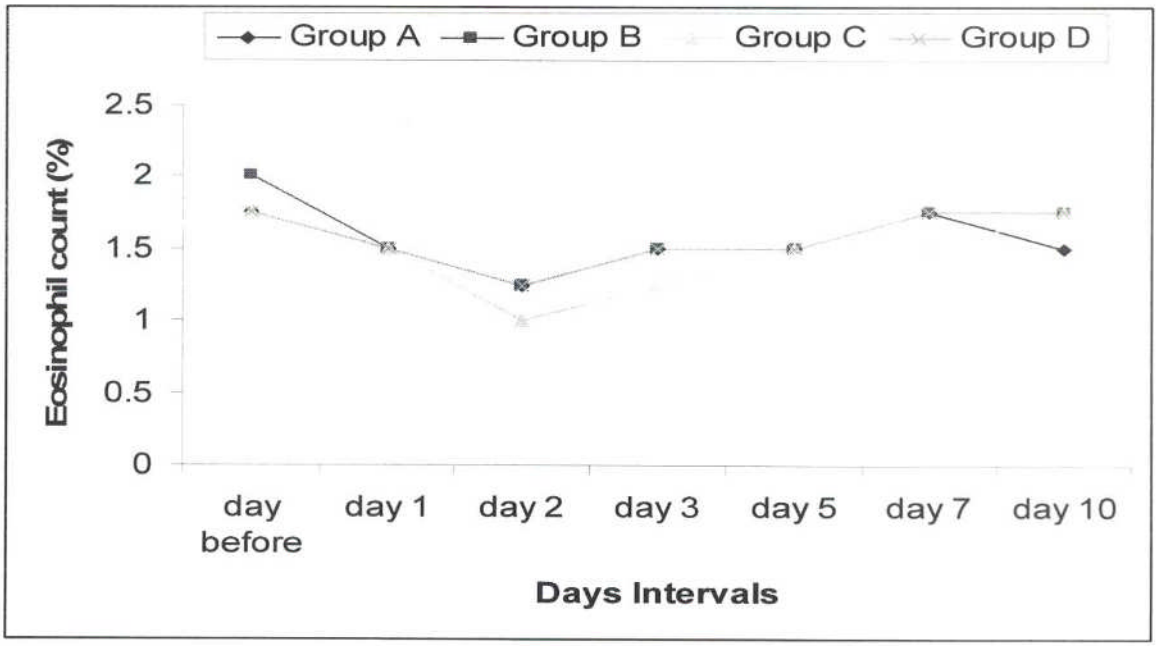


Fig.-14: Eosinophil count (%) in different groups of bitches at different intervals.

Table 18.

Mean values ± SE of Glucose (mg/100ml) in different groups of bitches at different intervals.

Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
preoperative	82.35 ± 4.02	87.24 ± 3.59	84.27 ± 3.09	87.97 ± 2.40
1st day (24hrs)	91.43 ± 5.06	96.92 ± 1.86	97.47 ± 3.12	96.86 ± 2.32
2nd day	87.53 ± 5.36	93.09 ± 2.96	91.56 ± 4.08	90.91 ± 2.81
3rd day	85.25 ± 4.34	90.91 ± 2.61	88.43 ± 3.46	87.32 ± 3.49
5th day	84.10 ± 5.36	91.40 ± 2.41	89.16 ± 3.64	88.31 ± 3.43
7th day	84.27 ± 4.88	90.18 ± 2.35	88.16 ± 3.83	89.25 ± 3.31
10th day	84.89 ± 5.41	90.82 ± 2.42	88.10 ± 3.74	90.13 ± 2.43

1. Values are mean ± SE of four replicates.

Table 19.

Mean values ± SE of Serum Creatinine (mg/100ml) in different groups of bitches at different intervals.

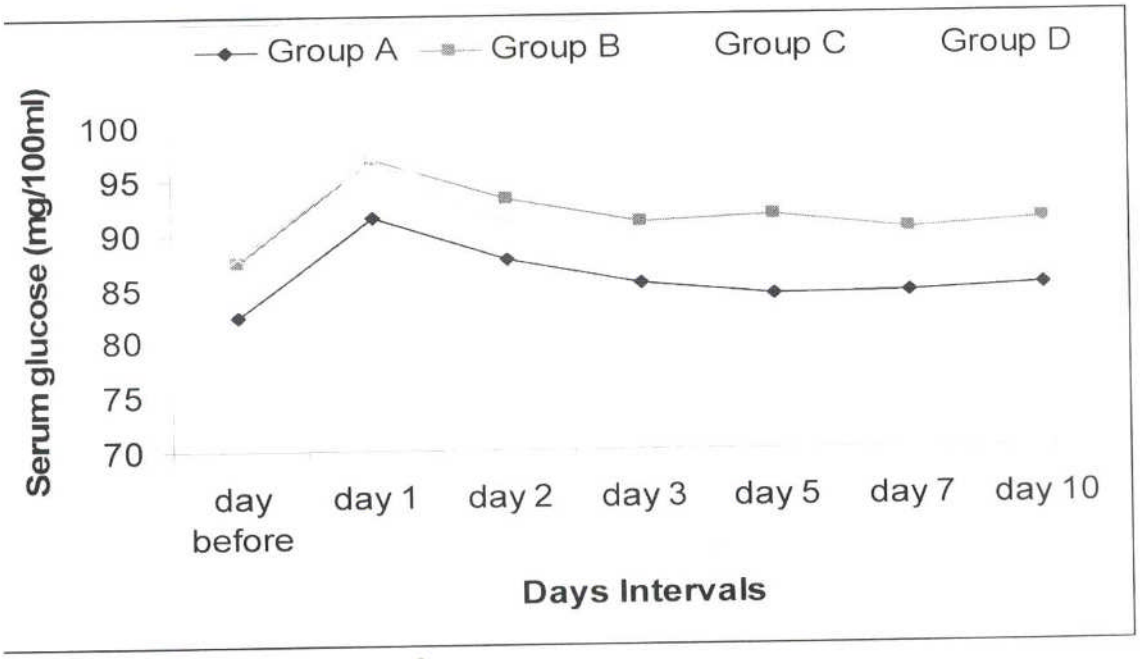
Intervals (Days)	Flank approach		Midline approach	
	group A (subcuticular suture)	group B (external skin suture)	group C (subcuticular suture)	group D (external skin suture)
preoperative	0.63_{a} ± 0.06	0.62_{a} ± 0.04	0.75_{a} ± 0.06	0.67_{a} ± 0.04
1st day (24hrs)	1.10_{b}^{B} ± 0.11	1.00_{b}^{B} ± 0.10	0.85_{a}^{A} ± 0.06	0.79_{a}^{A} ± 0.06
2nd day	1.12_{b}^{B} ± 0.10	1.23_{b}^{B} ± 0.12	0.88_{b}^{A} ± 0.08	0.85_{b}^{A} ± 0.08
3rd day	1.22_{b}^{B} ± 0.12	1.24_{b}^{B} ± 0.11	0.96_{b}^{A} ± 0.06	0.89_{b}^{A} ± 0.10
5th day	1.23_{c}^{B} ± 0.13	1.24_{c}^{B} ± 0.12	0.98_{b}^{A} ± 0.11	0.96_{b}^{A} ± 0.11
7th day	1.15_{b}^{B} ± 0.11	1.20_{b}^{B} ± 0.11	0.95_{b}^{A} ± 0.08	0.85_{b}^{A} ± 0.06
10th day	1.10_{a} ± 0.10	1.02_{a} ± 0.10	0.87_{a} ± 0.06	0.75_{a} ± 0.08

1. Values are mean ± SE of four replicates.

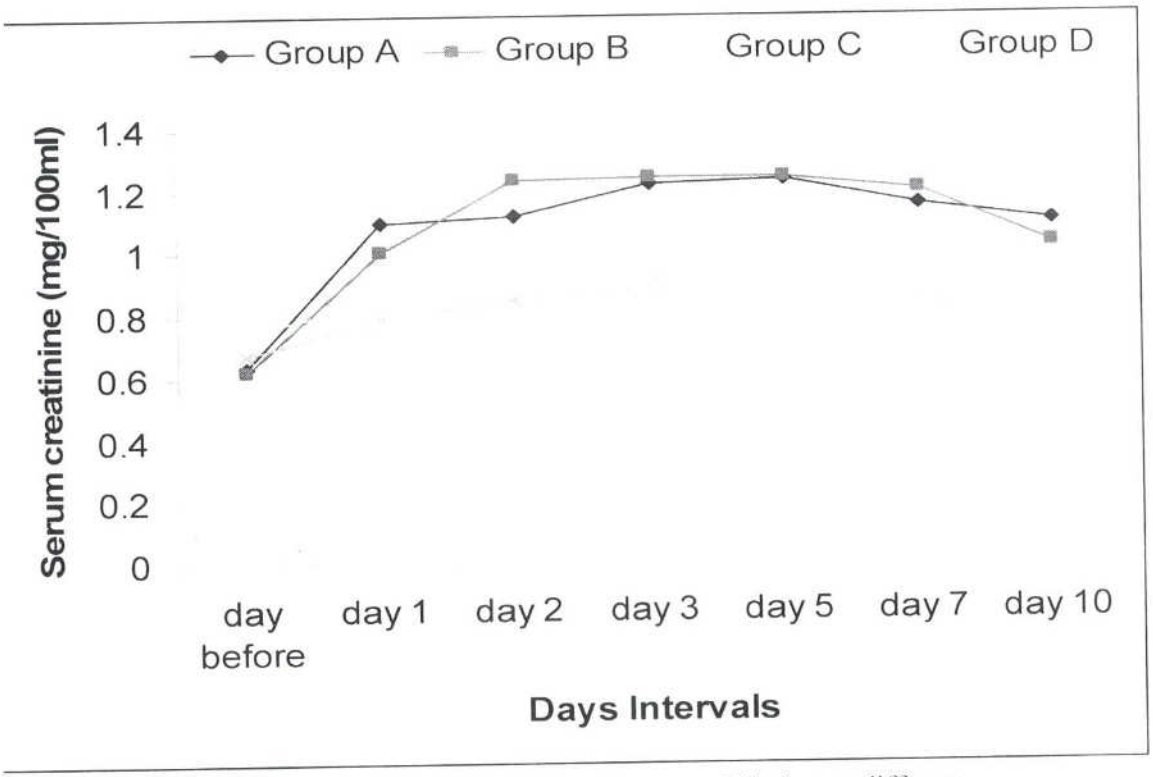
2. Values sub / super scribed with same alphabets do not differ significantly ($P<0.05$)

3. Small letters are used to differentiate within groups at different time intervals.

4. Capital letters are used to differentiate between groups at different time intervals.



g.-15: Glucose (mg/100ml) in different groups of bitches at different intervals.



ig.-16: Serum Creatinine (mg/100ml) in different groups of bitches at different ntervals.

(7) Biochemical observations:

(a) Serum glucose:

The observations on the serum glucose for different groups of animals are presented in Table-18. No significant differences in the values of serum glucose were recorded in different groups of animals and at different postoperative periods. However a non significant increase in serum glucose was observed after 24hrs (2^{nd} day) of operation after that it decline gradually up to 10^{th} day of operation which was still greater than normal preoperative values, in all groups of animals.

(b) Serum Creatinine:

The values of serum creatinine of different groups of animals are recorded in Table-19. A significant ($P<0.05$) increase in the values of creatinine was observed in A and B groups in comparison to C and D groups at 1^{st},2^{nd},3^{rd},5^{th} and 7^{th} day after operation. There was a statistically significant gradual increase in the serum creatinine values in all groups of animals in comparison to preoperative values up to 5^{th} postoperative day then the values gradually and significantly return to their preoperative values by 10^{th} postoperative day.


DISCUSSION

CHAPTER-6

DISCUSSION

Present study envisages a clinical evaluation of different techniques of ovariohysterectomy in bitches (OH) in terms of various parameters like duration of operation, operative and postoperative complications, and status of wound healing and cost effectiveness of operative procedure.

In the present study ovariohysterectomy techniques involved were right flank and mid-ventral approach and were discussed on the basis of various parameters selected for the study as below:

Physiological effects:

The changes in **rectal temperature** after surgery as well as after anaesthesia and analgesia were influenced by the presence of infection, endogenous pyrogens, and due to changes in metabolic rate and peripheral blood circulation. Increase metabolic rate associated with inflammation also increases body temperature (Vegad, 1998). During the period of general anaesthesia and during recovery, there is hypothermia because of generalized distribution of blood as a result of peripheral vasodilation and decreased metabolic activity. In the present study no significant increase or decrease in the values of rectal temperature were recorded at any postoperative stage in any groups of animals, only minor fluctuations within normal physiological limits were observed in all the groups of animals which underwent OH either by flank approach or mid-ventral approach.

Heart and **respiration rates** are the important physiological functions, which use to fluctuate because of many variables, namely ambient temperature, anxiety, fear, stress, blood oxygen tension, body acid-base balance and due to the effect of analgesic or anaesthetic drugs. During surgery and anaesthesia all the above-mentioned factors play an important role. In the present study the values of heart and respiration

rates do not differ significantly at different time intervals in all the groups of animals. However only slight decrease in heart rate and respiration rate was observed at day of operation which might be due to depressive effect of xylazine on cardiovascular and respiratory centers as reported by Demirkan *et al.* (2002), and slight increase in heart rate at 24hrs after operation which might be due to inflammatory response of laparotomy wound in all the groups of animals. The differences in respiration and heart rates in the present study were not alarming and were within normal physiological limits.

Duration of operation:

The duration of operation in the present study was significantly less in flank approach in comparison to midline approach as duration of operation was directly proportional to the length of incision, as comparatively longer incision was required for ovariohysterectomy through midline approach which takes more time for suturing. The group A animals consume least time of surgery due to short operative incision, quick exteriorization and ligation of ovarian and uterine stumps and use of subcuticular suture for apposition of skin edge.

Rubin and Maplesden, 1978; Flynn, 1995; Hoque, 1991 also favoured the use of a short incision for OH in bitches as short incision take less time to suture and provide less surface area for possible contamination with better cosmetic appearance.

For dogs, the right flank approach is found to be more suitable for easy location and delivery of the ovaries as the suspensory ligamentous attachment of right ovary is short and right ovary is more cranially situated in bitches, hence could be quickly exteriorized through right flank. Since the left kidney is floating and left ovarian suspensory ligamentous attachment is loose as compared to right side, the left ovary can be much more easily exteriorized through right flank incision than right ovary through a left flank incision. These findings are in agreement with Wilson and Balasubramanian, 1967 and Rajoria, 2002.

The Dorn, (1975) also made use of right flank approach for spaying in dogs due to the same reason and was further pointed out that flank approach from the left side was more difficult because the omentum covered the viscera and the spleen would lie in the surgical field.

In the animals of present study a uniform sequence of ligation and severing of ovarian and uterine stumps were followed in both right flank and mid-ventral ovariohysterectomies, first the right ovarian stump was exteriorized, ligated and severed followed by left ovarian stump and then uterine body. This technique was found more satisfactory as it minimized time of exteriorization and ligation, because the right ovary has a shorter attachment and is more difficult to manipulate than the left ovary as has been also reported by Rubin and Maplesden (1978) and Stone (1985) but contrarily Smith (1974) and Fingland (1997) conducted OH by exteriorization and ligation of left ovarian stump first.

Intraoperative hemorrhage:

In the present study the intraoperative hemorrhages from laparotomy wounds were observed significantly higher in the groups of animals where OH done through flank approach (groups A and B) in comparison to mid-ventral approach animals, similar findings were reported by Hansson, (2005), and were due to more tissue bleeding when passing the abdominal wall through flank. Hansson, (2005) further opined that the midline approach had more complications with respect to bleedings from the ligatures during surgery. To minimized hemorrhafge, muscle separation technique for entering the abdominal cavity through flank approach for ovariohysterectomy was reported by Wilson and Balasubramanian 1967, Krazaczynski, 1974, Dorn 1975 and Hoque 1991.

Status of wound healing:

Status of cutaneous wound healing at different sites as evaluated with the help of inflammatory oedema, warmth of the site and pain on palpation revealed that initially the inflammatory response was significantly

more at flank incisions in comparison to midline incisions. Burrow *et al.* (2006) also reported higher pain score in cats neutered by flank approach in comparison to midline approach. This can be attributed to the differences in the vascularity of the different sites and interstitial separation of different tissue layers as more surgical maneuvers were required at flank laparotomy sites in comparison to mid-ventral sites. The postoperative wound healing in canine ovariohysterectomy following the use of buried continuous subcuticular suture pattern give better cosmetic results and eliminate the requirement for further appointments for suture removal as has also been reported by Sylvestre, *et al.* (2002). The skin incision at linea alba took 3-4 days more for healing than flank incisions as reported by Ormrod, (1966) and Hoque, (1991). Which can be attributed to decreased vascularity at linea alba. The animals which undergone OH through mid-ventral approach required more postoperative care in comparison to flank approach. However the healing pattern observed was usual and suture were removed in all animals at 10^{th} day after operation

Post-operative complications:

In the present study the associated postoperative complications were found highest in the animals which undergone ovariohysterectomies through midline approach and particularly in those where chromic gut sutures were used as has also been reported by Pearson, (1970). The most common postoperative complications observed in the present study in animals of midline approach was suture biting, suture dehiscence and seroma formation and has also been reported by Pollari *et al.*, 1996 and Fingland, 1997. Abdominal wound dehiscence characterized by herniation and prolapse on 4^{th} to 6^{th} postoperative day was reported to be the main hazard of OH through median incision (Wilson and Balasubramanian, 1967). These complications were possibly due to pressure of viscera on suture line (Dorn, 1975), contamination of suture line due to sitting habit of dogs and due to violent abdominal muscle contraction during barking. In the present study the ovarian and uterine stumps were ligated by

absorbable suture i.e. Chromic catgut and Vicryl in order to minimize the formation of sinuses which resulted with the use of nonabsorbable sutures like silk, multifilament nylon and strong linen as they cause marked tissue reaction due to irritant nature and may cause formation of uterine and ovarian stump granuloma and sinus formation at sub lumbar region as reported by Joshua 1965, Pearson 1970 and 1973, Borthwick 1972 and Osborne and Polzin 1979.

Total cost of surgery incurred:

The cost of surgery involves the cost of anaesthesia, site preparation, suture material and postoperative medicines. In the present study minimum cost was incurred in animals of group A where the OH done through flank approach with the use of subcuticular suture. Though the animals of this group utilized subcuticular absorbable suture, the cost of surgery was still lower than animals of group B which undergone OH through right flank with external skin suture, it might be due to the fact that subcuticular suture take less time to suture and therefore less amount of anaesthesia used and another important factor is that minimum postoperative care was required in the animals which use subcuticular suture therefore less postoperative medicines were used. The subcuticular suture are more advantageous in comparison to external skin suture in present study as external skin suture cause more amount of irritation and the habit of dogs to chew the sutures located on skin. Similarly, Rice and Dewell, (1976) used subcuticular gut suture instead of skin suture for closure of laparotomy wound in puppies for the same reason. Aronsohn and Fagella, (1993) also used 3-0 nylon for closure of subcutaneous layer following OH in Kittens. Theran, (1993) also stressed that skin should be closed with a subcuticular layer of sutures after spay operation. While Olson and Bruce (1986) used tissue adhesive and autoclips for closure of laparotomy wound following OH in rodents and rabbits. In the present study the subcutaneous suture were use employing Vicryl (P.G.A.) as a better choice over chromic catgut because Vicryl was found more soft pliable and

flexible as compared to catgut which is agreement with the observations of Chawla and Rajoria, (2002). Freeman *et al.* (1987) was also used subcuticular Vicryl suture to avoid seroma formation. Although there was a marginal differences in the cost of surgery in the groups of animals which undergone OH through flank approach (group A and B) or through mid-ventral approach (group C and D), respectively, however there was a significant more cost of surgery in mid-ventral approach in comparison to flank approach. The maximum cost of surgery was in group D animals where the OH done through mid-ventral approach with external skin sutures.

Hematological effects:

TLC and **DLC** are the indicators of infection, toxin, trauma, as well as hemorrhage. In the present study, no significant differences were observed in the values of TLC, neutrophil, lymphocyte, monocyte and eosinophil in different groups of animals. However, an increase in TLC, neutrophil, monocyte and decrease in lymphocyte and eosinophil was observed at 24 to 48 hrs after operation in all the groups of animals. The changes were significant in animals which underwent OH through flank approach possibly due to more inflammatory response and hemorrhage from this site. However the changes were within normal physiological limits and reflect the typical stress leukogram which is characterized by leukocytosis, neutrophilia, lymphocytopenia and eosinopenia as reported by Millis *et al.* (1992).

Slight decrease in **hemoglobin** and **PCV** after 24 hrs of ovariohysterectomy was observed in all the groups of animals. However the changes were statistically significant in animals of group A and B in comparison to C and D groups, although the changes were physiologically insignificant and can be attributed to hemodilution due to blood loss during surgery or shift of intracellular fluid from tissue to blood after blood loss (Millis *et al.* 1992), or due to fluid retention in response to ADH secretion in

response to sympathetic stimulation as reported by, Korval *et al.* (1974), Collins *et al.* (1997).

Biochemical effects:

Non-significant increases in present study in **serum glucose** value were observed in all the groups of animals with maximum increase after 24hrs of operation and still higher up to 10^{th} day after operation.The increase in serum glucose level in these groups of animals may be accounted for either to decreased glucose utilization, impaired insulin activity or increased level of adrenocortical hormones. Stimuli like pain, trauma, hypovolemia and anxiety can stimulate sympathetic nervous system, which trigger the release of catecholamine from adrenal medulla (Bright and Lantz, 1985). Excessive releases of catecholamines inhibit the pancreatic production of insulin and thereby cause hyperglycemia (Moore, 1972). Excessive sympathetic stimulation due to surgery can also increase the rate of metabolic activities, such as glycogenolysis in liver and muscles leading to realease of glucose into blood. Other factors that contribute to hyperglycemia during anaesthesia and surgery include peripheral carbohydrate breakdown by growth hormone and gluconeogenesis due to realease of glucagons (Clarke, 1970). An increase in blood glucose level has also been observed in dogs with xylazine-ketamine anaesthesia by Kumar and Thurman (1979).

Creatinine levels in serum is a measure of glomerular filtration rate (GFR) as well as protein metabolism (Blood et al, 1995) and it depends on renal function and rate of urine formation. It is a product of deamination of amino acids. Its level in serum is elivated in all types of renal insufficiencies and destruction of muscle tissues. Though in all the groups of animals, an increasing trend in the serum creatinine levels was seen but the values were higher in the groups which underwent ovariohysterectomy through flank approach and values in any of the groups at a given interval did not exceed 1.24 ± 0.12mg/100ml which is within physiological limit.

SUMMARY AND CONCLUSIONS

CHAPTER-7

SUMMARY AND CONCLUSIONS

The present investigation was conducted on 16 clinical cases, which were adult bitches, of 2-6 years of age and weighing between 14 kg to 25 kg. Out of 16 bitches, 8 were presented for neutering, 5 for pyometra and 3 cases of vaginal tumor. In all these cases surgical treatment by ovariohysterectomy was decided along with surgical removal of vaginal tumors (in cases of bitches having vaginal tumor) by episiotomy, and were randomly categorized into 4 groups, for 4 replications in each group and these groups were further subdivided into 2 subgroups based on suture material used and consisting of 2 animals in each subgroup for clinical evaluation of different techniques of ovariohysterectomy (OH), either by right flank or mid-ventral approach.

Ovariohysterectomy (OH) of these animals was done through right flank and midline approach under general anaesthesia. The bitches were premedicated with atropine sulphate (0.044mg/kg b.wt.) and xylazine hydrochloride (0.5mg/kg b.wt.) administered intramuscularly. After 10 minutes a combination of xylazine hydrochloride and ketamine hydrochloride (1:3) was administered intravenously till effect, followed by maintenance of general anaesthesia with the intravenous incremental dosages of same combination as per the need. The animals were restrained either in dorsal recumbency for mid-ventral approach or in left lateral recumbency for right flank approach.

Animals of group A and B underwent OH through right flank approach and closure of abdominal wound after operation was carried out in layers. In the first layer the transverse abdominal muscle with its attached peritoneum was closed and in second layer the internal and external oblique muscles of abdomen were closed together in simple continuous pattern. Whereas animals of group C and D underwent OH through mid-ventral approach and closure of abdominal wound after operation was also carried out in layers. In the first layer the peritoneum was closed and in second layer the linea alba was closed in simple

continuous pattern. Finally the subcuticular sutures were applied for skin apposition by using absorbable suture (half of animals utilized Vicryl and other half utilized chromic gut) in animals of group A and C. While skin was apposed with horizontal mattress suture using nylon in animals of group B and D.

Clinical evaluation of different techniques of OH was done on the basis of clinical observations recorded during preoperative, operative and postoperative periods as below:

1. Clinical observations of the temperature, pulse and respiration rates were recorded before operation, at day of operation and after operation on 1st (24hrs), 2nd, 3rd, 5th, 7th and 10th day of operation in all the bitches. The average of morning and evening values were calculated and presented as reading of the day for each parameter.

2. Observations during operation included, the time taken for surgery (duration of operation) and amount of hemorrhage during entire procedure.

3. Observations after operation included,

(a) Status of healing of cutaneous wound: It was evaluated on the basis of oedema, pain on palpation and warmth of incision site at1st (24hrs), 2nd, 3rd 5th, 7th and 10th day after operation in all the bitches.

(b) Postoperative complications: The animals were observed for postoperative complications viz., hemorrhage, exudation from incision line, suture biting, suture dehiscence, evisceration and peritonitis and death at different postoperative intervals with status of wound healing.

(c) Total cost of operation: For individual group of animals was computed based on the cost of anaesthetic, site preparation, suture material and postoperative medicines used.

(4.) The different hematological and serum biochemical parameters viz., Hb, PCV, TLC and DLC and serum glucose and serum creatinine were evaluated at preoperative and postoperative i.e. on

1st (24hrs), 2nd, 3rd, 5th, 7th and 10th day after operation from blood and serum, respectively.

In all the approaches the temperature, heart and respiration rates did not reflect any significant difference within and between different groups at different time intervals. However, the values of heart and respiration rates were slightly decreased at day of operation and increased at 24hrs after operation in comparison to preoperative values.

In the present study, the animals underwent OH through right flank approach (A and B groups), the mean time taken for completion of surgery were 31.25 ± 1.99 and 32.25 ± 1.61 minute, the mean amount of hemorrhage during surgery were 50.00 ± 2.29 and 51.25 ± 1.79 gm. and mean cost of operation were 236.17 ± 7.44 and 250.19 ± 5.41 rupees, respectively. Whereas the animals underwent OH through mid-ventral approach (C and D groups), the mean time taken for completion of surgery were 39.00 ± 2.14 and 44.25 ± 2.17 minute, the mean amount of hemorrhage during surgery were 23.00 ± 2.19 and 25.25 ± 1.65 gm. and mean cost of operation were 280.82 ± 5.15 and301.41 ± 7.60 rupees, respectively.

By applying Duncan's New Multiple Range Test (DMRT), it was inferred that the mean time taken for surgery, cost of operation and amount of hemorrhage during surgery in animals of group A and B, and C and D, did not differ significantly among themselves. However, the mean time taken for surgery and cost of operation was significantly lower in group A and B (right flank approach) in comparison to group C and D (mid-ventral approach). The animals of group A underwent OH through right flank, had lowest mean time of surgery and cost of operation followed by group B, C and D, respectively. However, the mean amount of hemorrhage had lowest in group C, followed by group D, A and B, respectively.

The exteriorization of right ovarian stump up to cutaneous incision was easy with minimum manipulation of abdominal viscera in right flank approach, in comparison to mid-ventral approach. In mid-ventral approach the right ovarian stump was relatively difficult to exteriorize and ligate and

required larger incision for completion of operation and more time for suturing especially when interrupted external skin sutures were placed in place of continuous subcuticular suture.

In the present study, the animals underwent OH through right flank approach, had superiority with respect to status of wound healing and postoperative complications, particularly in those where subcuticular sutures (group A) were used, as the recovery was uneventful without any postoperative complications and with satisfactory wound healing with minimum scar formation. The animals where vicryl absorbable suture have been used have less postoperative complications in comparison chromic gut suture.

The animals of group D, had maximum postoperative complications with regard to suture biting, suture dehiscence and evisceration as has been observed in No.A16 (Chromic catgut) animal and exudation in No.A15 (Chromic catgut) animal, followed by group C, in which exudation was observed in No.A11 and A12 (Chromic catgut) animals, then group B in which suture biting was observed in No.A5 (Vicryl) animal whereas group A animals were recovered uneventfully without any complication.

There was no significant change in hematological and serum biochemical studies in the present study.

On the basis of the observations of the present study it is concluded that the right flank approach with # 2-0 Vicryl subcuticular technique for ovariohysterectomy is best as this technique was less time consuming and cost effective (economic) without any postoperative complication. Although # 2-0 chromic catgut used for abdominal closure, did not shown any adverse effect in right flank approach. It is therefore recommended that the OH for sterilization and in the treatment of pyometera and vaginal tumors should be undertaken through right flank approach with # 2-0 Vicryl subcuticular technique for ovariohysterectomy.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