COMPARATIVE EVALUATION OF CHROMIC CATGUT POLYGLYCOLIC ACID POLYGLACTIN 910 AND POLYDIOXANONE SUTURES FOR LAPAROTOMY WOUND CLOSURE IN CANINES

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CERTIFICATE

This is to certify that the thesis entitled ***"COMPARATIVE EVALUATION OF CHROMIC CATGUT POLYGLYCOLIC ACID POLYGLACTIN 910 AND POLYDIOXANONE SUTURES FOR LAPAROTOMY WOUND CLOSURE IN CANINES"*** submitted by **Mr. MALLIKARJUN, I.D. No. MVNK-1106** in partial fulfillment of the requirements for the award of **MASTER OF VETERINARY SCIENCE** in **VETERINARY SURGERY AND RADIOLOGY** of the **Karnataka Veterinary, Animal and Fisheries Sciences University, Bidar** is a record of bona-fide research work done by him during the period of his study in this University under my guidance and supervision, and the thesis has not previously formed the basis of the award of any degree, diploma, association ship, fellowship or other similar titles.

BIDAR
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Affectionately Dedicated to

My Beloved Parents,

Brother, Sisters and Guide

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LIST OF ABBREVIATIONS

%	-	Per cent
@	-	At the rate of
±	-	Plus or minus
≤	-	Lesser than or equal to
^{0}F	-	Degree Farenheit
dl	-	Deciliter
g	-	Gram
kg	-	Kilogram
mg	-	Milligram
ml	-	Milliliter
µl	-	Microliter
µg	-	Microgram
min	-	Minute
No.	-	Number
Sl.	-	Serial
viz.	-	Namely
et al.	-	Co workers
P	-	Level of significance
SE	-	Standard Error

Introduction

I. INTRODUCTION

Sutures are probably the materials used widely in all fields of surgery (Yugv and Cavaliere, 1983). The search for new and improved suture materials began in Egypt 2000 B.C. where they used linen to close wounds. The ideal suture for an abdominal wound does not exist, it has been clearly shown that suture material in a wound affects the local condition and leads to infection (James and Macleod, 1961). Sutures serve to maintain tissue approximation until a wound attains sufficient tensile strength to prevent dehiscence during normal physiological activity (Wallace *et al*., 1970).

Even today, there is a search for ideal suture material. Every surgeon tries to use the best suture material for the patient in given circumstance. Suture materials may be classified according to their behaviour in tissue (absorbable or nonabsorbable), their structure (monofilament or multifilament) or their origin (synthetic, organic or metallic) (Fossum, 1997). Absorbable sutures are those that undergo degradation and rapid loss of tensile strength within 60 days (Bennett, 1988). Nonabsorbable suture are those that retain tensile strength for longer than 60 days (Edlich , 1979).

Natural absorbable suture materials are surgical gut and collagen. Synthetic absorbable suture materials are polyglycolic acid suture, polyglactin 910, polydioxanone, polyglyconate, and polyglecaprone 25 (Slatter, 1993). Galen, Circa 75 AD, was the first to experiment with catgut (Goldenberg, 1959). The word catgut is derived from the term Kitgut or Kitstring (the string used on a kit or fiddle). Catgut has been the most commonly used suture material for visceral organs however, synthetic absorbable sutures are now being widely accepted (Kobayashi *et al*., 1981).

A large number of suture materials have emerged in the last fifty years that differ in physical and biological properties, a new era dawned in 1968 with introduction of polyglycolic acid suture the first synthetic absorbable suture, soon followed by polyglactin 910 following the commercialization of braided, synthetic, absorbable sutures came the introduction of absorbable, synthetic monofilament sutures in the 1980'S both polydioxanone and polyglyconate (Busic *et al*., 2003).

Polyglycolic acid suture is a braided multifilament polymer of glycolic (hydroxyacetic) acid. Absorption is by hydrolysis, not phagocytosis, presumably through esterase activity (Stashak and Yturraspe., 1978). Polyglactin 910 is a braided synthetic fiber composed of glycolic and lactic acids in a ratio of 9:1 (Craig, 1975). Polyglactin 910 coating of polyglactin 370 and calcium sterate gives polyglactin 910 excellent handling and smooth tying properties (Moy *et al.,* 1991). Polydioxanone is polymer of paradioxanone available as a monofilament (Ray, 1981).

The ideal suture is one that can be used under all circumstances in every operation, such a suture showed include high tensile strength, low tissue reactivity, ability to form secure knots, easy handling, elasticity, flexibility, low cast and lack of package memory, till date no one suture possesses all these attributes.

Formation of tissue adhesions after abdominal surgery is usually considered a complication. Post-operative adhesions can result in impaired intestinal motility and low obstruction and predispose the animal to inadvertent incision of abdominal organs during entry to the abdomen in subsequent surgical procedures (Mark *et al.,* 1996).

Chromic catgut suture material has several undesirable characteristics (Laufmann and Rubel, 1977). Absorption of catgut from tissue is unpredictable and its strength varies, especially in the presence of enzymes, secretions and infection (Burrows and Harvey, 1973). Its tensile strength, strength retention and knot security in tissue are less dependable and generally inferior to those characteristics of synthetic absorbable suture materials (Swanson and Tromovitch, 1975). References to available literature revealed that no systematic work has been carried out to study the comparative evaluation of different suture material for laparotomy wound closure in canine. The present study was undertaken with the following objectives:

1) To evaluate different suture materials for canine laparotomy wound closure and to evolve a better suture material for canine laparotomy wound closure.

2) To assess haematological, biochemical and physiological changes before and after surgery in order to assess the inertnesss of suture material for canine laparotomy.

3) Histopathological assessment of suture material reaction in the tissue.

Review of Literature

II. REVIEW OF LITERATURE

The review of literature available is presented under the following headings.

2.1 Chromic catgut

2.2 Polyglycolic acid

2.3 Polyglactin 910

2.4 Polydioxanone

2.5 Surgical technique

2.6 Anesthetic protocol

2.7 Physiological, biochemical and haematological parameter

2.8 Histopathology

2.1 Chromic catgut

Madsen (1953) reported intense inflammatory reactions to chromic catgut suture materials in rabbits.

Goldenberg (1959) reported that in 1869, Lister developed the concepts of both impregnating chromic acid in catgut and sterilizing suture materials.

Lawrie *et al.* (1960) reported intense inflammatory reactions to chromic catgut suture materials in rats.

Lawrie *et al*. (1960) reported that catgut causes less tissue inflammation in dogs than in rats and rabbits.

Lilly *et al. (*1968) postulated that multi-filament suture materials permitted a ‘wicking phenomenon’ that could advance oral fluids and bacteria along the suture filaments by capillary action. The wicking effect was evidenced by swelling and fragmentation with necrotic debris and exudates among the suture fibers.

Goligher (1975) showed that catgut had insufficient strength for use in abdominal wound closure, being assosciated with 11% incidence of dehiscence and reported that catgut as the only suture material that was associated with an unacceptably high rate of wound failure. Reported 11 bursts in 100 paramedian incisions closed with catgut. The reason for that high incidence of dehiscence was that catgut did not retain its tensile strength longer than 10 days and that was not long enough for the linea alba or rectus sheath to heal strongly.

Hastings *et al*. (1975) reported that prolene and plain and chromic catgut produced the least and silk produced the greatest cellular reaction in the stomach and colon of mongrel dogs.

Tauber *et al.* (1975) concluded that catgut caused inflammatory reactions within the wound area leading to loss of strength of the sutured tissue, thus incisions of the abdominal fascia should not be closed with catgut.Whereas, polyglycolic acid produced minimal inflammatory reaction. The nonabsorbable suture material polyester showed satisfactory strength and minimal tissue reaction.

Winkle *et al.* (1975) studied in subcuticular closure of abdominal incisions in dogs both plain and chromic catgut, produced only a mild cellular reaction in dogs after 21 days.

Knowles (1976) studied that chromic catgut was degraded by the activity of proteolytic enzymes, macrophages and giant cells. In many species, the tissue reaction was locally severe because, except in cattle and sheep it acts as xenograft.

Stashak and Yturraspe (1978) reported chromic gut was an organic, monofilament, absorbable suture material that was commonly used in mammals and birds.

Varma *et al.* (1981) studied in the infected wounds with implants of plain catgut, chromic catgut, silk and braided Dacron there were large numbers of neutrophils even in chronic implantation, indicating persistence of local infection.

Varma *et al.* (1981) studied the degree of swelling in surgical wounds after implantation of suture material, which were inoculated with 3 dilution of Staphylococcus aureus. Monofilament nylon and multifilament steel produced the least amount of swelling, plain and chromic catgut produced the most. The BPGA (braided polyglycolic acid), BD (braided Dacron) and silk caused swellings that were intermediate in degree.

Bellinger (1982) reported that catgut, which was made from bovine or ovine intestine, was treated with chromium salts to increase its tensile strength, delay its absorption, and decrease its tissue reactivity.

Freeman *et al.* (1987) studied in the linea alba of cats the chromic gut sutures induced an inflammatory reaction that progressed from moderately purulent to severely fibromononuclear to fibrous in 14 days.

Mathes *et al*. (1987) reported the tensile strength half-life for various sutures was chromic gut, 1 week; poliglecaprone (monocryl), 1 week; polyglactin (Vicryl) and polyglycolic acid (Dexon), 2-3 weeks; polyglyconate (Maxon), 3-4 weeks; polydioxanone (PDS) 4-5 weeks. Some nonabsorbable suture also lost tensile strength over time when implanted in tissues.

Scheidel and Hohl (1987) stated that catgut suture was made by twisting together strands of purified collagen taken from the serosal or submucosal layer of small intestine of healthy ruminants (cattle, sheep and goats) or from beef tendon. They also stated that with advent of synthetic absorbable suture materials, they had no doubt that catgut and chromic catgut can be declared obsolete in gynaecological surgery

Bennett (1988) reported that chromic catgut initially causes greater tissue reactivity than do synthetic absorbable sutures. The reaction was believed to result from antigenic response to the proteins in the suture.

Aldahash *et al.* (1990) studied the comparison of chromic catgut, silk and nylon for uterine closure in ewe and concluded that, tissue reaction on day 7 was more intense to catgut and was the least to nylon.

Gilliland (1994) showed that in largemouth bass maintained at 18°C, chromic gut had a mean absorption time of five weeks, suggesting that sutures of the material might be suitable when healing was expected to be rapid and rapid absorption of the suture was desirable.

Greenwald *et al*. (1994) evaluated 10 suture materials in rats, chromic gut induced the most severe inflammation, with extensive cellular infiltrates and 'wide reaction zones.'

Bennett *et al*. (1997) studied in rock doves tissue reaction to chromic catgut induced a prominent granulocytic infiltrate that gradually resolved after 30 days.

Wainstein *et al*. (1997) reviewed the hypothesis that the use of plain catgut would result in many complications because of its animal origin.

Outlaw *et al*. (1998) showed an increased diameter of chromic catgut and multifilament sutures that was attributed to the infiltration of inflammatory tissue which had an impact on breaking strength.

Selvig *et al*. (1998) studied braided sutures seemed to conduct bacterial migration to a great extent than monofilament sutures. Even immobile bacteria were transported inside multifilament suture materials, where the cellular and immunological defence against them was considerably impaired.

Hurty *et al*. (2002) reported chromic gut sutures might not be appropriate for use in koi (Cyprinus carpio) skin, especially if the sutures were required to remain in place for longer than seven days.

Yaltrik *et al*. (2003) studied that catgut suture materials showed the highest inflammatory reactions and expansion in soft tissues of rats compared with the synthetic ones such as Vicryl and Polypropylene and were identified as the most inconvenient suture materials.

Subhash *et al.* (2004) reported that in mini laparotomy spaying, suture line dehiscence was observed in 42 out of 6475 bitches which underwent laparotomy wound closure by catgut.

Ajadi *et al.* (2006) compared chromic catgut and nylon suture materials for the closure of a pyloric incision in the dog reported that total leukocyte count was significantly higher on day five where chromic catgut 2-0 was used to close the pyloric incision.

Hadjipour *et al*. (2008) reported that raw materials of chromic catgut sutures which were from the intestine of particular animals were different, the pathological effects of these materials were variable and this matter was not matched with the rules of standards about the materials being used in medicine and veterinary medicine. So, these suture materials had better not be recommended anymore.

Asvin *et al*. (2009) studied chromic gut (CG), seems to sustain its strength better than PG (polyglactin) and PG-FA (polyglactin fast absorbing) after 2 weeks. PG-FA may not be a desirable suture if tensile strength was required after 10 days.

James *et al*. (2009) stated that in their study chromic catgut had straight pull strength of 4.11 kgf and Knot pull strength of 2.05 kgf. Chromic catgut took 7 to 10 days for 50% loss of tensile strength, 14 to 21 days for complete loss of tensile strength and 90-120 days for complete mass absorption, chromic catgut had initial stiffness of 2.77×10^4 kg/cm^2.

Akinrinmade and Lawal (2010) reported that the use of polypropylene with superior suture qualities should be considered instead of chromic catgut for procedure in which adhesion was desired in the dog.

Ramazan *et al.* (2010) concluded polypropylene proved to preserve its stability in vivo and in vitro surveys. Catgut and caprosyn lost its tensile strength in all media (bladder, stomach, intestine and bile duct, urine, bile of rat, pH 1 and pH 10) silk and biosyn lost its tensile strength in all conditions except the stomach and intestines. Maxon also lost its tensile strength in all condition except urine.

Archana kumari *et al.* (2011) revealed that unabsorbed suturing materials in addition of inflammatory tissue reaction in the incised area, formation of collagen fiber and haemorrhage was also marked in muscular tissue on 10^{th} post-operative day where laparotomy was duly repaired by chromic catgut. Whereas, study conducted on tissue sample (20^{th} day) marked with, vacant area formed by absorption of catgut and had been filled with fibroblast cells in the incised area.

Sagu *et al*. (2011) concluded that, non absorbable suture material (monofilament non braided silk) was comparatively more effective and successful in canine perineal herniorrrhaphy than absorbable (chromic catgut, polyglactin 910) sutures.

2.2 Polyglycolic acid

Forester *et al.* (1969) reported that the strength to the wound was provided by shape of fiber and weaving of wound collagen.

Katz (1970) studied the presence of aliphatic ester bonds in polyglycolic acid and polyglactin 910 suture materials renders them hydrolytically unstable, they degrade by hydrolysis from body fluids as a result, the tensile strength of the suture materials decreases with an increase in the duration of immersion in a saline solution.

Bergman *et al*. (1971) suggested that Dexon sutures were unsuitable for use in urogenital surgery because of the risk of calculus formation and accelerated hydrolysis.

Elert *et al*. (1971) reported that polyglycolic acid sutures were inert, non collagenous, non antigenic, non pyrogenic and had a high tensile strength and caused minimal tissue reactions which was confirmed clinically.

Edlich *et al.* (1973) reported that the polymorph reaction to bacteria in polyglycolic acid suture was less marked then either silk or multifilament nylon, which might represent an antibacterial effect of polyglycolic acid. Giant cell invasion was slowed in the infected sutures with a consequent decreased absorption, especially with polyglycolic acid.

Postlethwaite *et al*. (1975) studied the evidence of abrupt (48h) degeneration of chromic catgut placed intraluminally in the stomach of dogs and demonstrated that polyglycolic acid maintained half its initial breaking strength in dogs 7 days after implantation.

Urdhal (1975) studied tests on the tissue tolerance of an absorbable, synthetic suture material polyglycolic acid in horses and dogs, concluded that tissue reactions were much milder towards polyglycolic acid suture material than towards chromic catgut and

also concluded that polyglycolic acid was successfully used for operations on skin, muscle, fascia and gastro intestinal tract.

Milroy (1976) reported that the presence of infection within the bladder enhanced the dissolution of Dexon sutures, this early dissolution was tentatively attributed to an increase in bladder p^H level to as high as 8.39 as a result of urea splitting microorganism (proteus) degrading the urine to ammonia.

Salthhouse and Matalaga (1976) showed that primary biodegradation of PGA sutures occured independent of cellular activity, the only requirement for suture degradation was an aqueous environment.

Kronborg (1976) concluded the use of polyglycolic acid nearly eliminated the risk of suture granulomas without increasing the risk of wound dehiscences.

Williams *et al*. (1977) studied that certain enzymes such as esterase and carboxypeptidase were able to influence the rate of hydrolysis in PGA.

Kaminski *et al.* (1978) absorbable sutures were considered better suture materials than non absorbable when they were used in the urinary tract.

Pavan (1979) reported that polyglycolic acid and polyglactin 910 suture materials which elicit minimal tissue reactions retain better strength than catgut under certain physiologic and pathologic conditions and perform more consistently due to precise and reproducible manufacturing processes.

Cameron *et al*. (1980) reported there was no difference in the rate of infection, disruption or herniation between the polypropylene and polyglycolic acid for abdominal wound closure in human.

Bucknall (1981) reported polyglycolic acid loses over 90% of its strength within three weeks. Whereas, the abdominal wall fascia required about 120 days to regain its strength. The use of synthetic absorbable material for abdominal wall closure therefore required careful monitoring.

Varma *et al.* (1981) studied cellular response to wound infected with staphylococcus aureus using seven suture materials (braided polyglycolic acid, multifilament stainless steel, monofilament nylon, black braided silk, braided Dacron, and plain and chromic catgut) in 60 dogs and observed large number of neutrophils in wound containing plain catgut, chromic catgut, silk. Whereas, in nylon and polyglycolic acid the number of neutrophils decreased rapidly.

Askew (1983) reported that upper abdominal surgical wounds should be closed with a continuous nylon suture, there was significantly greater incidence of wound infection and incisional hernia in the group closed with polyglycolic acid sutures in a trial of 104 consecutive patients undergoing upper abdominal surgery.

Lober and Fenskena (1986) reported that polyglycolic acid was an absorbable braided synthetic homopolymer of glycolic acid, it was supplied as an uncoated or coated form. Coated polyglycolic acid suture was undyed or dyed green, violet or bicolored.

Sharma *et al*. (1986) concluded that biopsy collected from flank region of male buffalo calf, polyglycolic acid sutured wounds exhibited slightly higher values of hexosamine than catgut sutured wounds.

Sharma *et al.* (1991) reported that polyglycolic acid produced very mild inflammatory and higher proliferative changes with laying down of fibroplasias in clean wounds when compared to moderate inflammatory changes produced by catgut.

Wetter *et al.* (1991) reviewed that suture material could influence the incidence of wound infection and that was less frequent when polyglycolic acid sutures were used regardless of whether the appendix was inflamed or normal compared with catgut and nylon for appendicectomy wound closure.

Sharma *et al.* (1992) studied polyglycolic acid and catgut, polypropylene and linen in clean, infected and infected experimental wounds in buffaloes. The tensile strength of these suture materials was measured on 3^{rd}, 6^{th}, 9^{th}, 15^{th}, and 90^{th} days after implantation. Polyglycolic acid had significantly higher tensile strength amongst absorbable sutures and prolene amongst nonabsorbable sutures.

Kiilholma *et al.* (1994) studied Maxon was as safe and efficacious as Dexon plus for abdominal hysterectomy and its handling properties were even better than those of dexon plus.

Luijendijk (2000) reviewed polyglycolic acid were the most commonly used rapidly absorbable suture materials by surgeons, such materials were lost for 15 to 90 days, although most of their tensile strength was lost in 14 to 21 days.

Rucinski *et al*. (2001) reported rapidly absorbable suture materials had been associated with increased rates of incisional hernia formation when compared with nonabsorbable sutures.

Stridsberg and Maria (2002) stated that polyglycolic acid could be obtained through ring opening polymerization of glycolic acid.

Gunatillake and Adhikari (2003) explained that polyglycolic acid degradation process took place in two steps during which the polymer was converted back to its monomer glycolic acid. First water diffused in to amorphous regions had been eroded, leaving the crystalline portion of the polymer susceptible to hydrolytic attack. The degradation product, glycolic acid was nontoxic and it could enter the tricarboxylic acid cycle, after which it was excreted as water and carbon dioxide.

Tan *et al.* (2003) stated that polyglycolic acid was a braided suture composed of the homopolymer of glycolic acid and it was rapidly absorbed and quickly lost its tensile strength.

James *et al*. (2009) stated that in their study polyglycolic acid suture took 14 to 21 days for 50% loss of tensile strength, 28 days for complete loss of tensile strength and 60 to 90 days for complete mass absorption, polyglycolic acid had an initial stiffness of 1.15×10^2 kg/cm^2.

Tiberiu (2011) stated that polyglycolic acid degradation was faster in vivo than in vitro and this phenomenon was thought to be due to cellular enzymatic activity.

Balamurugan *et al.* (2012) reported polyglycolic acid suture was more superior for minor oral surgical procedures compared with silk. It had less tissue reaction, better handling characteristics and knotting capacity.

2.3 Polyglactin 910

Watts (1975) reported that residual tensile strength of a polyglactin 910 suture was consistently greater than that of polyglycolic acid suture.

Winkle *et al*. (1975) concluded polyglactin a synthetic absorbable suture, produced a moderate tissue reaction and uniformly disappeared between the twenty eighth and seventieth days where polyglactin suture was used as subcuticular closure of abdominal incisions in dogs.

Bellenger (1982) reported that polyglactin 910 was considered a strong material that gave good knot security and induced a mild tissue reaction in many species.

Driscoll *et al***.** (1982) evaluated the uterus tissue reaction in rabbits using absorbable and nonabsorbable microsutures (nylon and coated polyglactin 910) and reported that after 35-70 days there was a lesser tissue reaction if absorbable sutures were used.

Schoenenberger *et al*. (1985) treated blunt injuries of kidney in 12 pigs with polyglactin mesh and catgut sutures. There were fewer adhesions, less kidney scarification and less kidney atrophy if the polyglactin 910 mesh was used.

Freeman *et al*. (1987) reported tissue reaction with polyglactin 910 suture in the linea alba of cats developed from a mildly purulent reaction after one day to a severely fibromononuclear response after 14 days.

Moy *et al.* (1991) stated polyglactin 910 suture (Vicryl rapid) was a copolymer of lactide and glycolide with a lower molecular weight than vicryl suture, manufactured with a coating of polyglactin 370 and calcium sterate. This lubricant coating gave vicryl excellent handling and smooth tying properties.

Ansari (1992) studied polyglactin 910 and polyamide 6 sutures were compared employing both mass and layered closure technique. Layered closure with polyglactin 910 developed maximum number of adhesions to laparotomy scar.

Sahlin (1993) concluded that closure of an abdominal incision could be effected by a monofilament continuous absorbable suture polyglyconate (maxon) more quickly than by multifilament (polyglactin 910) interrupted absorbable sutures without an increased risk of wound dehiscence or incisional hernia.

Miro *et al*. (1995) evaluated the wound breaking strength and healing after suturing non injured tissues in mice. Silk sutures, polypropylene sutures and polyglactin 910 sutures were compared. The breaking strength for polyglactin 910 sutures was smaller than for polypropylene sutures.

Denardro *et al.* (1996) stated polyglactin 910 was a synthetic, multifilament absorbable copolymer of glycolide and lactide. It was slowly degraded by hydrolysis and was more susceptible to break down and absorption in alkaline environments and

reported that the tissue reaction to polyglactin 910 sutures placed in the oral cavity of cats was moderate to marked and that bacteria were consistently observed in the interstices of the braided suture.

Guyuron and Vaughan (1996) reported that handling of polyglactin 910 suture was more acceptable to most surgeons than catgut. However, it was a subjective rather than objective remarks. They also observed that, polyglactin 910 was nonpyrogenic and nonantigenic and there was minimal tissue reaction during resorption.

Knote and Bohmert (1996) reported that polyglactin 910 (vicryl rapid) was a relatively new, rapid absorption synthetic multifilament skin suture material.

Bennet *et al.* (1997) showed that in the body wall of rock doves, the reaction to polyglactin 910 was characterised by fibroblasts, macrophages and multifilament giant cells and was the most severe induced by the suture materials they evaluated.

Wainstein *et al*. (1997) concluded polyglactin 910 appeared to be the best suture for pyeloplasty in the rabbit because of the mild inflammatory response and rapid tissue reabsorption complete by 5 weeks.

Aderriotis and Sandar (1999) studied irradiated polyglactin 910 (Vicryl rapid) was a useful suture material with both intra and extra oral applications in the pediatric and adult populations.

Gabel *et al*. (2000) compared polyglactin 910 and nylon in the closure of punch biopsy sites, for evaluation of redness, infection, dehiscence, scar, hypertrophy and

patient satisfaction and concluded that absorbable sutures were good alternative in the primary closure of skin biopsy sites.

Sand *et al.* (2001) concluded polyglactin 910 mesh was found to be useful in the prevention of recurrent cystoceles.

Hurty (2002) reported that in the koi (Cyprinus Carpio) the median inflammatory score of the tissue reactions to polyglactin 910 was moderately severe after seven days and moderate after 14 days.

Rothenburger *et al.* (2002) concluded that coated polygalctin 910 suture with triclosan provides antimicrobial effect sufficient to prevent in vitro colonization by Staphylococcus aureus and Saphylococcus epidermidis.

Busic *et al.* (2003) observed by the microscopic analysis that there were more inflammatory reactions (foreign body granulomas) with catgut and there was not any difference in scar formation in sheep between subcuticular plain catgut and polyglactin 910 sutures.

Yaltirik *et al.* (2003) concluded Vicryl as a suture material produced the mildest tissue reaction during early healing period in soft tissues of rats compared with catgut, silk and polypropylene sutures.

Pamela *et al.* (2004) studied four different suture materials were used to close surgical wounds in Juvenile sea turtles. Gross and histopathologic results indicated that

poliglecaprone 25 and polyglyconate caused significantly less crust formation and panniculus inflammation than chromic gut and polyglactin 910.

Brackeen *et al.* (2005) reviewed irradiated polyglactin 910 provides option for the placement of full thickness skin grafts without the need for suture removal.

Drazen *et al.* (2005) studied polypropylene (prolene) and polyglactin 910 (vicryl rapid) for skin surgery in dogs. The handling characteristics of prolene suture was less satisfactory than vicryl rapid suture. Vicryl rapid had a better control of knotting then prolene. In dogs whose wounds were sutured with vicryl rapid, the threads only have to be wiped away or have already fallen off by themselves. Vicryl rapid suture were interesting for veterinary skin surgery in wild and zoo animals, non-socialized pets and wounds under casts.

Nathalie and Leroux (2006) studied synthetic suture material had better results than catgut in cervix surgery.

Aboutalebi and Wells (2007) studied irradiated polyglactin 910 (IRPG) produced low inflammatory response, rapid degeneration, soft feel, and easy workability. It was an ideal suture option for closure of mucosal defects and areas where short term wound support was desired.

Eric *et al*. (2009) reported the maxon suture showed greater suture tensile strength when compared with vicryl, catgut, caprosyn and Biosyn. The presence of E.coli and higher urine p^H led to greater suture degradation however, these were not statistically significant.

Sagul *et al.* (2011) advised that nonabsorbable (monofilament non braided silk) suture should be preferred over absorbable suture (chromic catgut and vicryl) for standard perineal herniorrhaphy in dogs.

Bilal *et al.* (2012) reported that absorbable polyglactin 910 sutures could be used for wound closure after thyroid surgery as it would not require suture removal and thus prevents patient anxiety and discomfort.

Justinger *et al.* (2012) reported fast absorbable sutures with antibacterial coating (triclosan) do not increase the hernia rate after mid line abdominal incision compared with slowly absorbable sutures, when wound infection rates were decreased by coating the fast absorbable suture with triclosan. The development of incisional hernia was significantly increased in patients with a body mass index >30 kg/m.

2.4 Polydioxanone

Cassie (1977) reported that polydioxanone caused a minimal tissue reaction, unlike catgut and therefore should be less likely to promote infection.

Corman (1981) concluded that long term absorbable suture material maybe the most appropriate choice for abdominal wall closure after bowel operations because the absence of suture sinus formation and the failure to demonstrate an increased incidence of wound dehiscence or incisional hernia.

Leese *et al*. (1984) reported that synthetic absorbable sutures were known to carry an increased risk of wound failure and it would be of value to determine the incidence of late herniation following polydioxanone closure compared with nonabsorbable sutures.

David *et al.* (1985) reported that polydioxanone suture might be an alternative suture to nylon for laparotomy closure, no wound failures occured after nylon closures but two occurred in wounds closed with polydioxanone although they were probably not attributable to suture failure.

Knoop *et al.* (1987) studied in vivo experiments on rats showed a slight tissue reaction and a dissolution time of 120-180 days for maxon and 180-240 days for polydioxanone. With maxon, tensile strength was measurable for 42-49 days, while the period for polydioxanone amounted to 65-80 days.

Krukowski *et al*. (1987) stated suture sinus formation had been reported following abdominal wall closure with polypropylene in humanbeings and dogs and reported that polydioxanone become one of the preferred suture materials for abdominal closure in human beings.

Rodeheaver (1987) reviewed the polydioxanone suture was prepared by polymerizing and extruding the monomer, paradioxanone, in the presence of a suitable catalyst. Stiffness of the polydioxanone suture was 60 per cent greater than that of the modified polyglycolic acid (trimethylene carbonate and glycolide) suture.

Aarnio *et al*. (1988) concluded polydioxanone was a suitable suturing material for small luminal arterial anastomoses and was superior to polypropylene suturing material because it caused no tissue or other late changes on the flow surfaces.

Bourne *et al.* (1988) studied the invivo half–life tensile strength of the monofilament absorbable sutures polyglyconate (maxon) and polydioxanone (PDS) were

3 and 6 week respectively whereas, those of the braided absorbable sutures polyglycolic acid (Dexon plus) and polyglactin 910 (vicryl) was 2 weeks.

Sanz e*t al.* (1988) reviewed that Maxon and polydioxanone elicited a lower degree of chronic inflammation when compared with vicryl and chromic catgut. Maxon and polydioxanone retained a larger percentage of tensile strength during the long post-operative period whereas, vicryl and chromic catgut were mostly absorbed.

Schoetz (1988) stated slowly absorbable suture material (polydioxanone and polyglyconate) were the most commonly used slowly absorbable suture material by surgeons. Absorption of such material took about 180 days and they maintained 50 per cent of their tensile strength for about 4 weeks. Polydioxanone had showed that it had 1.7 times higher tensile strength than prolene.

Shuhaiber *et al.* (1989) reported that synthetic monofilament suture materials evoke less tissue reaction and had less capillary than either natural or synthetic multifilament suture materials, and bacteria were less likely to adhere to them.

Smeak *et al*. (1989) reported that suture sinus formation could be associated with any suture material however, the prevalence was higher following use of multifilament or braided suture materials than following use of monofilament suture material. They recommended suture material in contaminated or infected wounds was synthetic monofilament suture material.

Vipond *et al.* (1991) reported that an absorbable (polydioxnone) subcuticular suture might be safely used in all cases of wound.

Israelsson and Jonssun (1994) reported that suture of midline laparotomy wounds was as safe with polydioxanone as it was with nylon. Incisional hernia was associated more with suture technique than with the material used.

Trostle *et al*. (1994) reported biomechanical studies indicated that the initial strength of polyglyconate and polydioxanone was similar to that of polypropylene and concluded that slower degrading, synthetic monofilament absorbable suture materials such as polyglyconate and polydioxanone, should be considered as alternative to polypropylene to close abdominal incision in horses.

Edwards *et al*. (1995) reported that when nylon sutures were used for skin closure there was an increased incidence of erythema. Polydioxanone had few complications and was recommended in both the young and elderly patients.

Wainstein *et al.* (1997) compared the influence of suture material on pyeloplasty in rabbits. They noted that the most marked inflammatory reaction was produced by chromic catgut while the least reaction was evoked by polyglycolic acid and polydioxanone sutures.

Iwase *et al.* (1999) compared continuous closure of clean and contaminated abdominal wounds using polydioxanone with interrupted closure using braided silk. Incidence of late suture sinus formation in the polydioxanone group was significantly lower than that in the silk group.

Hodgson (2000) stated no statistically significant difference in the incidence of incisional hernia formation, wound dehiscence or infection between the slowly absorbable and the non absorbable suture materials.

Hsiao *et al*. (2000) concluded that abdominal closure with a late absorbable polydioxanone loop suture might be beneficial to patients with a malignant disease for preventing incisional hernia than with polyglactin 910 suture.

Molea *et al*. (2000) reported that with respect to clinical characteristics, tissue inflammatory reaction and suture absorption times, poliglecaprone 25 and glycomer 631 suture materials were found to be less reactive than polydioxanone in rat skin.

David *et al*. (2002) reported for ventral midline closure polyglycolic acid was significantly stronger than polydioxanone and monofilament nylon however, polydioxanone had better mechanical performance after implantation.

Thiede *et al*. (2002) reported that synthetic absorbable suture materials were the first choice in the majority of the procedure by causing lesser tissue damage and inter filament bacterial transport, monofilament sutures were preferable than multifilament ones.

Chelsea *et al*. (2004) concluded polydioxanone, polyglyconte and glycomer 631 might be acceptable for urinary bladder closure in the presence of sterile neutral and E.coli contaminated urine.

John *et al*. (2004) reported polyglactin 9l0 and polydioxanone appear ideal for short term and medium term tissue apposition respectively. Panacryl will be suited to long term tissue apposition.

Tolga *et al.* (2004) studied polydioxanone was the strongest suture material in pancreatic juice among plain, chromic catgut, polyglactin 910, polyglycolic acid, polypropylene and silk sutures. These sutures were incubated in pancreatic juice and bile that was collected from patients.

Fierheller and Wilson (2005) studied larger suture materials (polydioxanone size 7, polyglactin 910 size 6) had higher breaking strengths than smaller suture materials (polydioxanone size 2, polyglactin 910 size 3) and stiffness was also affected by suture material and size.

Sylvain and David (2007) concluded polyglecaprone 25 was an inappropriate suture material for use in teat surgery, polyglycolic acid suture should be avoided in cattle with mastitis, among polyglecaprone 25, polyglycolic acid and polydioxanone. Polydioxanone was best suited for use in teat surgery.

Kosan *et al*. (2008) observed lower degrees of inflammation and foreign body reaction with 4/0 polydioxanone and no stone formation on rat bladder compared with chromic catgut and polyglactin 910.

Justinger *et al*. (2009) reported the use of triclosan coated polyglactin 910 suture materials and antibacterial activity vicryl plus (antibiotic coated) loop suture for abdominal wall closure could decrease the number wound infections after abdominal surgery.

Benito *et al.* (2011) reported that Huaiyin polydioxanone had a greater ($p < 0.001$) ultimate load on days 0, 1, and 7 and constantly greater stiffness on day 14, 21 and 28.

Compared with Ethicon polydioxanone. Ethicon polyglactin 910 had a greater ($p < 0.001$) ultimate load on 0, 1, 7, 14, 21 and 28 and greater stiffnes ($p < 0.001$) at days 0, 1, 7 and 14 compared with Huaiyin polyglactin 910. Huaiyin polydioxanone and polyglactin 910 had greater knot breaking strength than Ethicon equivalent.

Sajid *et al*. (2011) concluded that polydioxanone, prolene and nylon were equally effective for the closure of abdominal fascia following laparotomy, there were no significant difference between two suture materials in terms of risk of incisional hernia, wound dehiscence, pre-operative complications, suture sinus formation and surgical site infection.

2.5 Surgical Technique

Cawley *et al.* (1958) reported complications from the use of nonabsorbable material for tying of the bitch's reproductive tract.

Miller *et al. (*1964) concluded right flank was preferred for better access to the right ovary as it was located slightly cranial than the left ovary.

Schneider *et al.* (1969) reported that ovariohysterectomy not only prevents unwanted pups and kittens from being born, but protects the bitch or queen from future ovarian and uterine disease. If performed early in life, ovariohysterectomy also reduces the risk of mammary tumors in dogs.

Pearson (1970) reported that ovarian pedicle and uterine body could be ligated with either absorbable or nonabsorbable sutures. However, for most ovariohysterectomies absorbable ligatures were preferred by Pearson.

Dorn (1975) concluded that by approach from left side, the omentum covers the viscera and spleen lies there, which get enlarged with induction of barbiturate anesthesia, thus creating difficult approach to ovaries.

Bucknall *et al.* (1982) studied 1129 abdominal operations and demonstrated that layered closure was associated with a significantly higher dehiscence rate compared with mass closure (3.81% Vs 0.76%).

Becker (1985) studied the suturing technique in horse's abdominal wall closure layer by layer with running suture catgut, linea alba with monofilament polydioxanone or polyglycolic acid, subcutaneous layer with running suture of polyglycolic acid. He observed that there were no complications in wound healing in any of the suture materials used.

Jonathan *et al*. (1985) concluded that layered closure of a paramedian incision results in a lower incidence of incisional hernias than mass closure of a midline incision.

Wandstrom (1990) reported the choice of suture material for closing the abdominal fascia should be made in the light of what is known about fascial healing and the physical properties of suture materials (strength, durability, ease of handling and resistence to infection).

Rucinski *et al.* (2001) reported absorbable materials were designed to approximate the fascia during the critical early healing period and subsequently to undergo absorption to avoid the complications of sinus formation, pain and button hole hernia associated with nonabsorbable sutures. The incidence of chronic wound pain and

suture sinus formation had been found to be significantly less with absorbable suture material.

Bohanes *et al*. (2002) reported that the best method of closure of laparotomy was use of absorbable suture material in continuous pattern.

Anurag *et al*. (2004) reported that the risk of abdominal burst less with interrupted suture (1 burst out of 46 patient) methods of abdominal wound closure compared to continuous suture (8 burst out of 54 patient) method.

Vandana (2005) reported that right flank approach for ovariohysterectomy was found to be a simple economical and efficient procedure require very small skin incision (1.6-2.8 cm), less manipulation of visceral organs, short operation period (25-50 min) and less post-operative care with no complications.

Coomer *et al.* (2007) reported that no significant difference in prevalence or rate of wound suppuration in 2 layer closures without subcutaneous suture compared to conventional 3 layer closure.

Fong *et al.* (2008) concluded that closure of an abdominal wound would be best commenced with a loop knot, using gauge 1 polyglyconate.

Singh *et al.* (2011) studied both the ultra sound guided biopsy (USGB) and ultra sound guided fine needle aspiration biopsy (USG-FNAB) were quite effective tools for histoparhological and cytological diagnosis of neoplastic and non-neoplastic abdominal masses.

Murthy *et al.* (2012) compared ventral midline and flank approach of ovariohysterectomy in dogs. A negligible bleeding was noticed flank approach in compared ventral midline due to surgical trauma to the muscles and there was no any serious complications were encountered during and post-operatively in both groups.

2.6 Anesthetic protocol

Sagner *et al*. (1968) stated that the use of xylazine alone produces poor analgesia, and the muscle relaxant effect of xylazine has been reported due to suppression of the interneural transmission of impulses and not probably due to paralysis of neuromuscular transmission.

Miysaka and Momino (1968) reported depressant effect of ketamine was due to the effect of central nucleus of hypothalamus.

Soma (1971) stated that atropine was an anticholinergic (parasympatholytic) drug frequently used to reduce the secretion in the respiratory tract and salivary glands and also to inhibit the vagal stimulation of the cardiovascular and respiratory system.

Antonaccio *et al.* (1973) reported vagal stimulation after xylazine administration.

Kumar *et al*. (1979) reported combination of xylazine and ketamine has been used successfully for variety of clinical surgical procedures in dogs.

Raghavan *et al.* (1979) studied the influence of xylazine as a preanaesthetic drug in dogs in different dosage levels. The dose rate of 1 mg/kg body weight produced a considerable increase in anesthetic period.

David (1980) reported Ketamine and xylazine in anesthetic combination has gained much popularity in veterinary practice because of the very fact that the pressor effect of ketamine seems to offset the cardiovascular depressor activity of xylazine.

Peshin *et al*. (1980) studied xylazine produced good sedation and muscle relaxation at the dose rate of 1-2 mg/kg in the dog.

Booth (1982) reported xylazine acts by stimulating presynaptic α 2 adrenergic receptors thus increasing feedback inhibition of release of norepinephrine from the adrenergic nerve endings.

Kolata and Rawling (1982) reported xylazine (1.1 mg/kg IV) in combination with atropine sulphate (0.045 mg/kg SC) and Ketamine (11mg/kg IV) for dogs.

Mohinder Singh *et al.* (1997) reported the onset of xylazine- ketamine anesthesia occurred within 5-10 minutes and the duration of surgical anesthsia lasted for 30-45 minutes. There was moderate depression of palpebral reflex and absence of pedal and cough reflexes during surgical anesthesia. The eyeball and corneal reflexes disappeared and tongue protruded out of the buccal cavity, the eyes however remained open.

Sharma *et al.* (1997) reported atropine, xylazine- ketamine combination could safely be used as balanced surgical anesthesia for any surgical manipulation in canine under field conditions.

Pathak *et al.* (1998) opined that xylazine could be safely used at the dose rate of 1 mg/kg body weight intramuscularly prior to intravenous barbiturate or dissociative anesthetic in dogs.

Tranquilli *et al.* (2007) reported mild sedative effect in dogs has been observed after atropine administration.

Udegbunam and Igwe (2009) stated that the effect of ketamine anesthesia on the haemogram of dogs has not been adequately studied although daily oral administration of ketamine to mongrel dogs caused a decrease in packed cell volume, total leucocytes and erythrocyte counts.They stated that ketamine was useful for anesthesia in patients in intensive care units and for short duration surgery.

2.7 Haematobiochemical parameter

Gerber (1964) reported creatine kinase activity was very high in skeletal muscle, myocardium, brain and intestine. It was concluded as a sensitive and specific indicator of muscle damage in dogs and horses in their study.

Spurr (1972) reported that increase in glutamic oxaloacetic transaminase (SGOT), glutamic pyruvate transaminase (SGPT), and lactate dehydrogenase (LDH) with hyperthermia might be due to liver, perhaps cardiac tissue damage and that increased body temperature, rather than increased metabolic activity was the prime cause of increased serum enzymes due to heat exposure.

Krausaz *et al*. (1977) reported increase in the levels of transaminases suggests an increase in gluconeogenesis from proteins due to these increased metabolic demands.

Deswal and Chohan (1981) reported transaminases and dehydrogenases were involved in metabolic changes in the body to maintain homeostasis and plasma levels of glutamic oxaloacetic transaminase (SGOT), glutamic pyruvate transaminase (SGPT),

andlactate dehydrogenase (LDH) have been reported to increase with temperature stress in animals.

Schmidt and Booker (1982) reported significant elevation in total leukocyte count, serum cortisol and aspartate aminotransferase level upto 72 hours following ovariohysterectomy in bitches.

Sorenseen, (1989) reported higher level of lactate dehydrogenase indicates the possible presence of malignancy.

Muir and Hubbel (1991) stated that the decrease in erythrocyte count after induction of anesthesia was due to relaxation of splenic capsule leading to pooling of erythrocytes in the spleen of dogs.

Millis *et al.* (1992) studied post-operative haematological profiles of dogs undergoing ovariohysterectomy and stated that post-operative leukocyte differential counts were typical of stress leukograms and were characterized by leukocytosis, neutrophilia, lymphopenia and eosinopenia. Mild decrease in packed cell volume, red blood cell count and haemoglobin concentration were consistent with minor blood loss during surgery or fluid retention and haemodilution post-operatively. Authors concluded that celiotomy and routine ovariohysterectomy in healthy dogs did not alter hematoogical profiles 24 hours after surgery.

Bhaga (1995) concluded that bullock with lower normal blood values of glutamicoxaloacetic transaminase (SGOT), glutamic-pyruvate transaminase (SGPT), and lactate dehydrogenase (LDH), had higher heat tolerance ability.

Selvig *et al.* (1998) suggested that the acute tissue response to sutures early on 1 to 2 post-operative days might be attributed to suturing trauma, which would be similar to all materials.

Dharmaceelan *et al.* (2000) reported significant reduction in the red blood cell count during surgery and post-operative day, with no significant difference in packed cell volume, hemoglobin, neutrophils, lymphocytes, eosinophils, basophils and monocytes between pre and post-operative levels.

Tas *et al.* (2001) reported lactate dehydrogenase catalyses the reversible transformation of pyruvate to lactate, having a principal position in the anaerobic cellular metabolism and found to correlate with malignancy detection, poor prognosis and resistence to chemotherapy and radiotherapy in various neoplastic diseases like germ cell cancer, lung cancer and lymphoma in human beings.

Austain *et al.* (2003) reported that the difference between pre and post-operative creatine kinase varied greatly between bitches (123-2195 U/L) which underwent laparoscopic ovariohysterectomy and stated that creatine kinase was not a predicatable indicator of surgical stress.

Roy *et al.* (2004) concluded serum lactate dehydrogenase value was found 6.67 times increase in malignant conditions and 3.01 times increase in benign disease compared with normal groups in canines.

Dhindsa *et al.* (2008) studed biochemical alteration in bovine caesarean operation with reference to suture material and intra peritoneal lubricant and found increased

concentration of blood urea nitrogen, plasma creatinine and peritoneal fluid fibrinogen and decreased total plasma protein where chromic catgut was used alone.

Alves *et al.* (2009) reported that highest creatine kinase (CK) was found after 3 hour of conventional ovariectomy in queens compared to laparascopic ovariectomy.

2.8 Histopathology

2.8.1 Light Microscopic Study

Edlich *et al.* (1973) reviewed many light microscope histological studies all sutures caused a varying degree of inflammatory reaction in the tissues in which they were implanted due to the trauma of insertion and partly to the physicochemical properties of the suture.

Varma *et al.* (1981) concluded, cellular reaction varied with different suture materials. In general neutrophils were the predominant cells in acute infection however, later macrophages and fibroblasts predominated. Occasionally, plasma cells, lymphocytes, eosinophils, and giant cells were present.

Nary *et al.* (2002) reported fibrosis, angioblastic and fibroblastic proliferation and also the intensity of inflammation was less with poleglecaprone 25 suture followed by polyglactin 910 and polytetra fluorethylene in the dorsal region of subcutaneous tissue of rat which was stained with H and E.

Oloumi *et al*. (2003) studied the histopathological changes in an Iranian brand catgut suture materials for suturing ventral abdominal muscle of rats. Concluded that, the

short term reaction to the suture material ethicon chromic catgut was superior to supra-chromic catgut.

2.8.2 Electron Microscopic Study

Bucknall (1981) reported that scanning electron microscope was ideal, it provided a high resolution, three dimensional image of a relatively large volume of material at high magnification and at low magnification it was possible to correlate the appearance these with those reported using light microscopy.

Chu *et al.* (1982) concluded scaning electron microscopic study of hydrolytic degradation of polyglycolic acid suture was the formation of surface cracks on filaments, cracks were not observed that had not been subjected to hydrolysis.

Bucknall *et al.* (1983) studied electron microscopically that in infected wound, braided sutures of silk, nylon and polyglycolic acid even after 70 days were seen to contain bacteria and polymorphonuclear cells.

Stone *et al*. (1985) studied scanning electron micrographs of zero gauges of chromic gut and glycerine treated chromic gut after 7 days of implantation. reported that uniform surface was present for the glycerine treated chromic gut.

Sharma *et al.* (1986) stated ultra structural studies of biopsy specimens collected on 9^{th}, 30^{th}, 90^{th} post-operative day revealed slightly delayed formation of collagen bundles in early stages of healing in dexon treated wounds as compared to prolene treatment. However, no difference in the healing pattern was observed on 90^{th} post-operative day in buffalo calves.

Miguel *et al*. (2006) concluded by scanning electron microscopy study that, polyglactin 910 stimulated the formation of multinucleated giant cells and its filaments underwent cleavage and dissolution. In polydioxanone a few inflammatory cells and scar fibrosis was observed and triangular cracks appeared on its surface, around the poliglecaprone 25 a diffused infiltration of a few mononuclear cells and fibrosis was recorded and formation of craters was observed on its surface and chromic gut induced necrosis and granulation tissue. These alterations were studied on 1^{st}, 2^{nd}, 3^{rd}, 7^{th} and 14^{th} day after suture implantation.

Materials and Methods

III. MATERIALS AND METHODS

The objective of the present study was to compare chromic catgut, polyglycolic acid, polyglactin 910 and polydioxanone absorbable suture for laparotomy wound closure in canines. Clinical, haematological, biochemical, physiological and histopathological, electronmicroscopic assessment of suture material reaction to the tissue were made before and after surgery in order to assess the tissue reaction caused by the suture material.

3.1 Sources of Research Animals

Clinical cases of canines which were presented to Department of Surgery and Radiology, Veterinary College Bidar and Agriculture Product Marketing centre (APMC) Hospital Bidar for ovariohysterectomy.

3.2 Schedule of the experiment

Twenty four clinical dogs were randomly divided in to four equal groups of six animal each. The details of design of experiment are given in table no 1.

Table 1: Design of technical programme of clinical study

Sl. No	Groups	Number of animal	Surgery performed	Suture used to close laparotomy wound
1	I	06	Ovariohysterectomy	Chromic Catgut[1]
2	II	06	Ovariohysterectomy	Polyglycolic acid[2]
3	III	06	Ovariohysterectomy	Polyglactin 910[3]
4	IV	06	Ovariohysterectomy	Polydioxanone[4]

[1]Trugut (chromic catgut) Suture India Pvt Ltd., peenya industrial Area, Bengaluru.

[2]Petcryl (polyglycolic acid) Futura Surgicare Pvt Ltd., Yeshwanthpur Indl.suburb, Bengaluru.

[3]Petcryl 910 (polyglactin 910) Futura Surgicare Pvt Ltd., Yeshwanthpur Indl.suburb, Bengaluru .

[4]Monofilament polydioxanone (polydioxanone) Futura Surgicare Pvt Ltd., Yeshwanthpur Indl. suburb, Bengaluru.

3.3 Procedure

3.3.1 Preparation of animal and surgical site

All the dogs were restricted for solid food for 12 hours and water for six hours before the surgery. Right flank was shaved, scrubbed with two per cent chlorhexidine (savlon[5]) solution followed by application of surgical spirit.

3.3.2 Pre-operative care

All the animal were kept under antibiotic prophylaxis of ceftriaxone sodium[6] 25 mg per kg body weight IV prior to surgery, anti- inflammatory meloxicam[7] 0.2 mg per kg body weight IM and in post operative period.

3.3.3 Anesthetic procedure

All the animals were premedicated with atropine sulphate[8] 0.045 mg per kg body weight IM. Xylazine hydrochloride[9] 1 mg per kg and ketamine[10] 10 mg per kg mixture in a single syringe administered as single induction bolus and anesthesia maintained with incremental doses of xylazine - ketamine anesthesia by IV route.

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[5]Savlon-Chlorhexidine gluconate soln.IP, 1.5% v/v, Johnson and Johnson Ltd., Baddi.

[6]Intacef -0.5- Ceftriaxone sodium USP, 500mg, Intas pharmaceuticals Ltd., Ahmedabad.

[7]Melonex- Meloxicam IP, 5mg /ml, Intas Pharmaceuticals Ltd., Ahmedabad.

[8]Atropine sulphate IP, 1 mg/ml, 10 ml vial Domesto Ltd., Vijayawada.

[9]Xylaxin-Xylazine injection U.S.P, 23.32 mg/ml, 30 ml vial Indian immunological Ltd., Hyderabad.

[10]Aneket-Ketamine hydrochloride IP, 50 mg/ml, 10ml vial, Neon Laboratories Ltd., Thane.

3.3.4 Surgical procedure

Ovariohysterectomy: The animal was placed in left lateral recumbency. The surgical site was prepared as per standard procedure. The right flank laparotomy was performed. The site of incision was drapped. Oblique incision was made at surgical site (three finger width caudal to the last rib and ventral to lumbar transverse process). Skin and subcutaneous tissues were separated, stab incision was made to the abdominal muscle incision extended cranial and caudal to the stab with Mayo scissors. Right abdominal wall elevated by grasping the peritoneum with thumb forceps. The right uterine horn was located by means of the index finger. A clamp was placed on the proper ligament of the ovary and was used to retract the ovary while the suspensory ligament was broken with the index finger. A window was made in the mesovarium caudal to the ovarian vessels. The ovarian pedicle was triply clamped and the pedicle was severed between the clamp closer to the ovary and the middle clamp. The clamp most distant from the ovary was removed so that the transfixation ligature placed in its groove. Silk was used for transfixation ligature in all the group. The pedicle was grasped with small haemostats, the remaining clamp was removed and the pedicle was inspected for bleeding. The pedicle was gently replaced in to the abdomen and the haemostat was released. Same procedure was repeated on the left ovarian pedicle. Three clamps were placed on the uterine body just cranial to the cervix. The uterine body was severed between the proximal and middle clamps. Both the uterine arteries were ligated separately caudal to the most caudal clamp. The caudal clamp was removed and transfixation of uterine end was done just cranial to the cervix using silk in the groove. The uterine pedicle was grasped with a small haemostat above the clamp, the clamp was removed, and the pedicle was inspected for bleeding. The pedicle was gently replaced in

to the abdomen, and the haemostat was removed. Abdominal wall closed with routine procedure viz., peritoneum and transverse abdominus muscle closed by simple continuous suture pattern, obliques abdominus externus and obliques abdominus internus muscle together by interrupted suture pattern,, using chromic catgut number 1 (plate 1), polyglycolic acid number 1 (plate 2), polyglactin 910 number 1 (plate 3) and polydioxanone number 1 (plate 4) in group I, group II, group III and group IV respectively. Skin wound was approximated by simple interrupted suture using nylon suture material. Post-operatively ceftriaxone sodium was administered at the dose rate of 25 mg/kg body weight IV twice daily (Rebuelto *et al.*, 2002), anti inflammatory meloxicam 0.2 mg/kg I/M to all dogs for three days. Surgical wound was dressed on alternative days using povidone iodine[11] ointment till satisfactory wound healing was observed. Skin sutures were removed on 14th post-operative day.

3.3.5 Biopsy

Obliques abdominus externus muscle biopsy was collected before suturing and on 14th day after suturing by making stab incision at the site of suture under xylazine – ketamine anesthesia.

3.4 Parameters studied

3.4.1 Clinical parameter

Respiratory rate (breaths/minute)**,** heart rate (beats/minute) and rectal temperature **(**°F**)** were recorded in dogs of group I, group II, group III and group IV. According to the standard clinical procedure prior to surgery (0 hours) and 24 hours, 48 hours, 72 hours and 14th days after the surgery.

[11] Betadine - Povidone iodine ointment USP, 5% w/w, Win medicare Ltd., New Delh.

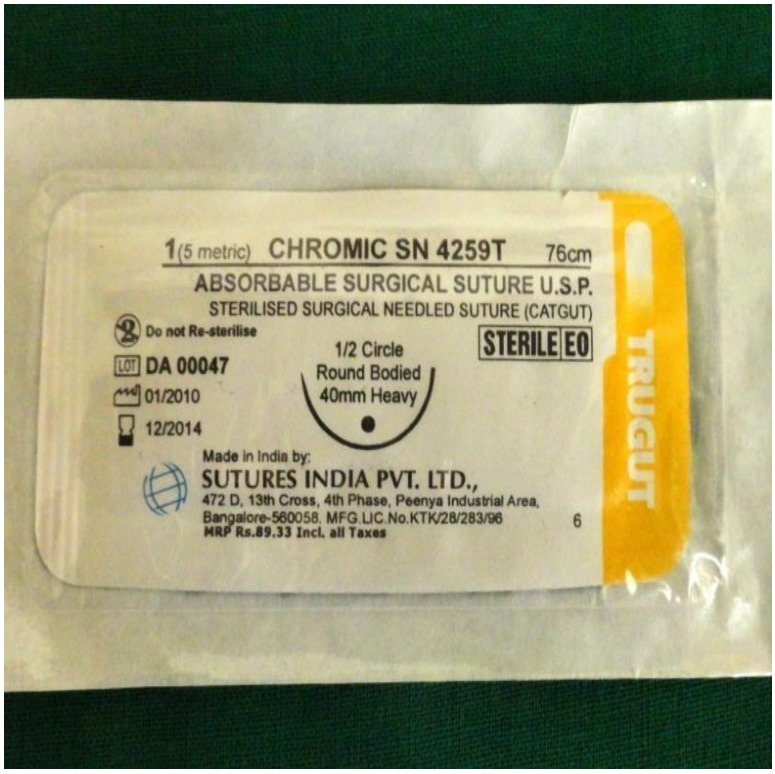


Plate 1: Natural absorbable suture Chromic Catgut (Trugut)

Plate 2: Synthetic absorbable suture polyglycolic acid (petcryl)

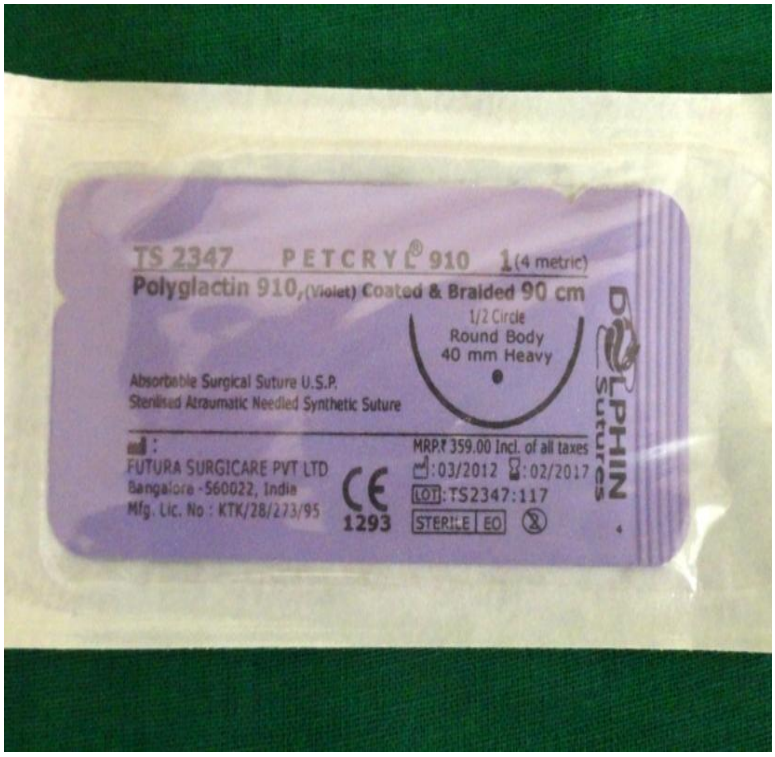


Plate 3: Synthetic absorbable suture Polyglactin 910 (Petcryl 910)

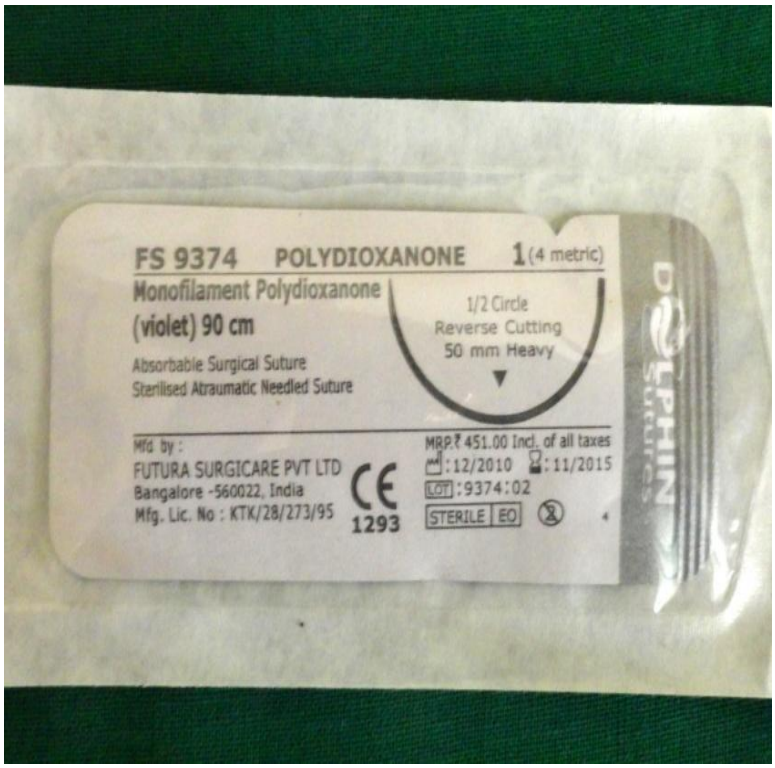


Plate 4: Synthetic absorbable suture Polydioxanone (Polydioxanone)

Group I

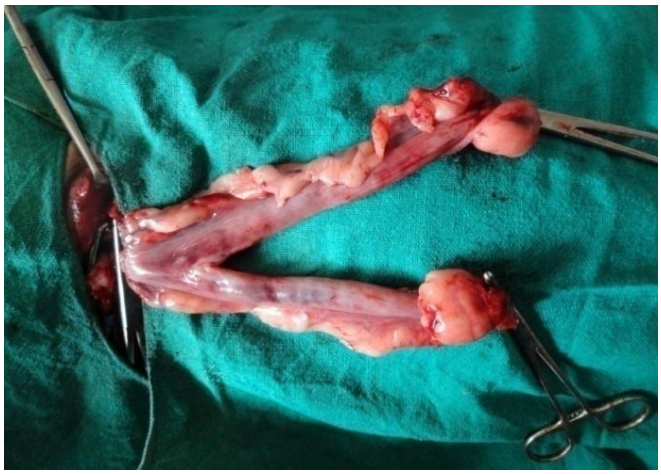


Plate 5: Exteriorised uterus

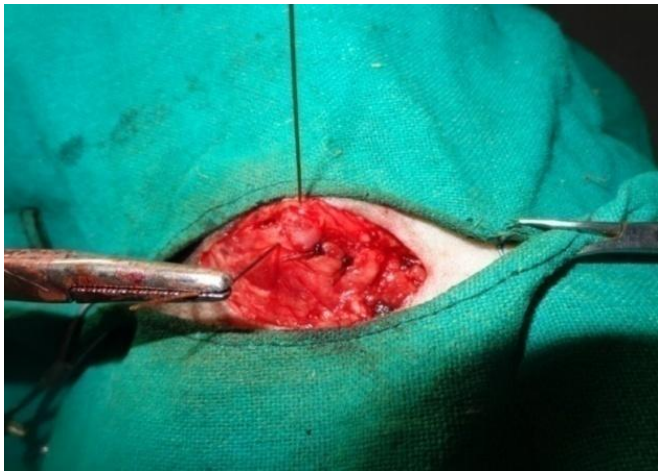


Plate 6: Laparotomy wound closure with chromic catgut

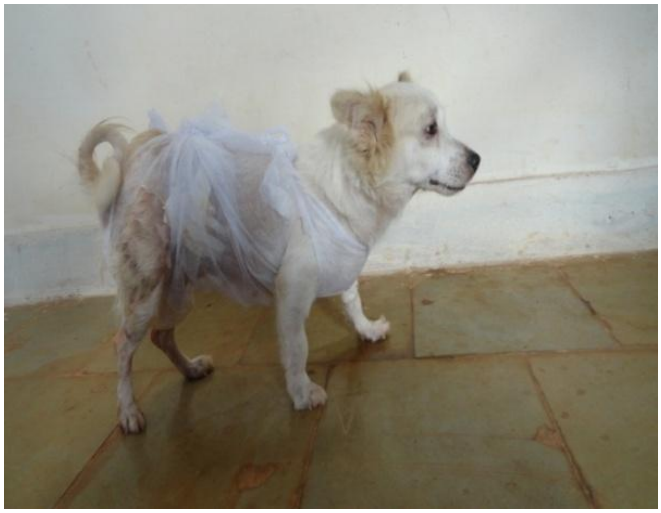


Plate 7: 14th post- operative day

Group II

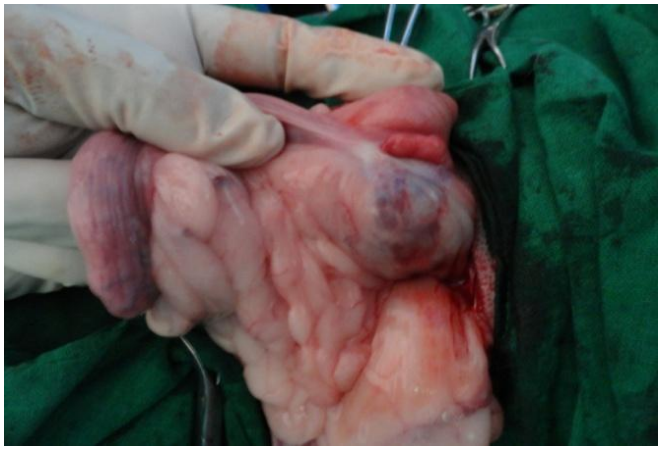


Plate 8: Exteriorised cystic ovary

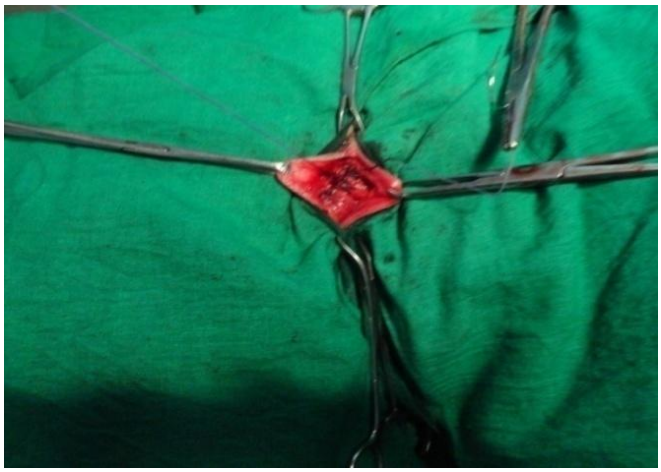


Plate 9: Laparotomy wound closure with polyglycolic acid

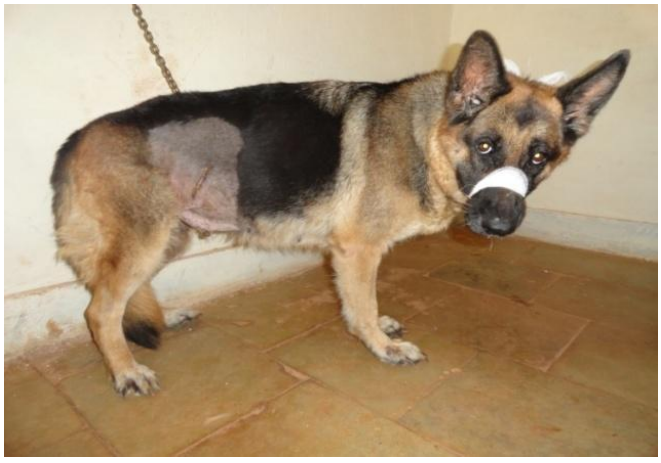


Plate 10: 14th post- operative day

Group III

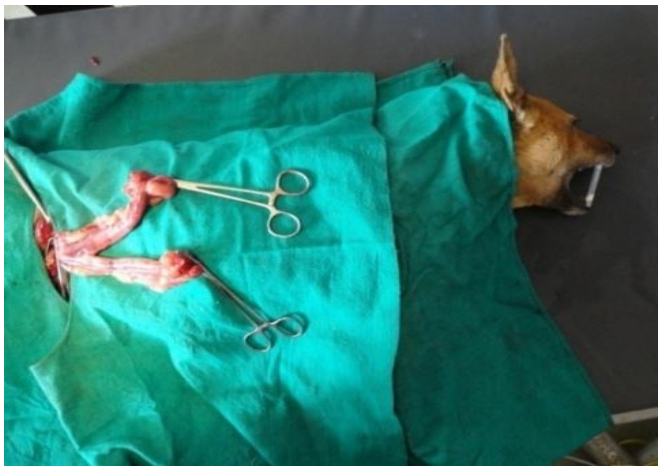


Plate 11: Exteriorised uterus

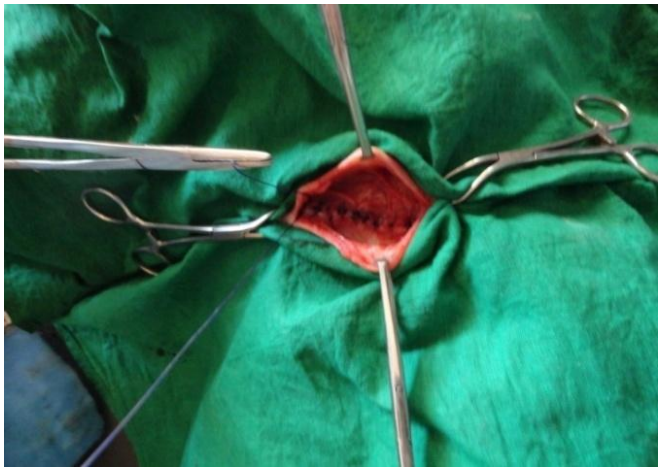


Plate 12: Laparotomy wound closure with polyglactin 910

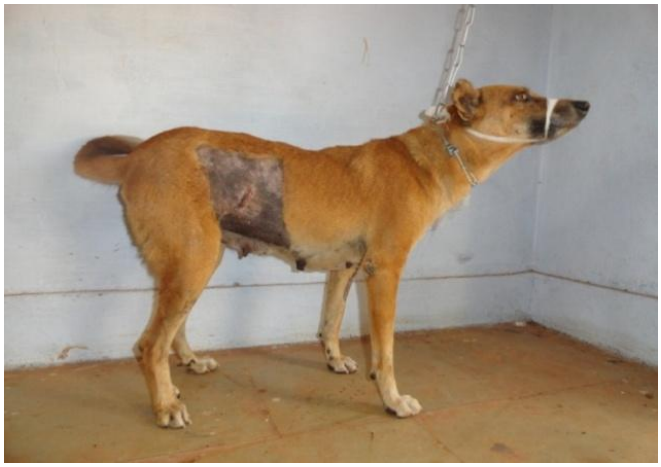


Plate 13: 14th post-operative day

Group IV

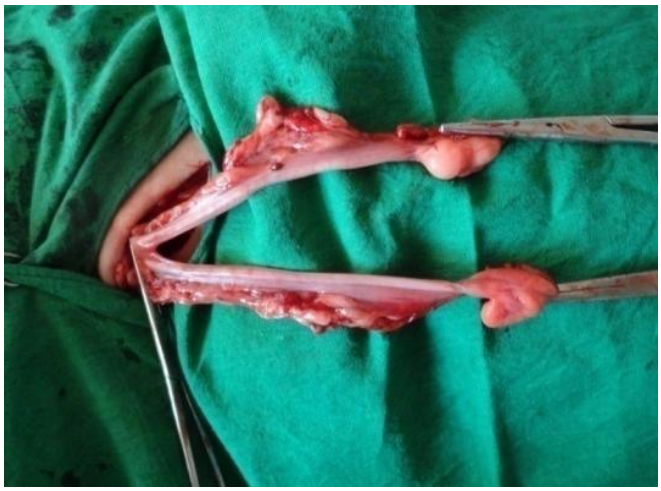


Plate 14: Exteriorised uterus

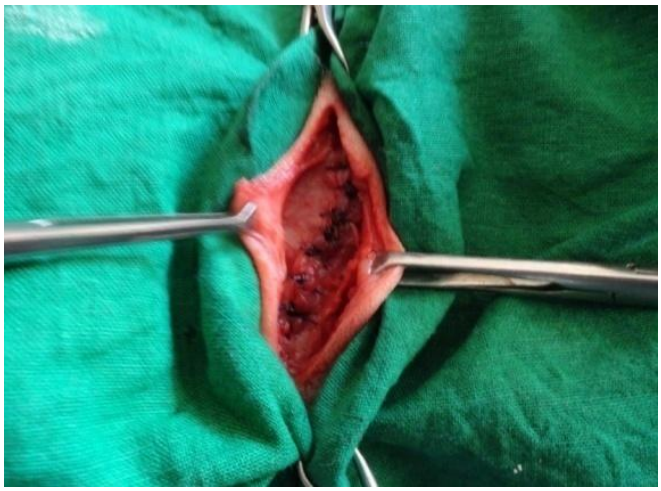


Plate 15: Laparotomy wound closure with polydioxanone

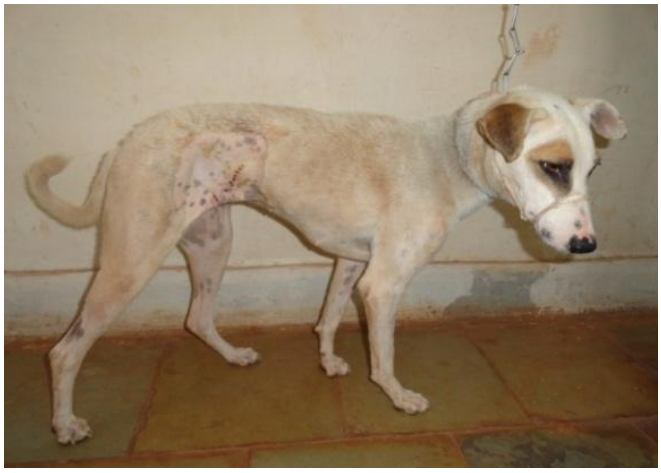


Plate 16: 14th post-operative day

3.4.2 Haematological parameter

Using disposable syringes with aseptic measures two ml of blood sample was drawn from cephalic vein and collected in EDTA vials for estimation of haemogram and leukogram. Blood sample were collected daily from 0 hours and 24 hours, 48 hours, 72 hours and 14^{th} days after the surgery.

3.4.2.1 Total Erythrocyte Count (millions cells / micro litre)

Total erythrocyte count was estimated as per the procedure described by Schalm *et al.* (1975) using Neubar's slide and the values were expressed as million cells per micro litre of blood ($10^{6/}$µl).

3.4.2.2 Haemoglobin (g/dl)

Haemoglobin was estimated by Shali's haemoglobinometer as per the standard method recommended by Schalm *et al.* (1975) and the values obtained were expressed as g/dl.

3.4.2.3 Packed Cell Volume (%)

Packed cell volume was estimated by haematocrit method and the values obtained were expressed as percentage.

3.4.2.4 Total Leucocyte Count (thousands cells / micro litre)

Total leucocyte count was estimated as per the procedure described by Jain (2000) and the values were expressed as thousand cells per micro litre of blood (10^{3}/µl).

3.4.2.5 Differential Leucocyte Count

Blood smears for differential leucocyte count were stained with Giemsa stain and cells were counted using Battlement method as described by Jain (2000) and the individual cells were expressed in percentage.

3.4.3 Biochemical parameters

Blood samples were collected and serum was separated before the surgery and 14^{th} days after surgery for estimation of aspertate aminotransferase (IU/L), lactate dehydrogenase (IU/L) and Creatine Phospho Kinase (IU/L). These parameter were estimated by using ARTOS biochemical analyzer (M/S Swemed diagnostics, Bengaluru) using respective diagnostic kit as per manufacture's instruction.

3.5 Clinical evaluation

Evaluation of skin wound for oedema, infection, dehiscences and late complications of muscular tissue infection were evaluated.

3.6 Histopathological evaluation

Obliques abdominus externus muscle biopsy was collected before suturing (plate 17) and on 14^{th} day after suturing in group I (plate 18), group II (plate 19), group III (plate 20) and group IV animal (plate 21). The collected material was fixed in 10% neutral buffered formalin and tissues were processed by routine paraffin embedding technique. 6-8 micron thick sections were cut and stained by Haematoxilin and Eosin for histopathology study and Mallory's phosphotungstic acid haematoxylin method (PTAH) for collagen and muscle fiber (Luna, 1968).

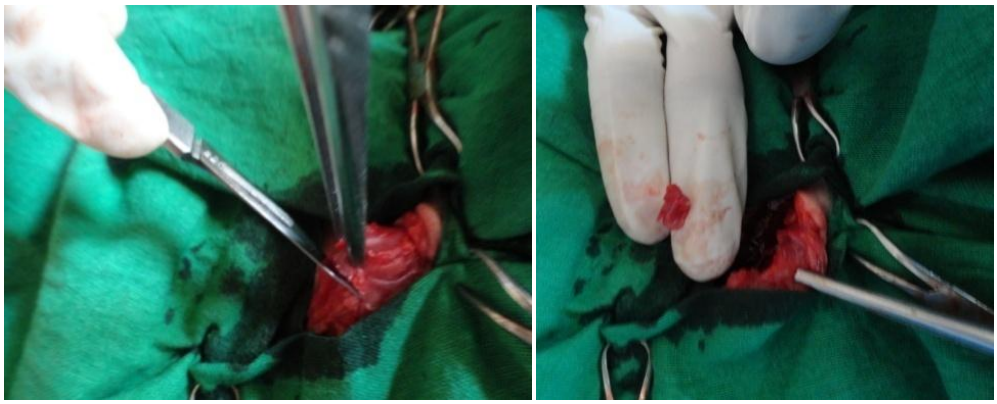


Plate 17: Biopsy collection on Zero day

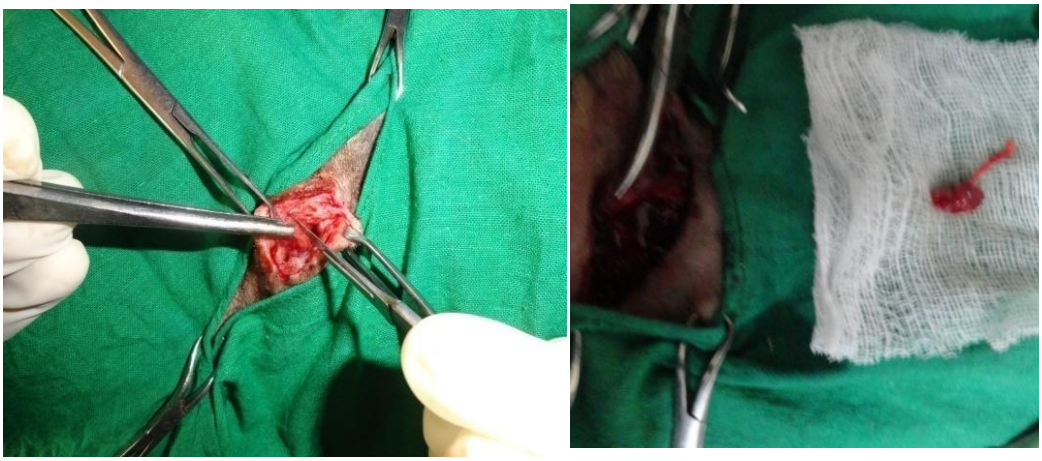


Plate 18: Biopsy collection on 14th day in group I animal

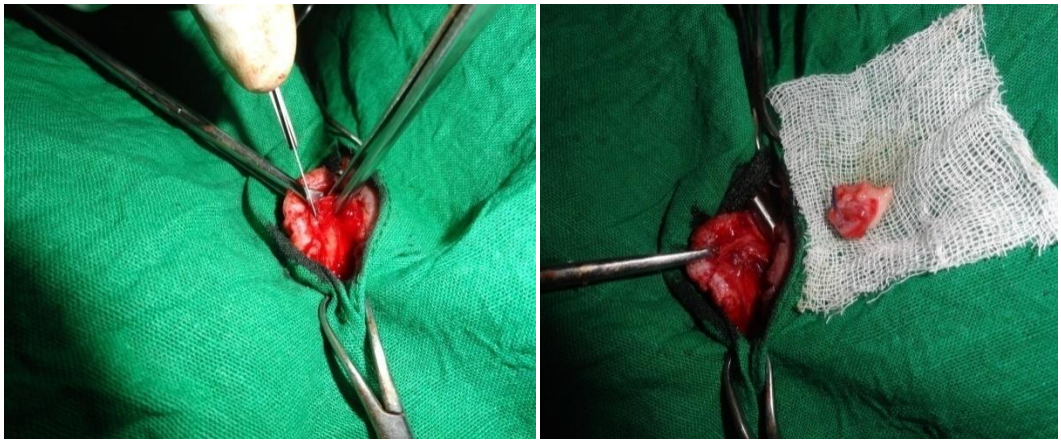


Plate 19: Biopsy collection on 14th day in group II animal

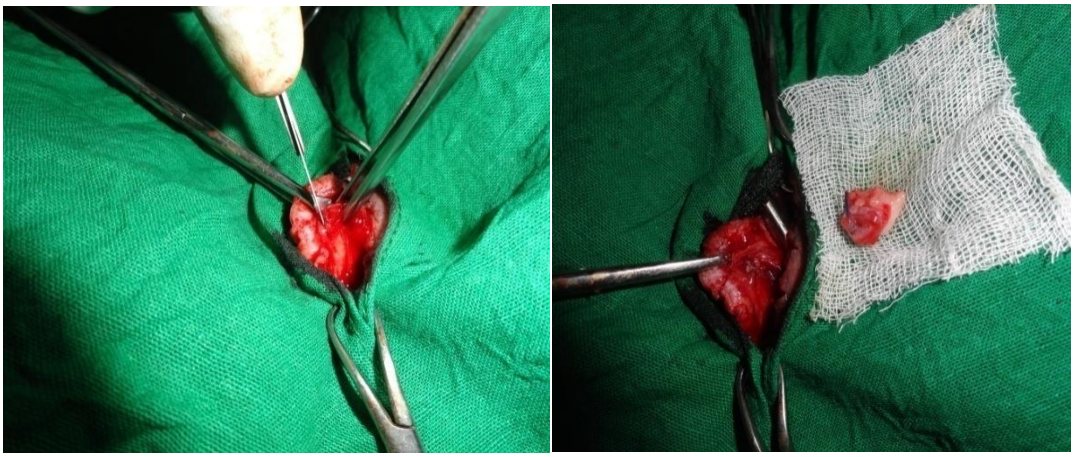


Plate 20: Biopsy collection on 14th day in group III animal

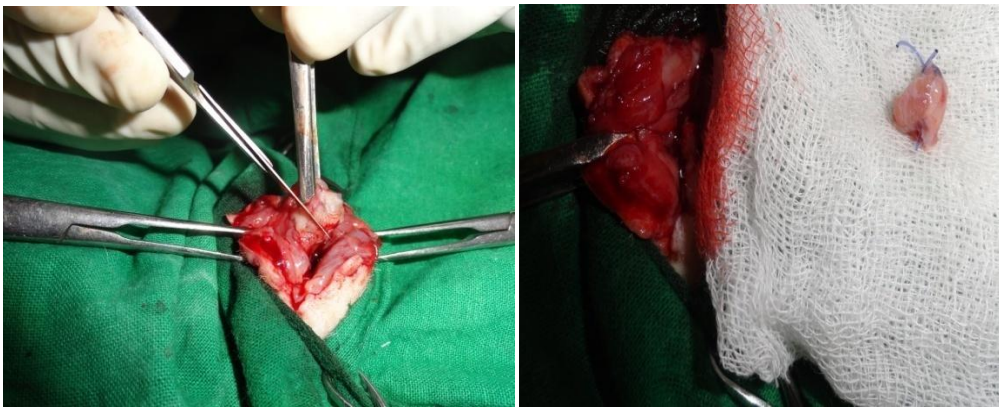


Plate 21: Biopsy collection on 14th day in group IV animal

3.7 Electron Microscopic Evaluation

Electron microscopic study purpose the biopsy was collected in 2.5% glutaraldehyde.

3.8 Statistical analysis

The data obtained was tabulated and was subjected to statistical analysis, using student ‘t’ test as per standard procedure described by Snedecor and Cochran (1989).

Results

IV. RESULTS

4.1 Parameters

4.1.1 Physiological parameters

The physiological parameters like respiratory rate, heart rate and rectal temperature were recorded at 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively in animals of groups I, group II, group III and group IV.

4.1.1.1 Respiratory Rate (breaths/minute)

The mean ± SE., values of respiratory rate (breaths/minute) are given in table (2) and fig (1).

The mean ± SE., values of respiratory rate (breaths/minute) of group I animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 28.16±1.07, 29.33 ± 0.42, 29.67±0.42, 28.67±1.14 and 27.00 ±1.06 respectively.

The mean ± SE., values of respiratory rate (breaths/minute) of group II animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 22.50±1.68, 25.83 ± 1.19, 24.16±1.97, 29.00 ±1.29 and 27.67±0.84 respectively.

The mean ± SE., values of respiratory rate (breaths/minute) of group III animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 26.67±1.25, 28.33± 0.88, 28.50±1.40, 28.16±1.19 and 27.67±0.75 respectively.

The mean ± SE., values of respiratory rate (breaths/minute) of group IV animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 25.16±1.40, 28.83± 0.79, 29.00±1.15, 28.16±1.92 and 28.67±1.20 respectively.

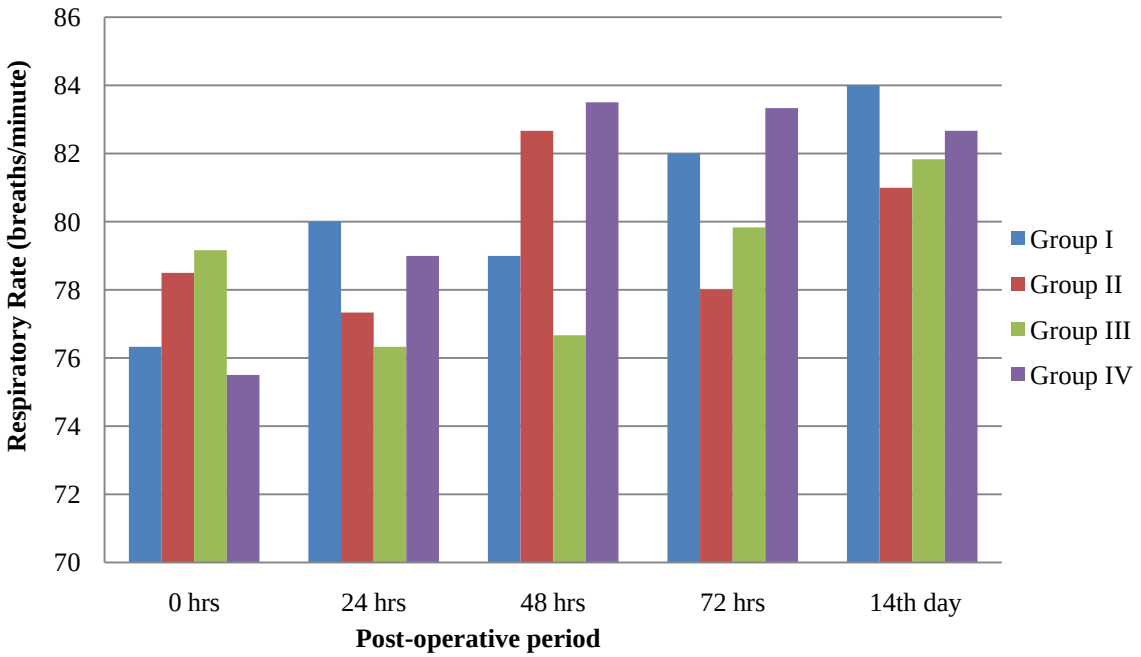
Table 2: Mean ± SE., values of Respiratory Rate (breaths/minute) in different groups of animals at different intervals

Groups	0 hours	24 hours	48 hours	72 hours	14th day
I	28.16 ±1.07[a]	29.33±0.42 [a]	29.67±0.42 [a]	28.67±1.14 [a]	27.00±1.06 [a]
II	22.50 ±1.68 [b]	25.83 ±1.19 [b]	24.16 ±1.97 [b]	29.00 ±1.29* [a]	27.67±0.84* [a]
III	26.67 ±1.25 [a]	28.33±0.88 [a]	28.50±1.40 [a]	28.16 ±1.19 [a]	27.67±0.75 [a]
IV	25.16±1.40 [a]	28.83±0.79* [a]	29.00 ±1.15 [a]	28.16±1.92 [a]	28.67±1.20 [a]

*= Means bearing superscript * differs significantly (p≤0.05) from 0 hours within the group.

a, b = Means bearing superscript a, b differs significantly (p≤0.05) between groups I and II and groups III and IV at corresponding intervals.

Fig. 1: Respiratory Rate (breaths/minute) in different Groups of animals at different intervals



The comparison of respiratory rate within the groups from pre-operative period (0 hours) to different post-operative intervals (24 hours, 48 hours, 72 hours and 14th day) showed non significant (P≥0.05) difference in group I and group III. However, the animals of group II and group IV showed significantly (p≤0.05) higher respiratory rate at 72 hours and 14th day. Whereas, group IV animals showed significantly (p≤0.05) higher respiratory rate at 24 hours.

The comparison between the group showed that the respiratory rate between group I, group III and group IV did not vary significantly (P>0.05) at all the intervals of study. However, the animals of group II at pre-operative period and at 24 hours and 48 hours post-operative period had lower respiratory rate when compared to group I, group III and group IV.

4.1.1.2 Heart Rate (beats/minute)

The mean±SE., values of heart rate (beats/minute) are given in table (3) and fig (2).

The mean ± SE., values of heart rate (beats/minute) of group I animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 76.33±2.28, 80.00±2.13, 79.00±2.33, 82.00±2.71 and 84.00±2.15 respectively.

The mean ± SE., values of heart rate (beats/minute) of group II animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 78.50±2.86, 77.33±2.92, 82.67±1.62, 78.00±2.85 and 81.00±2.88 respectively.

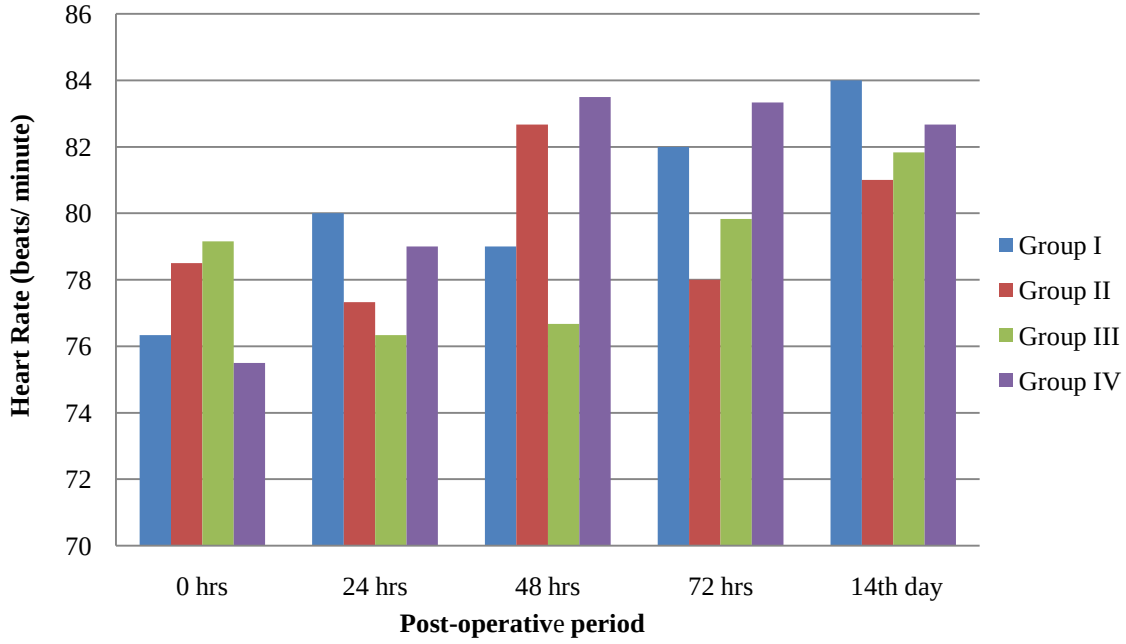
Table 3: Mean ± SE., values of Heart Rate (beats/minute) in different groups of animals at different intervals

Groups	0 hours	24 hours	48 hours	72 hours	14th day
I	76.33±2.28 [a]	80.00 ±2.13 [a]	79.00 ±2.33 [a]	82.00 ±2.71** [a]	84.00 ±2.15* [a]
II	78.50±2.86 [a]	77.33 ±2.92 [a]	82.67 ±1.62 [a]	78.00 ±2.85 [a]	81.00 ± 2.88 [a]
III	79.16±2.68 [a]	76.33 ±3.10 [a]	76.67 ±3.11 [a]	79.83 ±2.48 [a]	81.83 ±3.75 [a]
IV	75.50±2.55 [a]	79.00 ±2.51 [a]	83.50 ±1.76* [a]	83.33 ±2.01* [a]	82.67 ±1.99 * [a]

*= Means bearing superscript * differs significantly ($p \leq 0.05$) from 0 hours within the group.

**= Means bearing superscript **differs significantly ($p \leq 0.01$) from 0 hours within the group.

Fig. 2: Heart Rate (beats/minute) in different groups of animals at different intervals



The mean ± SE., values of heart rate (beats/minute) of group III animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 79.16±2.68, 76.33±3.10, 76.67±3.11, 79.83±2.48 and 81.83±3.75 respectively.

The mean ± SE., values of heart rate (beats/minute) of group IV animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 75.50±2.55, 79.00±2.51, 83.50±1.76, 83.33±2.01 and 82.67±1.99 respectively.

The comparison of heart rate within the group from pre-operative (0 hours) level to different intervals (24 hours, 48 hours, 72 hours and 14th day) showed that, there were significantly higher heart rates at 72 ($p\leq0.01$) hours and on 14th day ($p\leq0.05$) in animals of group I and group IV. In addition animals of group IV has significantly ($p\leq0.05$) higher heart rate in compare to pre-operative level. The animals of group II and group III did not show any significant ($p\leq0.05$) variation between the pre-operative and post-operative period.

The comparison of heart rate between the groups at all the intervals of study showed non significant ($P\geq0.05$) difference in the heart rate. The heart rate in all the groups of animal remained within normal physiological limit.

4.1.1.3 Rectal Temperature (°F)

The mean ± SE., values of rectal temperature (°F) are given in table (4) and fig (3).

The mean ± SE., values of rectal temperature (°F) of group I animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 100.83±0.24, 101.50±0.21, 102.30±0.25, 101.78±0.27 and 101.28±0.24 respectively.

Table 4: Mean ± SE., Values of Rectal Temperature (°F) in different groups of animals at different intervals.

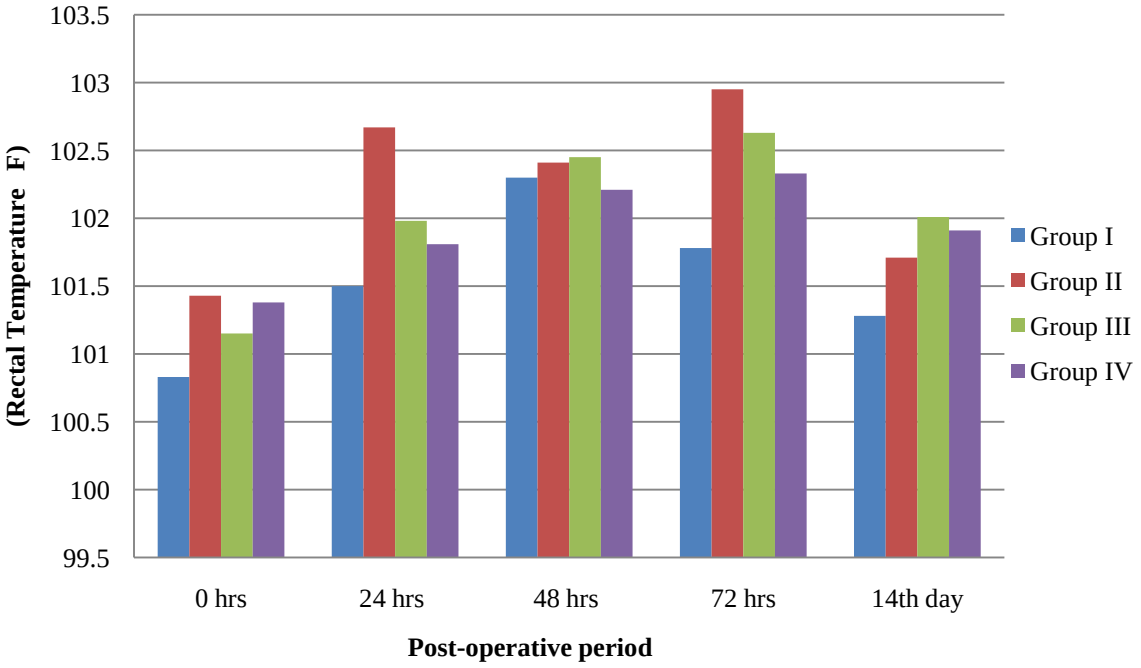
Groups	0 hours	24 hours	48 hours	72 hours	14th day
I	100.83 ± 0.24 [a]	101.50 ±0.21 [a]	102.30 ±0.25** [a]	101.78 ±0.27* [a]	101.28 ±0.24 [a]
II	101.43 ±0.18 [a]	102.67 ±0.22** [b]	102.41 ±0.21** [a]	102.95 ±0.25** [b]	101.71 ±0.15 [a]
III	101.15 ±0.21 [a]	101.98 ±0.17* [a]	102.45 ±0.09** [a]	102.63 ±0.22** [b]	102.01 ±0.07** [b]
IV	101.38 ±0.25 [a]	101.81 ±0.23 [a]	102.21 ±0.20* [a]	102.33 ±0.16* [a]	101.91 ±0.10 [b]

*= Means bearing superscript * differs significantly ($p \leq 0.05$) from 0 hours within the group.

**= Means bearing superscript **differs significantly ($p \leq 0.01$) from 0 hours within the group.

a, b = Means bearing superscript a, b differs significantly ($p \leq 0.05$) between groups I and II and groups III and IV at corresponding intervals.

Fig. 3: Rectal Temperature (° F) in different groups of animals at different intervals



The mean ± SE., values of rectal temperature (°F) of group II animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 101.43±0.18, 102.67±0.22, 102.41±0.21, 102.95±0.25 and 101.71±0.15 respectively.

The mean ± SE., values of rectal temperature (°F) of group III animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 101.15±0.21, 101.98±0.17, 102.45±0.09, 102.63±0.22 and 102.01±0.07 respectively.

The mean ± SE., values of rectal temperature (°F) of group IV animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 101.38±0.25, 101.81±0.23, 102.21±0.20, 102.33±0.16 and 101.91±0.10 respectively.

The rectal temperature was significantly ($p \leq 0.05$) higher at 48 hours and 72 hours post-operatively when compared to pre-operative level in all the groups of animals, in addition group II animals showed significantly ($p \leq 0.05$) higher rectal temperature at 24 hours when compared to 0 hours. Whereas, in group III animals significantly higher rectal temperature was observed at 24 hours ($p \leq 0.01$) and 14th post-operative day ($p \leq 0.05$) when compared to pre-operative period.

The comparison between the group revealed that at 24 hours group II animals had significantly ($p \leq 0.05$) higher rectal temperature when compared to other groups at 48 hours intervals there was non significant ($P \geq 0.05$) difference in rectal temperature between any of the group under present study.

At 72 hours group II and group III animals had significantly ($p \leq 0.05$) higher rectal temperature when compared to other groups of animals. On 14th post-operative day

group III and group IV animals had significantly ($p \leq 0.05$) higher rectal temperature when compared to other groups. The overall rectal temperature in all the groups of animals fluctuated within the normal physiological limits.

4.1.2 Haematological parameters

The haematological studies were carried out by estimation of total erythrocyte count, haemoglobin, packed cell volume, total leukocyte count and differential leukocyte count by collecting blood at 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively in animals of groups I, group II, group III and group IV.

4.1.2.1 Total Erythrocyte Count (million cells /micro litre)

The mean ± SE., values of total erythrocyte count (million cells/micro litre) are given in table (5) and fig (4).

The mean ± SE., values of total erythrocyte count (million cells/micro litre) of group I animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 5.79 ± 0.38, 5.22±0.44, 5.09±0.39, 5.27±0.35 and 6.17±0.52 respectively.

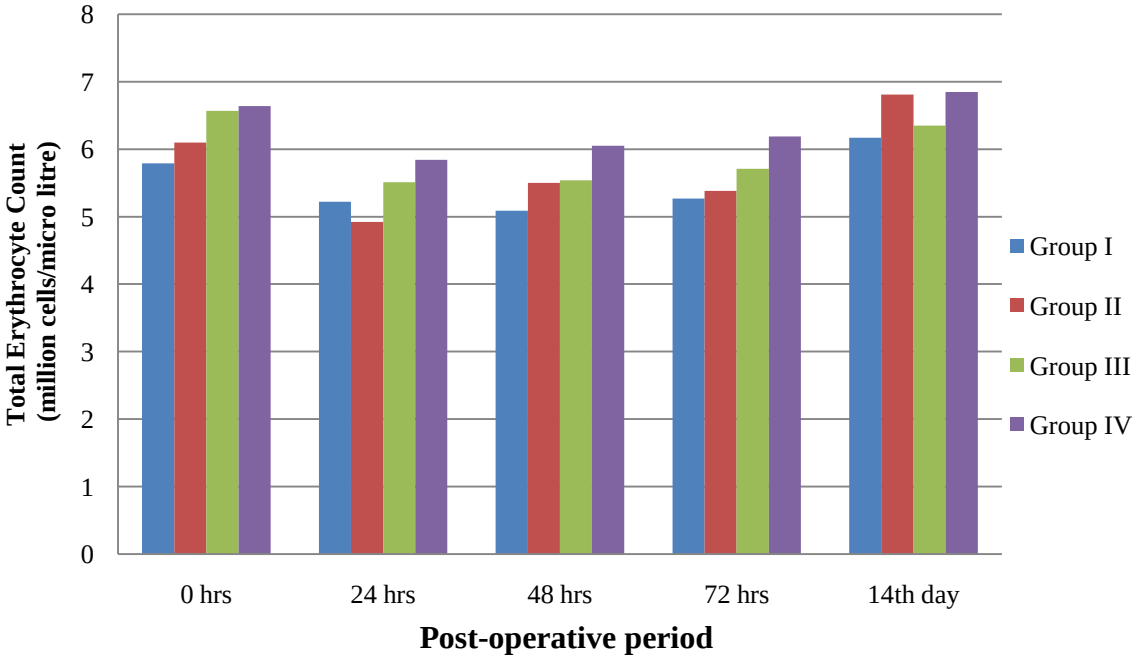
The mean ± SE., values of total erythrocyte count (million cells/micro litre) of group II animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 6.10±0.65, 4.92±0.43, 5.50±0.43, 5.38±0.44 and 6.81±0.61 respectively.

The mean ± SE., values of total erythrocyte count (million cells/micro litre) of group III animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 6.57±0.51, 5.51±0.34, 5.54±0.25, 5.71±0.33 and 6.35±0.55 respectively.

Table 5: Mean ± SE., values of Total Erythrocyte Count (million cells/micro litre) in different groups of animals at different intervals

Groups	0 hours	24 hours	48 hours	72 hours	14th day
I	5.79 ±0.38 [a]	5.22 ±0.44 [a]	5.09 ±0.39 [a]	5.27 ±0.35 [a]	6.17 ±0.52 [a]
II	6.10 ±0.65 [a]	4.92 ±0.43 [a]	5.50 ±0.43 [a]	5.38 ±0.44 [a]	6.81 ±0.61 [a]
III	6.57 ±0.51 [a]	5.51 ±0.34 [a]	5.54 ±0.25 [a]	5.71 ±0.33 [a]	6.35 ±0.55 [a]
IV	6.64 ±0.34 [a]	5.84 ±0.30 [a]	6.05 ±0.31 [a]	6.19 ±0.38 [a]	6.85 ±0.40 [a]

Fig. 4: Total Erythrocyte Count (million cells/micro litre) in different groups of animals at different intervals



The mean ± SE., values of total erythrocyte count (million cells/micro litre) of group IV animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 6.64±0.34, 5.84±0.30, 6.05±0.31, 6.19±0.38 and 6.85±0.40 respectively.

The comparison within the group from pre-operative period level to 24 hours to 14th day showed non significant (P≥0.05) difference in all the group of animal.

The comparison between the groups at different intervals of present study revealed that there was non significant (P≥0.05) difference in the total erythrocyte count at all the intervals of the present study. The total erythrocyte count in all the groups of animals and at different intervals (0 hours, 24 hours, 48 hours, 72 hours and14th day) of study fluctuated within normal physiological limits.

4.1.2.2 Haemoglobin (g/dl)

The mean ± SE., values of haemoglobin (g/dl) are given in table (6) and fig (5).

The mean ± SE., values of haemoglobin (g/dl) of group I animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 13.68±1.08, 12.31±1.26, 12.15±1.30, 12.73±1.31 and 13.59±1.40 respectively.

The mean ± SE., values of haemoglobin (g/dl) of group II animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 12.46±0.81, 9.90±0.69, 11.05±0.65, 10.90±0.76 and 12.15±0.62 respectively.

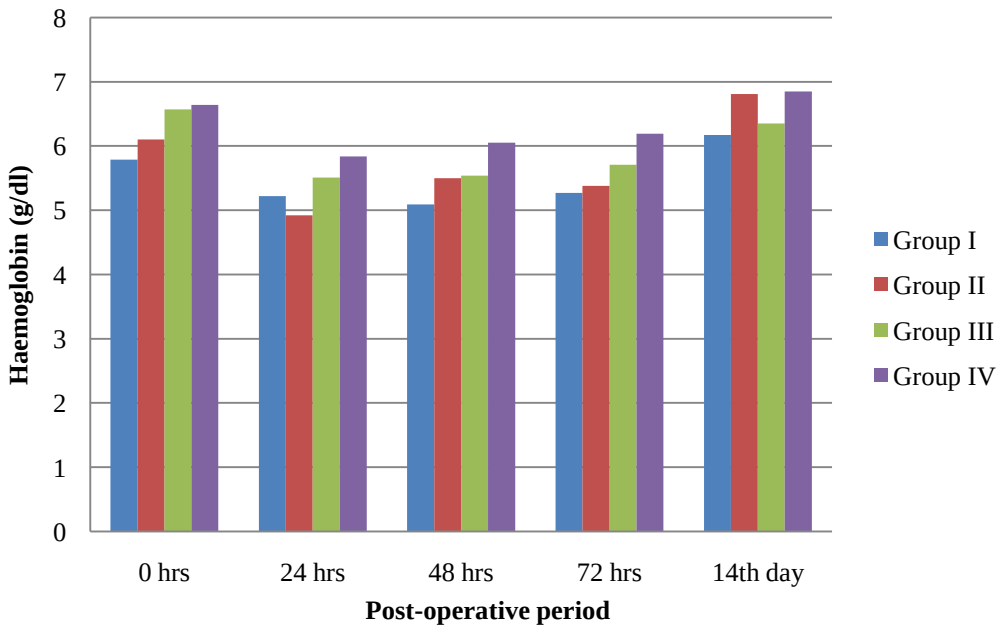
The mean ± SE., values of haemoglobin (g/dl) of group III animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 12.86±1.03,11.58±0.84 11.48±0.62, 11.61±0.67 and 12.63±0.99 respectively.

Table 6: Mean ± SE., values of Haemoglobin (g/dl) in different groups of animals at different intervals

Groups	0 hours	24 hours	48 hours	72 hours	14th day
I	13.68±1.08 [a]	12.31 ±1.26 [a]	12.15 ±1.30 [a]	12.73 ±1.31 [a]	13.59 ±1.40 [a]
II	12.46±0.81 [a]	9.90 ±0.69* [a]	11.05±0.65	10.90 ±0.76 [a]	12.15 ±0.62 [a]
III	12.86 ±1.03 [a]	11.58 ±0.84 [a]	11.48 ±0.62 [a]	11.61 ±0.67 [a]	12.63 ±0.99 [a]
IV	12.36 ±0.67 [a]	11.65 ±0.72 [a]	11.55 ±0.72 [a]	11.85 ±0.73 [a]	12.33 ±0.56 [a]

*= Means bearing superscript * differs significantly ($p \leq 0.05$) from 0 hours within the group.

Fig 5: Haemoglobin (g/dl) in different groups of animals at different intervals



The mean ± SE., values of haemoglobin (g/dl) of group IV animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 12.36±0.67, 11.65±0.72, 11.55±0.72,11.85±0.73 and 12.33±0.56 respectively.

The comparison of haemoglobin level within the group in general showed that, there was decreased haemoglobin level at all the post-operative (24 hour, 48hours, 72hours and 14th day) interval of study when compared to pre-operative level (0 hours).The decreased in haemoglobin level was statistically significant ($p \leq 0.05$) at 24 hours when compared to 0 hours in group II animal.

The comparison between the groups at different intervals (0 hours, 24 hours, 48 hours, 72 hours and 14th day) showed that there was non significant ($P \geq 0.05$) difference in the level of haemoglobin suggesting that haemoglobin level fluctuated within normal physiological limits in all the groups of animals.

4.1.2.3 Packed Cell Volume (%)

The mean ± SE., values of packed cell volume (%) are given in table (7) and fig (6).

The mean ± SE., values of packed cell volume (%) of group I animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 41.75±1.36, 37.65±1.77, 37.35±1.76, 38.01±1.45 and 40.46±1.59 respectively.

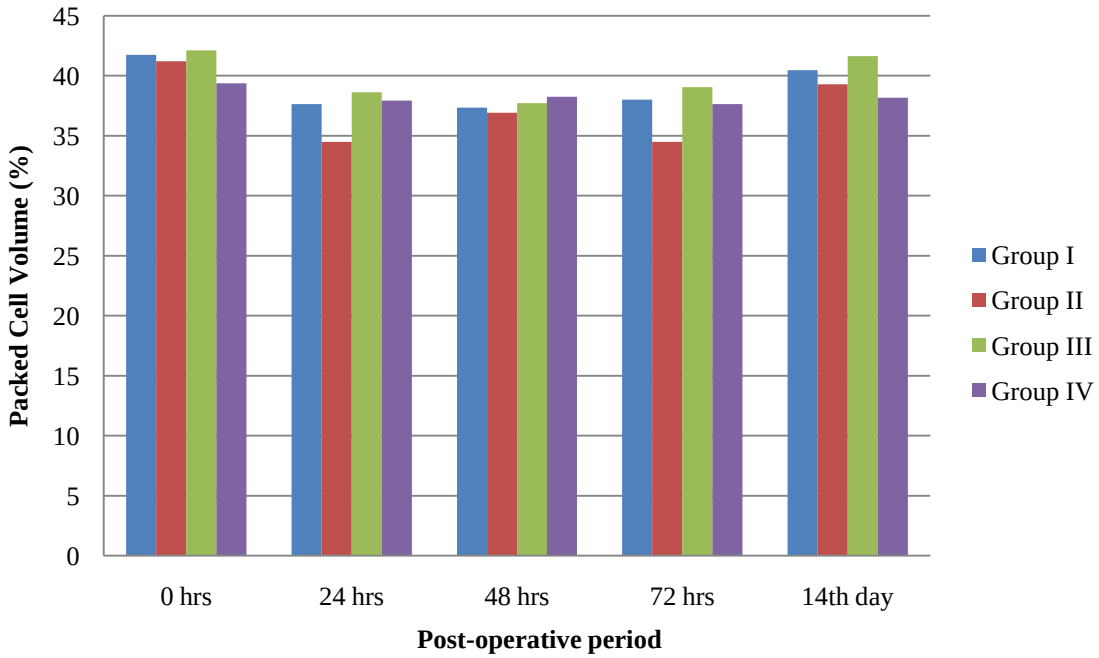
The mean ± SE., values of packed cell volume (%) of group II animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 41.20±3.28, 34.48±0.31, 36.91±2.81, 34.50±3.08 and 39.30±2.02 respectively.

Table 7: Mean ± SE., values of Packed cell volume (%) in different groups of animals at different intervals

Groups	0 hours	24 hours	48 hours	72 hours	14th day
I	41.75±1.36 [a]	37.65 ±1.77 [a]	37.35±1.76 [a]	38.01±1.45 [a]	40.46 ±1.59 [a]
II	41.20 ±3.28 [a]	34.48 ±0.31 * [a]	36.91 ±2.81 [a]	34.50±3.08 [a]	39.30 ±2.02 [a]
III	42.13 ±3.46 [a]	38.63 ±2.32 [a]	37.73 ±1.11 [a]	39.05 ±1.96 [a]	41.63 ±2.44 [a]
IV	39.38 ±1.16 [a]	37.93 ±0.80 [a]	38.26 ±0.74 [a]	37.63 ±0.56 [a]	38.18 ±0.74 [a]

*= Means bearing superscript * differs significantly ($p \leq 0.05$) from 0 hours within the group.

Fig. 6: Packed cell volume (%) in different groups of animals at different intervals



The mean ± SE., values of packed cell volume (%) of group III animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 42.13±3.46, 38.63±2.32, 37.73±1.11, 39.05±1.96 and 41.63±2.44 respectively.

The mean ± SE., values of packed cell volume (%) of group IV animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 39.83±1.16, 37.93±0.80, 38.26±0.74, 37.63±0.56 and 38.18 ±0.74 respectively.

The comparison of packed cell volume level within the group in general showed that, there was decreased packed cell volume level at all the post-operative (24 hour, 48hours, 72 hours and 14th day) interval of study when compared to pre-operative level. The decreased in packed cell volume level was statistically significant ($p \leq 0.05$) at 24 hours when compared to 0 hours in group II animal.

The comparison between the groups at different intervals (0 hours, 24 hours, 48 hours, 72 hours and 14th day) showed that there was non significant ($P \geq 0.05$) difference in the level of packed cell volume suggesting that packed cell volume level fluctuated within normal physiological limits in all the groups of animals.

4.1.2.4 Total Leukocyte Count (thousands cells/ micro litre)

The mean ± SE., values of total leukocyte count (thousands cells/ micro litre) are given in table (8) and fig (7).

The mean ± SE., values of total leukocyte count (thousands cells/micro litre) of group I animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 14.76±2.67, 26.38±2.81, 26.15±4.85, 21.95±3.08 and 13.48±1.97 respectively.

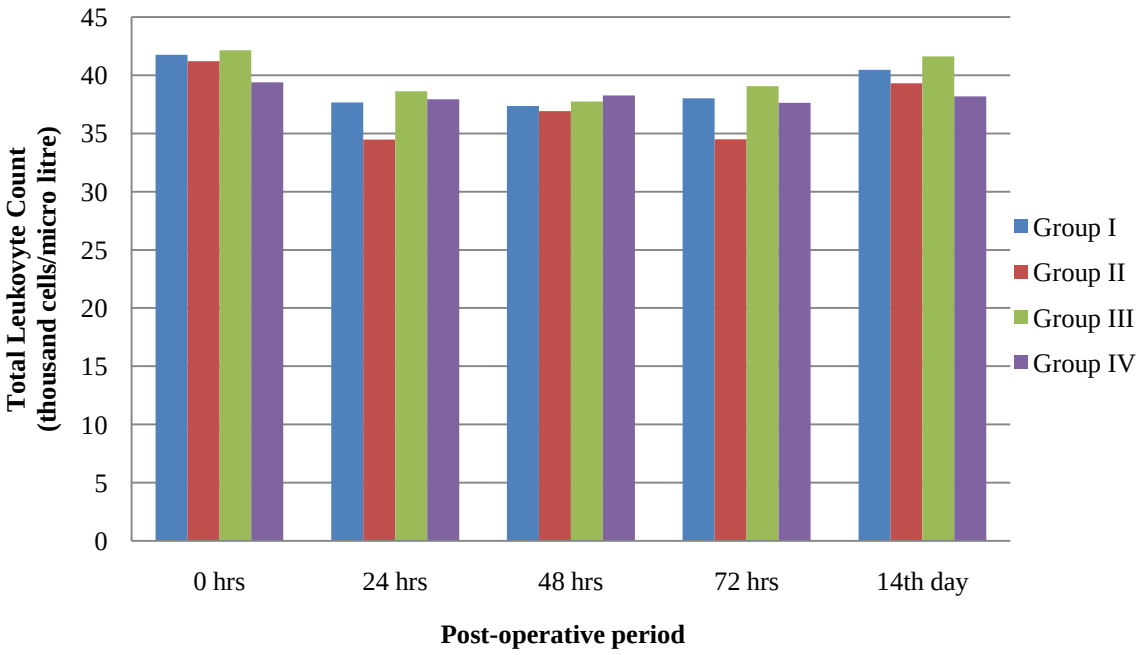
Table 8: Mean ± SE., values of Total Leukocyte Count (thousand cells/micro litre) in different groups of animals at different intervals

Groups	0 hours	24 hours	48 hours	72 hours	14th day
I	14.76 ±2.67 [a]	26.38 ±2.81* [a]	26.15 ±4.85 [a]	21.95 ±3.08 [a]	13.48 ±1.97 [a]
II	18.90 ±3.16 [a]	32.10 ±3.40* [a]	26.43 ±3.68 [a]	22.25 ±3.22 [a]	16.52 ±2.40 [a]
III	14.76 ±1.14 [a]	36.40 ±3.94** [a]	28.71 ±3.82** [a]	26.35 ±3.23** [a]	14.55 ±1.13 [a]
IV	10.61 ±0.89 [a]	33.00±2.47* * [a]	28.31 ±1.94** [a]	21.67±2.19** [a]	10.96 ±0.79 [a]

*= Means bearing superscript * differs significantly ($p \leq 0.05$) from 0 hours within the group.

** = Means bearing superscript **differs significantly ($p \leq 0.01$) from 0 hours within the group.

Fig. 7: Total Leukocyte Count (thousand cells/micro litre) in different groups of animals at different intervals



The mean ± SE., values of total leukocyte count (thousands cells/micro litre) of group II animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 18.90±3.16, 32.10±3.40, 26.43±3.68, 22.25±3.22 and 16.52±2.40 respectively.

The mean ± SE., values of total leukocyte count (thousands cells/micro litre) of group III animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 14.76±1.14, 36.40±3.94, 28.71±3.82, 26.35±3.23 and 14.55±1.13 respectively.

The mean ± SE., values of total leukocyte count (thousands cells/micro litre) of group IV animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 10.61±0.89, 33.00±2.47, 28.31±1.94, 21.67±2.19 and 10.96±0.79 respectively.

The comparison of total leukocyte count between the group pre-operative and post-operative intervals showed that, there was leukocytosis between 24 hours to 72 hours and the leukocyte count reached near normal by 14th day in all the groups of animal.

The leukocytosis was statistically significant ($p \leq 0.05$) at 24 hours when compared to 0 hours in group I and group II animals. Whereas, group III and group IV animal showed that, there was severe leukocytosis ($p \leq 0.01$) between 24 to 72 hours when compared to pre-operative level.

The comparison between the groups at different intervals of present study revealed that there was no significant ($P \geq 0.05$) difference in the total leukocyte count at all the intervals of the present study. The total leukocyte count in all the groups of

animals and at different intervals (0 hours, 24 hours, 48 hours, 72 hours and 14th day) of study fluctuated within normal physiological limits.

4.1.2.5 Neutrophils (%)

The mean ± SE., values of neutrophils (%) are given in table (9) and fig (8).

The mean ± SE., values of neutrophils (%) of group I animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 65.33±1.38, 76.00±0.85, 76.40±1.27, 74.50±5.45 and 66.67±3.61 respectively.

The mean ± SE., values of neutrophils (%) of group II animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 63.43±2.34, 65.38±1.74, 69.43±2.53, 71.36±2.58 and 64.23±1.78 respectively.

The mean ± SE., values of neutrophils (%) of group III animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 64.67±4.98, 67.33±0.58, 71.83±0.51, 77.67±2.60 and 69.50±4.95 respectively.

The mean ± SE., values of neutrophils (%) of group IV animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 67.83±1.57, 74.50±2.02, 81.00±1.88, 84.00±1.03, 72.00±1.52 respectively.

The comparison of neutrophil level within the group between 0 hours and at different post-operative (24 hours, 48 hours, 72 hours and 14th day) interval revealed that, in general there was neutrophilia between 24 to 72 hours. The neutrophil level by 14th day has reached normal in all the groups of animal.

Table 9: Mean ± SE., values of Neutrophils (%) in different groups of animals at different intervals

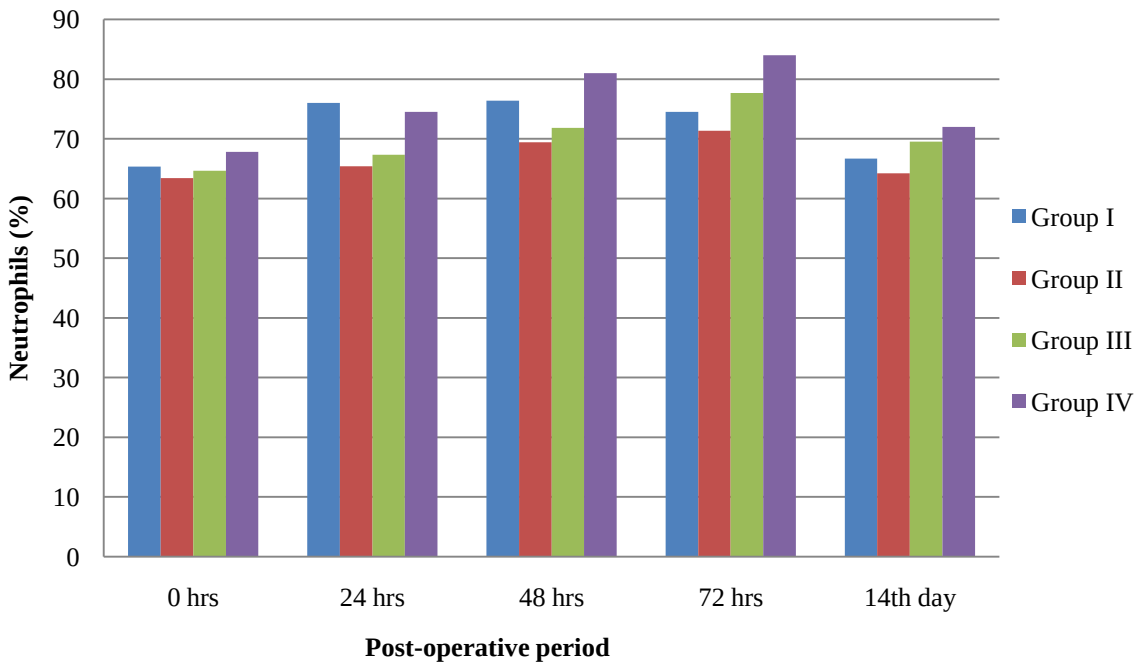
Groups	0 hours	24 hours	48 hours	72 hours	14th day
I	65.33 ±1.38 [a]	76.00±0.85** [a]	76.40 ±1.27** [a]	74.50 ±5.45 [b]	66.67 ±3.61 [b]
II	63.43 ±2.34 [b]	65.38 ±1.74* [b]	69.43 ±2.53** [b]	71.36 ±2.58** [a]	64.23 ±1.78 [a]
III	64.67 ±4.98 [a]	67.33 ±0.58* [c]	71.83 ±0.51** [a]	77.67 ±2.60* [b]	69.50 ±4.95 [c]
IV	67.83 ±1.57 [a]	74.50 ±2.02* [a]	81.00 ±1.88** [c]	84.00 ±1.03** [c]	72.00 ±1.52 [b]

*= Means bearing superscript * differs significantly (p≤0.05) from 0 hours within the group.

**= Means bearing superscript **differs significantly (p≤0.01) from 0 hours within the group

Means bearing different superscript, differs significantly (p≤0.05) between the group at different intervals.

Fig. 8: Neutrophils (%) in different groups of animals at different intervals



The comparison between group showed that, at 0 hours group II animals had lower neutrophil count compared to other group, at 24 hours the group II animal had neutrophil count lesser than group III animal.

Group III animal had lesser neutrophil count when compared to group I and group IV animals, at 48 hours interval highest neutrophilia was observed in group IV animal followed by group I, group III and group II animal respectively.

Group II had least neutrophil count when compared to all other group. At 72 hours highest neutrophilia was observed in group IV animals followed by group III and group I animal respectively. The least neutrophilia was observed in group II animal when compared to all other group. The similar trend was observed on 14^{th} day. The animals of group II had significantly ($p \leq 0.05$) lower neutrophil count when compared to all others groups. However, the neutrophil counts on 14^{th} day fluctuated within normal physiological limit.

4.1.2.6 Lymphocyte (%)

The mean ± SE., lymphocyte (%) values are given in table (10) and fig (9).

The mean ± SE., values of lymphocyte (%) of group I animals on 0 hours, 24 hours, 48 hours, 72 hours and 14^{th} day post-operatively were: 27.60±1.47, 17.83±0.87, 17.16±0.94, 21.00±3.60 and 29.16±3.97 respectively.

The mean ± SE., values of lymphocyte (%) of group II animals on 0 hours, 24 hours, 48 hours, 72 hours and 14^{th} day post-operatively were: 26.57±2.31, 25.45±3.21, 20.91±2.98, 16.31±2.33 and 29.28±3.13 respectively.

Table 10: Mean ± SE., values of Lymphocyte (%) in different groups of animals at different intervals

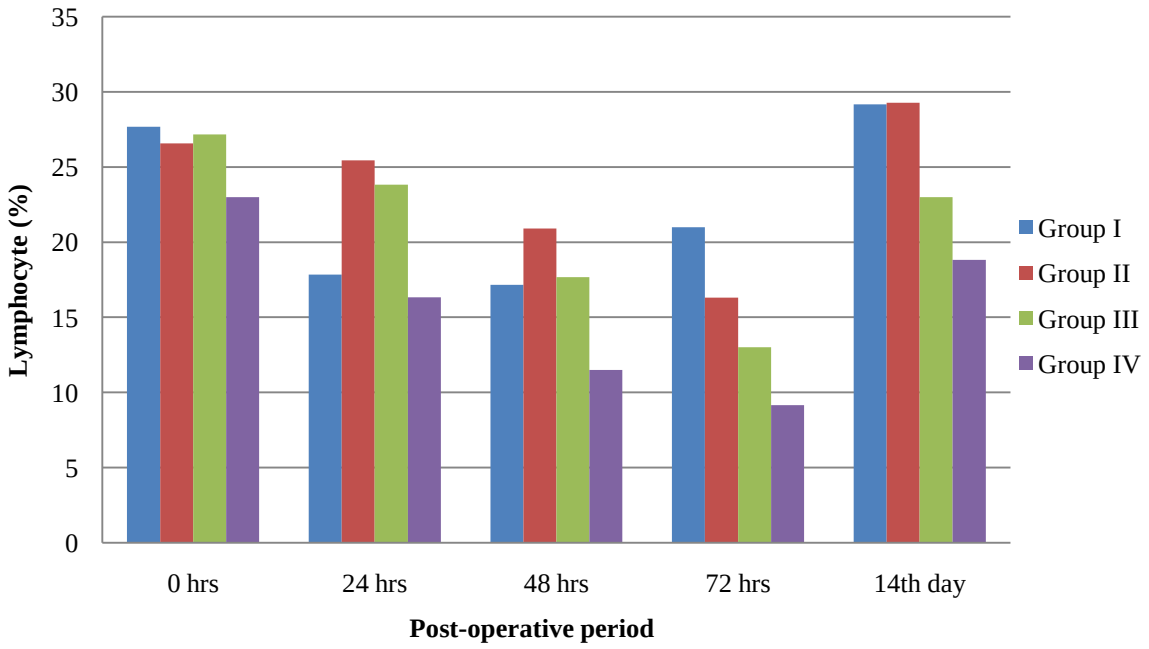
Groups	0 hours	24 hours	48 hours	72 hours	14th day
I	27.60 ±1.47 [a]	17.83 ±0.87** [a]	17.16±0.94** [b]	21.00 ±3.60 [c]	29.16 ±3.97 [b]
II	26.57 ±2.31 [a]	25.45 ±3.21 [b]	20.91±2.98** [c]	16.31 ±2.33** [c]	29.28 ±3.13** [b]
III	27.16 ±4.46 [a]	23.83 ±3.77 [a]	17.67 ±3.13** [b]	13.00 ±2.59* [b]	23.00 ±4.33* [b]
IV	23.00 ±1.34 [a]	16.33 ±1.81 [a]	11.50 ±1.82* [a]	9.16 ±1.22* [a]	18.83 ±2.18* [a]

*= Means bearing superscript * differs significantly (p≤0.05) from 0 hours within the group.

**= Means bearing superscript **differs significantly (p≤0.01) from 0 hours within the group.

Means bearing different superscript, differs significantly (p≤0.05) between the group at different intervals.

Fig. 9: Lymphocyte (%) in different groups of animals at different intervals



The mean ± SE., values of lymphocyte (%) of group III animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 27.16±4.46, 23.83±3.77, 17.67±3.13, 13.00±2.59 and 23.00±4.33 respectively.

The mean ± SE., values of lymphocyte (%) of group IV animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 23.00±1.34, 16.33±1.81, 11.50±1.82, 9.16±1.22 and 18.83±2.18 respectively.

The comparison of lymphocyte count within the group between 0 hours and different post-operative interval there was relative lymphocytopenia between 24 to 72 hours, in all the groups of animal.

The relative lymphocytopenia revealed even on group III and group IV animal. In group I, the relative lymphocytopenia was statistically significant ($p \leq 0.05$) between 24 to 48 hours. Whereas, it was statistically significant ($p \leq 0.05$) between 48 to 72 hours in group II, group III and group IV animal.

The comparison between the group at 24 hours, the relative lymphocytopenia was severe in group IV animals followed by group I and group III animals when compared to group II animal. The similar trend was also observed at 48 hours, at 72 hours relative lymphocytopenia was observed in group IV animal when compared to group III animal.

The group III animal had relative lymphocytopenia when compared to group I and group II animal. On 14th day relative lymphocytopenia revealed only in group IV animal.

4.1.2.7 Eosinophils (%)

The mean ± SE., eosinophils (%) values are given in table (11).

The mean ± SE., values of eosinophils (%) of group I animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 2.33±0.42, 2.16±0.30, 2.33±0.42, 0.67±0.33 and 2.33±0.42 respectively.

The mean ± SE., values of eosinophils (%) of group II animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 3.00±0.57, 2.50±0.49, 3.33±0.67, 3.50±0.49 and 1.16±0.16 respectively.

The mean ± SE., values of eosinophils (%) of group III animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 3.30±0.42, 3.83±0.60, 4.67±0.42, 3.83±0.65 and 4.00±0.57 respectively.

The mean ± SE., values of eosinophils (%) of group III animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 4.83±0.60, 5.00±0.57, 4.16±0.83, 4.16±0.60 and 4.83±0.60 respectively. The eosinophil (%) in all the groups of animal remained within normal physiological limit.

4.1.2.8 Monocyte (%)

The mean ± SE., values of monocyte (%) are given in table (12).

The mean ± SE., values of monocyte (%) of group I animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 4.67±0.71, 4.00±0.73,4.00±0.73, 3.83±1.75 and 1.83±0.30 respectively.

Table 11: Mean ± SE., values of Eosinophils (%) in different groups of animals at different intervals

Groups	0 Hours	24 hours	48 hours	72 hours	14 th day
I	2.33±0.42[a]	2.16±0.30 [a]	2.33±0.42 [a]	0.67±0.33 [a]	2.33±0.42 [a]
II	3.00±0.57 [a]	2.50±0.49 [a]	3.33±0.67 [a]	3.50±0.49 [a]	1.16±0.16 [a]
III	3.30±0.42 [a]	3.83±0.60 [a]	4.67±0.42 [a]	3.83±0.65 [a]	4.00±0.57 [a]
IV	4.83±0.60 [a]	5.00±0.57 [a]	4.16±0.83 [a]	4.16±0.60 [a]	4.83±0.60 [a]

Table 12: Mean ± SE., values of Monocyte (%) in different groups of animals at different intervals

Groups	0 Hours	24 hours	48 hours	72 hours	14th day
I	4.67±0.71[a]	4.00±0.73 [a]	4.00±0.73 [a]	3.83±1.75 [a]	1.83±0.30 [a]
II	7.00±1.39 [a]	6.67±1.08 [a]	6.33±1.11 [a]	8.83±1.70 [a]	5.33±0.88 [a]
III	4.01±0.67 [a]	5.00±0.44 [a]	5.83±0.60 [a]	5.50±0.56 [a]	3.50±0.42 [a]
IV	4.33±0.67 [a]	4.16±0.60 [a]	3.33±0.42 [a]	2.67±0.49 [a]	4.33±0.71 [a]

The mean ± SE., values of monocyte (%) of group II animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 7.00±1.39, 6.67±1.08, 6.33±1.11, 8.83±1.70 and 5.33±0.88 respectively.

The mean ± SE., values of monocyte (%) of group IV animals on 0 hours, 24 hours, 48 hours, 72 hours and 14th day post-operatively were: 4.33±0.67, 4.16±0.60, 3.33±0.42, 2.67±0.49 and 4.33±0.71 respectively.

The monocyte (%) in all the groups of animal remained within normal physiological limit.

4.1.2 Biochemical parameters

4.1.2.1 Aspertate Aminotransferase (IU/L)

The mean ± SE., values of aspertate aminotransferase (IU/L) are given in table (13) and fig (10).

The mean ± SE., values of aspertate aminotransferase (IU/L) of group I animals on 0 hours and on 14th day post-operatively were: 28.46±3.21 and 99.68±19.53 respectively.

The mean ± SE., values of aspertate aminotransferase (IU/L) of group II animals on 0 hours and on 14th day post-operatively were: 30.70±3.81 and 59.16±10.47 respectively.

The mean ± SE., values of aspertate aminotransferase (IU/L) of group III animals on 0 hours and on 14th day post-operatively were: 42.08±6.76 and 84.53±5.60 respectively.

Table 13: Mean ± SE., Aspertate Aminotransferase (IU/L) in different groups of animals at different intervals

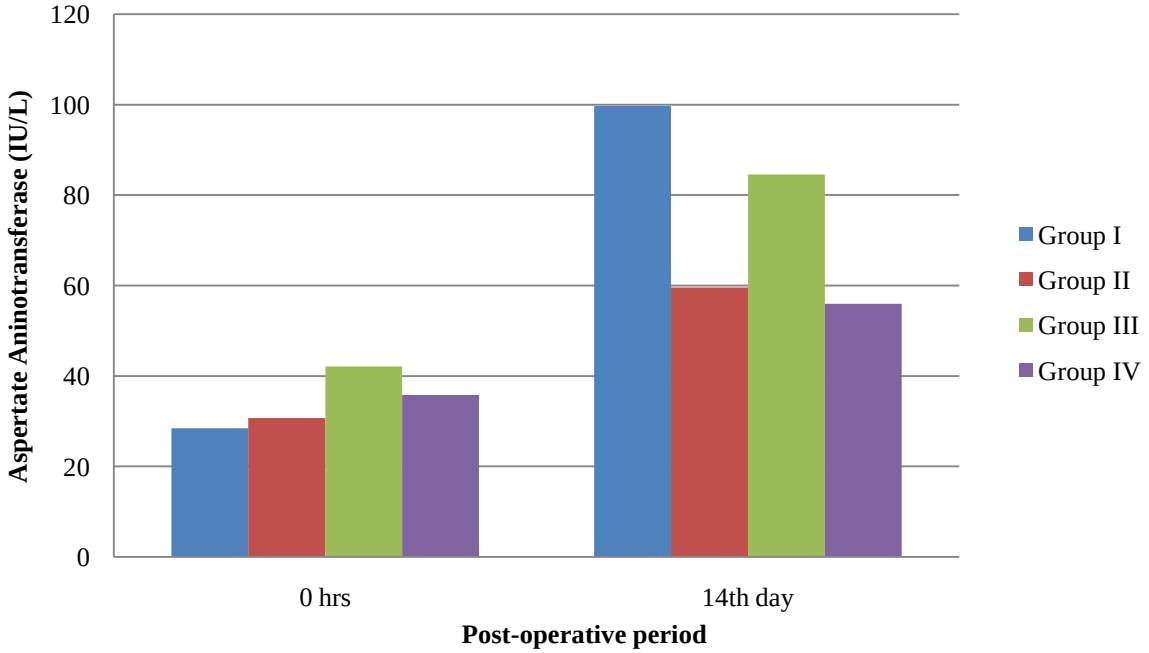
Groups	0 Hours	14th day
I	28.46±3.21	99.68 ±19.53** b
II	30.70±3.81	59.56 ± 10.47* a
III	42.08±6.76	84.53 ±5.60** b
IV	35.83±3.85	55.96 ±2.70** a

*= Means bearing superscript * differs significantly (p≤0.05) from 0 hours within the group.

**= Means bearing superscript **differs significantly (p≤0.01) from 0 hours within the group.

a, b = Means bearing superscript a, b differs significantly (p≤0.05) between groups I and II and groups III and IV at corresponding intervals.

Fig. 10: Aspertate Aminotransferase (IU/L) in different groups of animals at different intervals



The mean ± SE., values of aspertate aminotransferase (IU/L) of group IV animals on 0 hours and on 14th day post-operatively were: 35.83±3.85 and 55.96±2.70 respectively.

The comparison within the group showed that there was significant ($p \leq 0.01$) increase in aspertate aminotransferase level in all the groups of animal on 14th day when compared to 0 hours. The increased level was within the physiological limit in group II and IV animals. Group I and group III animal had higher aspertate aminotransferase level than the normal physiological limit.

The comparison between the group showed group II and group IV animals had significantly ($p \leq 0.005$) lower aspertate aminotransferse when compared to group I and group III animals.

4.1.2.2 Lactate Dehydrogenase (IU/L)

The mean ± SE., values of lactate dehydrogenase (IU/L) are given in table (14) and fig (11).

The mean ± SE., values of lactate dehydrogenase (IU/L) of group I animals on 0 hours and on 14th day post-operatively were: 102.11±20.36 and 195.16±13.21 respectively.

The mean ± SE., values of lactate dehydrogenase (IU/L) of group II animals on 0 hours and on 14th day post-operatively were: 125.92±19.62 and 208.13±19.23 respectively.

Table 14: Mean ± SE., values of Lactate Dehydrogenase (IU/L) in different groups of animals at different intervals

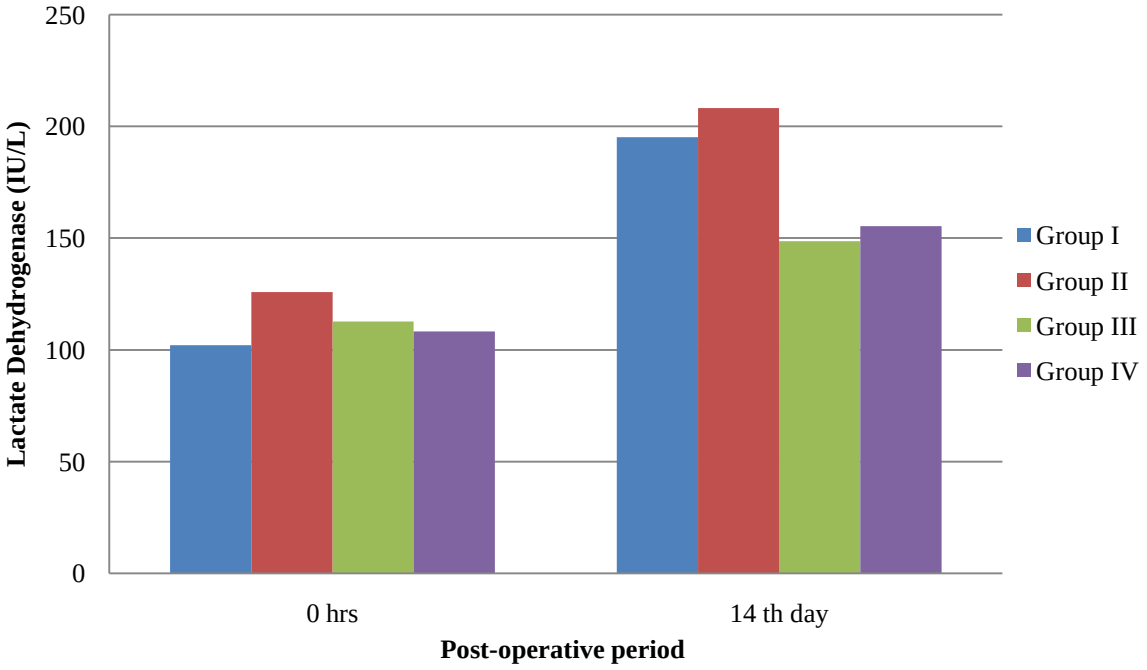
Groups	0 hours	14th day
I	102.11 ±20.36 [a]	195.16 ±13.21** [a]
II	125.92 ±19.62 [a]	208.13 ±19.23** [b]
III	112.74 ±9.71 [a]	148.59 ±8.48* [c]
IV	108.30 ±7.64 [a]	155.32 ±16.43* [c]

*= Means bearing superscript * differs significantly (p≤0.05) from 0 hours within the group.

**= Means bearing superscript **differs significantly (p≤0.01) from 0 hours within the group.

Means bearing different superscript, differs significantly (p≤0.05) between the group at different intervals.

Fig. 11: Lactate Dehydrogenase (IU/L) in different groups of animals at different intervals



The mean ± SE., values of lactate dehydrogenase (IU/L) of group III animals on 0 hours and on 14th day post-operatively were: 112.74±9.71 and 148.59 ±8.48 respectively.

The mean ± SE., values of lactate dehydrogenase (IU/L) of group IV animals on 0 hours and on 14th day post-operatively were: 108.30±7.64 and 155.32±16.43 respectively.

The comparison between 0 hours and 14th day within the group showed that, there was significant ($p \leq 0.05$) increase in lactate dehydrogenase level in all the groups of animal when compared to pre-operative level. However, the level of lactate dehydrogenase remained within the normal physiological limit in all the groups of animal.

The comparison between groups on 14th day showed that, the lactate dehydrogenase level in group III and group IV animals was significantly ($p \leq 0.05$) lower than group I and group II animal. The group I animals had significantly lower lactate dehydrogenase level when compared to group II animal.

4.1.2.3 Creatine Phosphokinase (IU/L)

The mean ± SE., values of creatine phosphokinase (IU/L) are given in table (15) and fig (12).

The mean ± SE., values of creatine phosphokinase (IU/L) of group I animals on 0 hours and on 14th day post-operatively were: 70.99±7.65 and 168.79±25.26 respectively.

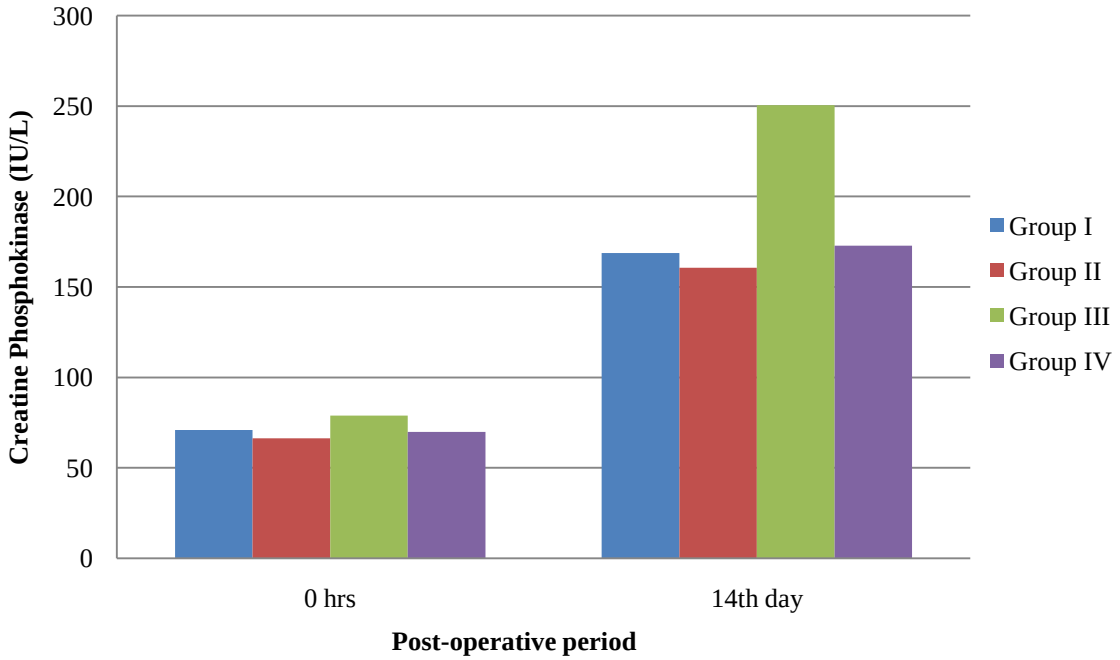
Table 15: Mean ± SE., values of Creatine Phosphokinase (IU/L) in different groups of animals at different intervals

Groups	0 hours	14th day
I	70.99±7.65	168.79 ±25.26** b
II	66.35±5.07	160.61 ±24.94** a
III	78.93±2.78	250.51 ±29.59** c
IV	69.92±7.57	172.88 ±5.13** c

**= Means bearing superscript **differs significantly ($p \leq 0.01$) from 0 hours within the group.

Means bearing different superscript, differs significantly ($p \leq 0.05$) between the group at different intervals.

Fig. 12: Creatine Phosphokinase (IU/L) in different groups of animals at different intervals



The mean ± SE., values of creatine phosphokinase (IU/L) of group II animals on 0 hours and on 14th day post-operatively were: 66.35±5.07 and 160.61±24.94 respectively.

The mean ± SE., values of creatine phosphokinase (IU/L) of group III animals on 0 hours and on 14th day post-operatively were:78.93±2.78 and 250.51±29.59 respectively.

The mean ± SE., values of creatine phosphokinase (IU/L) of group IV animals on 0 hours and on 14th day post-operatively were: 69.92±7.57 and 172.88±5.13 respectively.

The comparison within the group between 0 hours and 14th day showed that, on 14th day creatine phosphokinase level was significantly ($p \leq 0.01$) higher than pre-operative level.

The comparison between the group showed that, group II animals had significantly ($p \leq 0.05$) lower creatine phosphokinase level when compared to group I animal. The group I animals had significantly ($p \leq 0.05$) lower creatine phosphokinase when compared to group III and group IV animals.

4.1.3 Histopathological studies

Histopatholgical studies were conducted by collecting Obliques abdominus externus muscle biopsy in group I, group II, group III and group IV animal before suturing (0 day) and on 14th day after suturing.

Histopathology of obliques abdominus externus muscle biopsy collected on 0 day showed normal muscle fibers without any inflammatory reaction (plate 22) and blue coloured normal muscle fibers with striation and pink coloured collagen fiber (plate 23).

Histopathology of obliques abdominus externus muscle biopsy collected on 14th day in group I animal showed extensive fibroblast proliferation and collagen deposition with moderate infiltration of the mono nuclear cells (plate 24) and neovascularisation with deposition of collagen surrounding the suture area with severe mononuclear cell infiltration (plate 25).

In group II animal showed cellular infiltrate predominated neutrophils surrounding the suture (plate 27) and group III animal showed very mild inflammatory reaction surrounding the suture material with collagen deposition (plate 29) wheras, group IV animal showed very rare inflammatory cell and granulation tissue with new blood vessels (plate 31).

In group I (plate 26), group II (plate 28), group III (plate 30) and group IV (plate 32) animal with PTAH staining showed muscle fiber blue colour and collagen fiber pink colour.

4.1.3 Electron microscopic study

Obliques abdominus externus muscle biopsy with suture collected in group I, group II, group III and group IV animal on 14th day after suturing. Scanning electron microscopic observation of suture material surface on 14th day in group I(chromic catgut) animal showed disruption of suture fiber (plate 33) and circular electron dense material

over suture material (plate 34). Whereas, multifilament suture material on 14th day in group II (polyglycolic acid) and group III (polyglctin 910) showed intact suture filament without any disruption (plate 35), (plate 37) respectively, also showed Irregular electron dense material over filaments in group II (plate 36) and erythrocytes over and between the suture filament in group III animal (plate 38)**.** The monofilament suture material on 14th day in group IV (polydioxanone) animal showed homogenus surface (plate 39) and (plate 40).

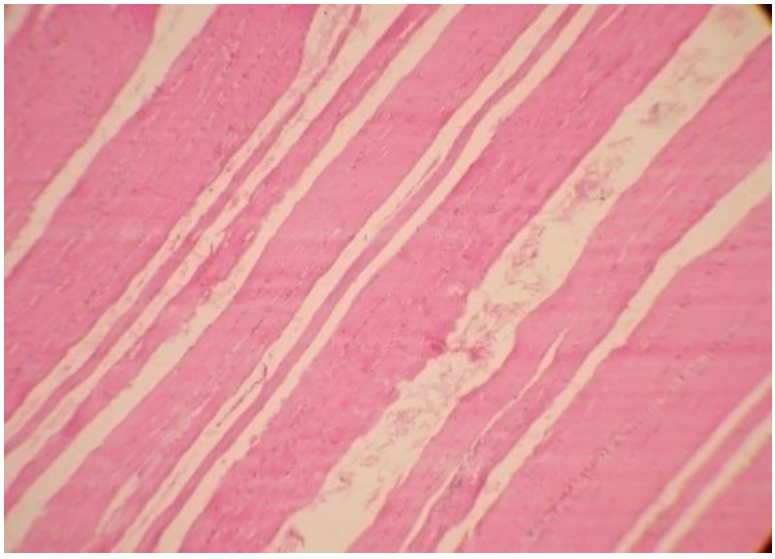


Plate 22: Photomoicrograph of abdominal muscle biopsy sample taken during laparotomy (0 day) showing normal muscle fibers without any inflammatory reaction. H&E, X10

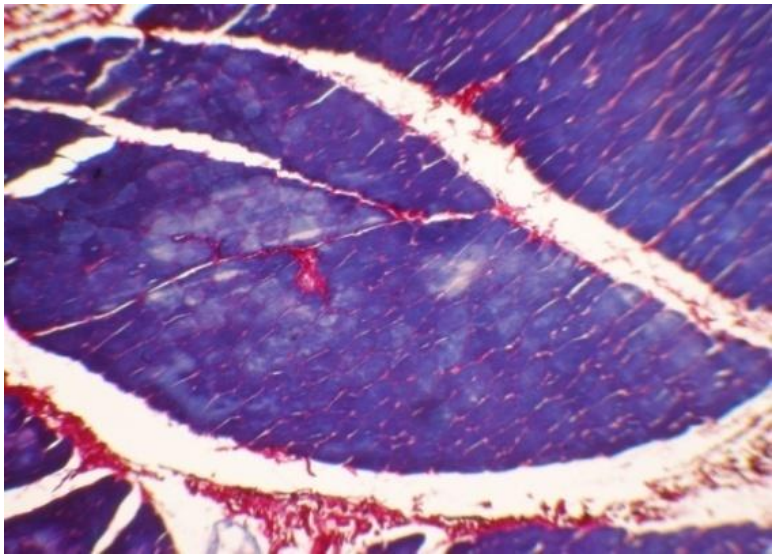


Plate 23: Photomoicrograph of abdominal muscle biopsy sample taken during laparotomy (0 day) showing blue coloured normal muscle fibers with striation and pink coloured collagen fiber. PTAH, X10

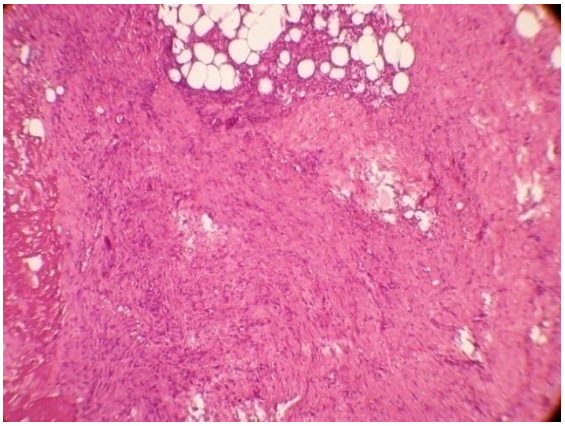


Plate 24: Photomicrograph of abdominal muscle biopsy sample taken on 14th post-operative day in a group I animal showing extensive fibroblast proliferation and collagen deposition with moderate infiltration of the mono nuclear cells. H&E, X10

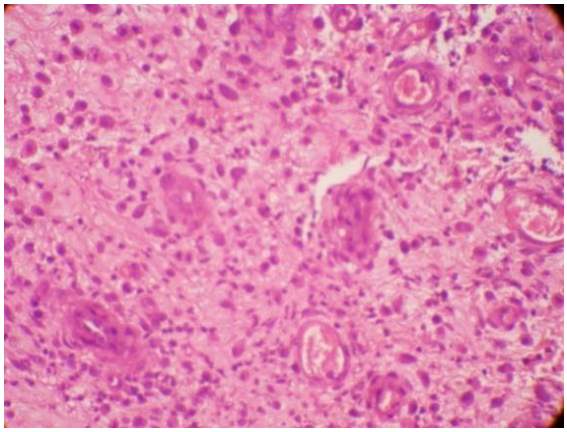


Plate 25: Photomicrograph of abdominal muscle biopsy sample taken on 14th post-operative day in a group I animal showing neovascularisation with deposition of collagen surrounding the suture area with severe mononuclear cell infiltration. H&E, X40

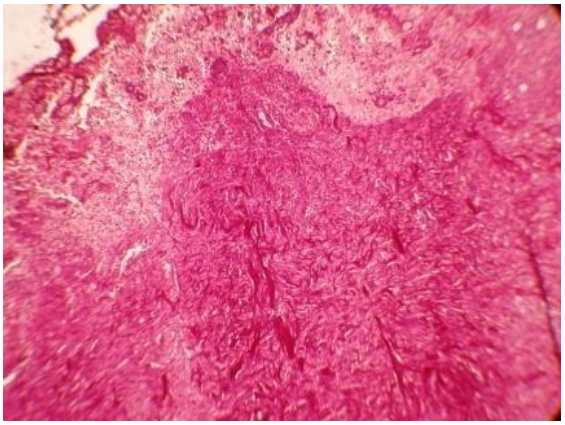


Plate 26: Photomicrograph of abdominal muscle biopsy sample taken on 14th post-operative day in a group I anima showing a pink coloured collagen fiber. PTAH, X10

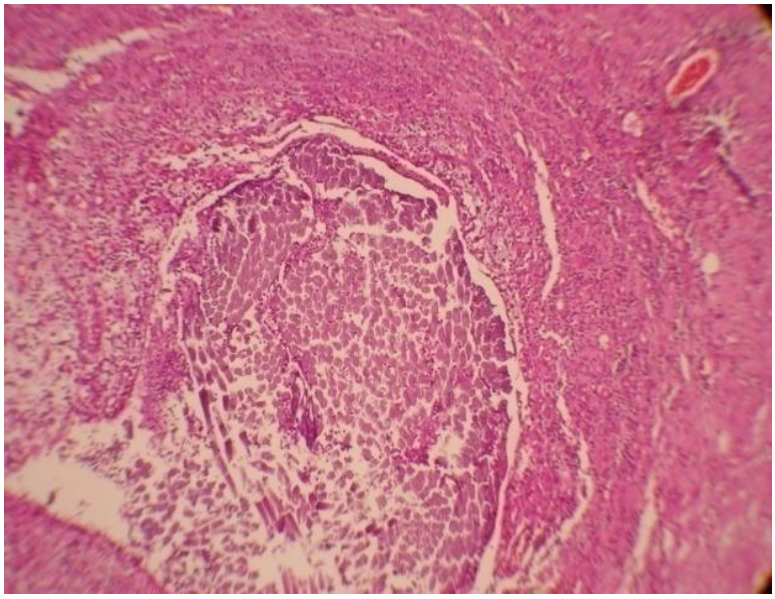


Plate 27: Photomicrograph of abdominal muscle biopsy sample taken on 14th post-operative day in a group II animal showing cellular infiltrate predominated neutrophils surrounding the suture. H&E, X10

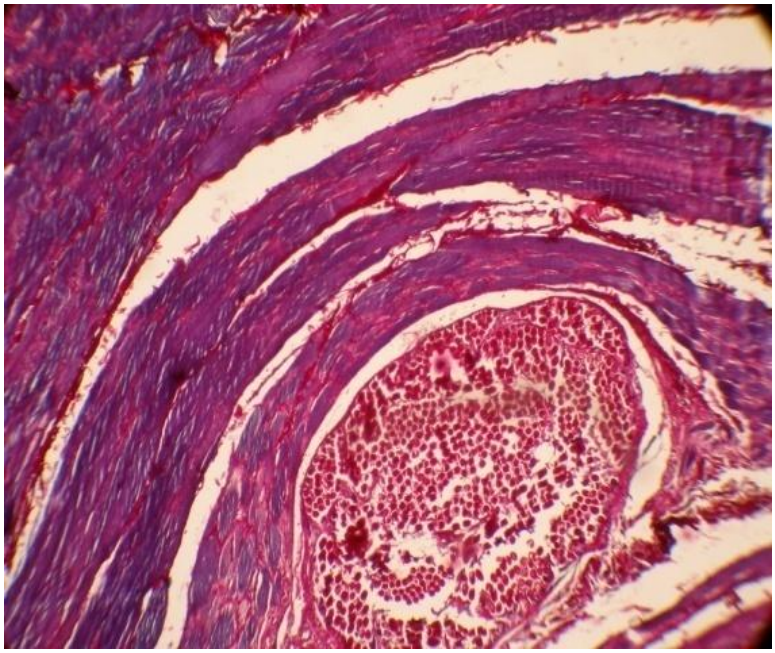


Plate 28: Photomicrograph of abdominal muscle biopsy sample taken on 14th post-operative day in a group II animal showing muscle fiber (blue) and wavy collagen fiber (pink) around the suture. PTAH, X10

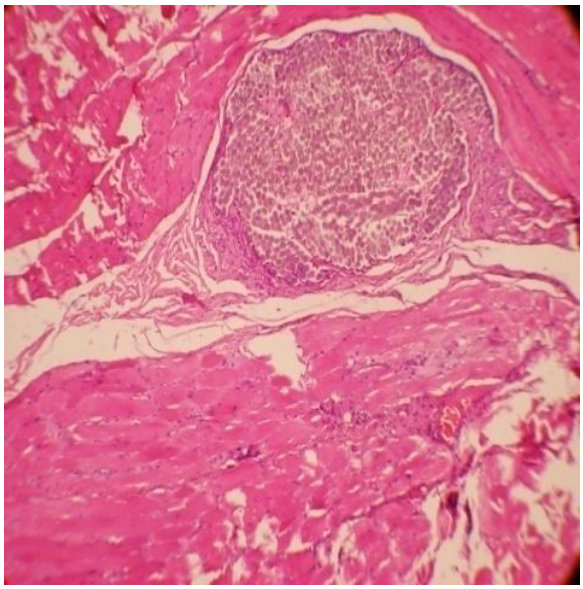


Plate 29: Photomicrograph of abdominal muscle biopsy sample taken on 14th post-operative day in a group III animal showing very mild inflammatory reaction surrounding the suture material with collagen deposition. H&E, X10

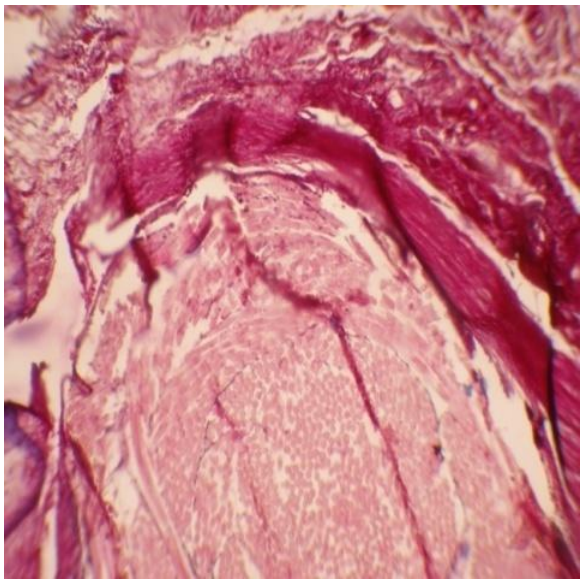


Plate 30: Photomicrograph of abdominal muscle biopsy sample taken on 14th post-operative day in a group III animal showing collagen fiber around the suture material. PTAH, X10

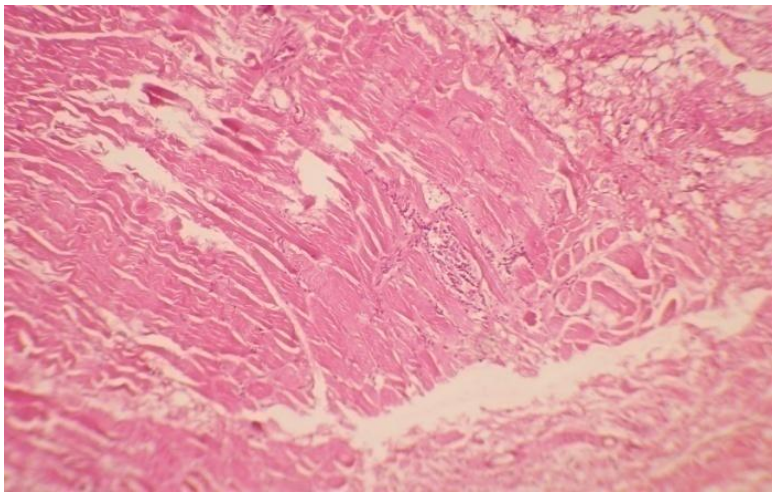


Plate 31: Photomicrograph of abdominal muscle biopsy sample taken on 14th post-operative day in a group IV animal showing very rare inflammatory cell and granulation tissue with new blood vessels. H&E, X10

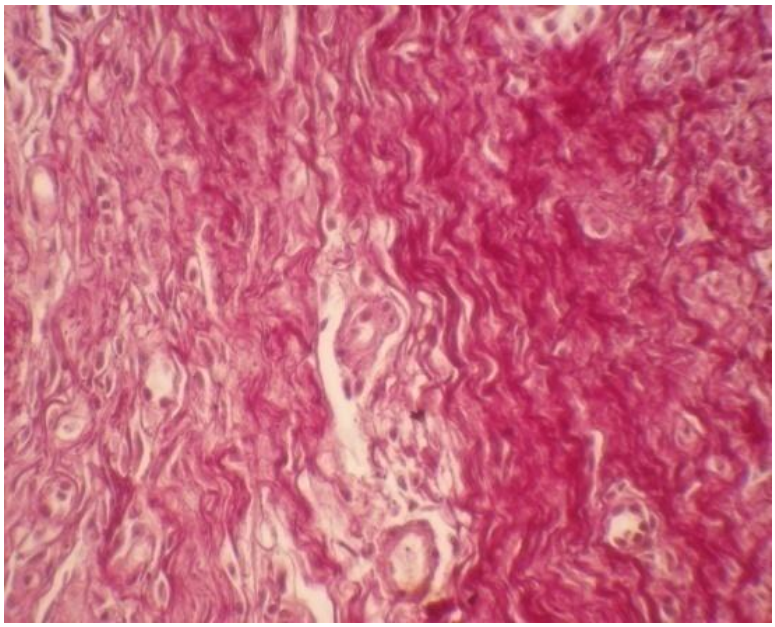


Plate 32: Photomicrograph of abdominal muscle biopsy sample taken on 14th post-operative day in a group IV animal showing collagen fiber. PTAH, X10

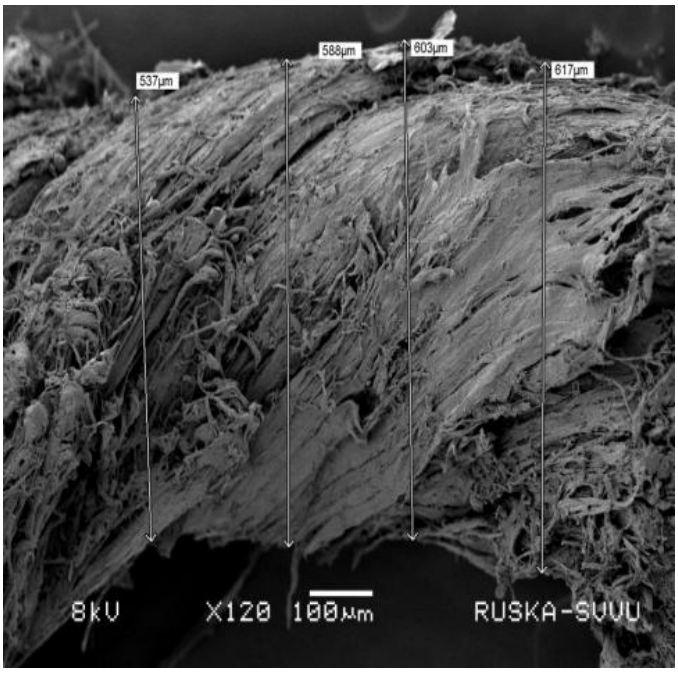


Plate 33: Scanning electron micrograph of chromic catgut on 14th day in group I animal showing disruption of suture material. X120

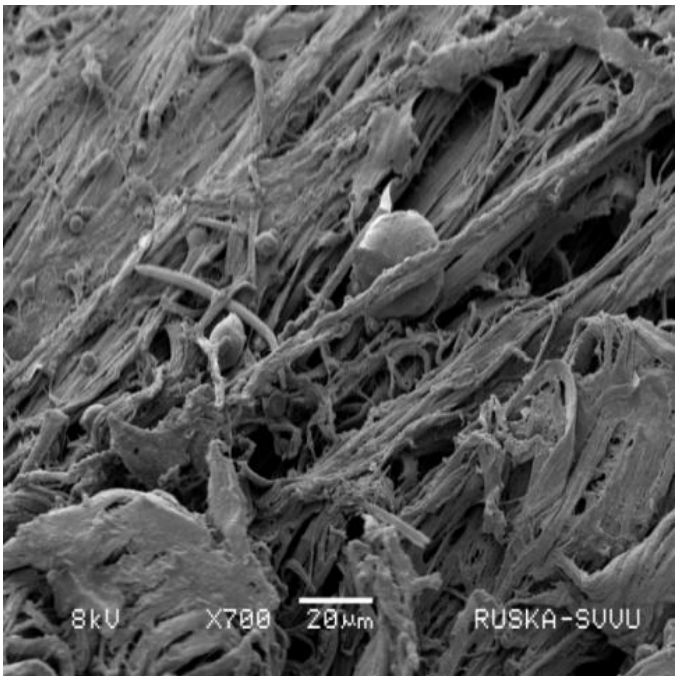


Plate 34: Scanning electron micrograph of chromic catgut on 14th day in group I animal showing circular electron dense material over suture material. X700

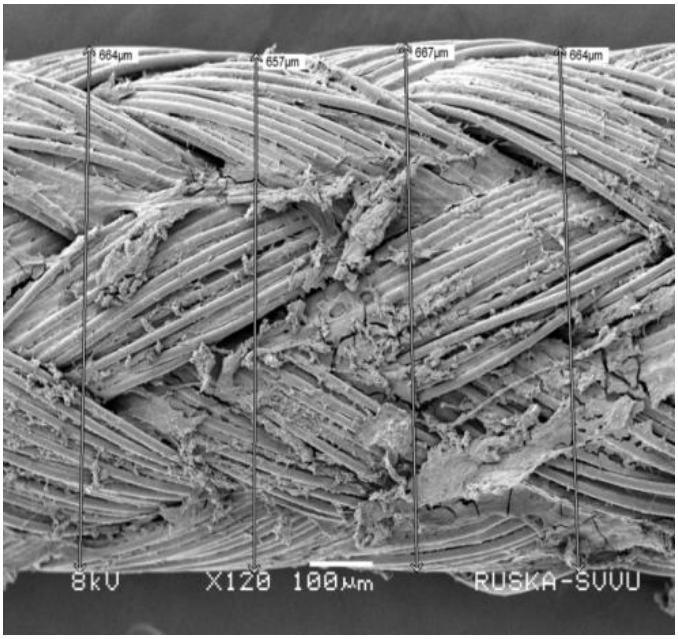


Plate 35: Scanning electron micrograph of polyglycolic acid on14th day in group II animal showing uniform tightly arranged suture filaments. X120

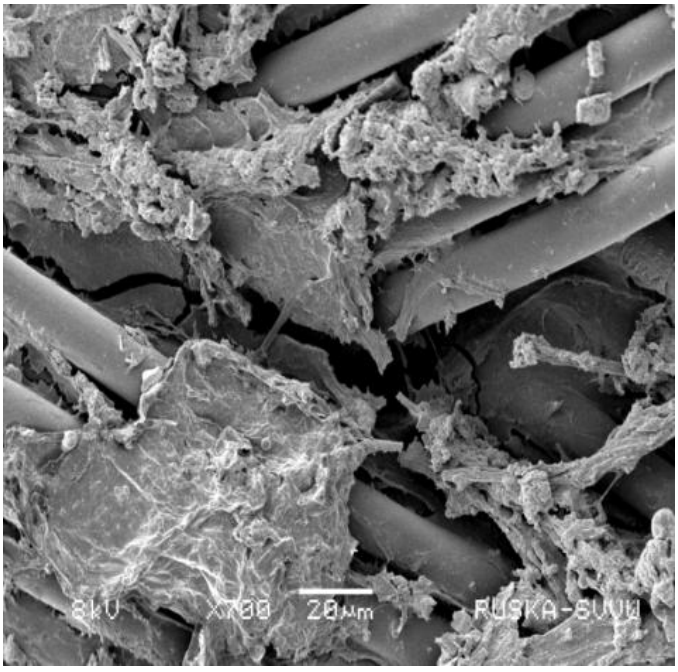


Plate 36: Scanning electron micrograph of polyglycolic acid on 14th day in group II animal showing irregular electron dense material over filaments. X700

Plate 37: Scanning electron micrograph of polyglactin 910 on 14th day in group III animal showing intact suture filaments. X100

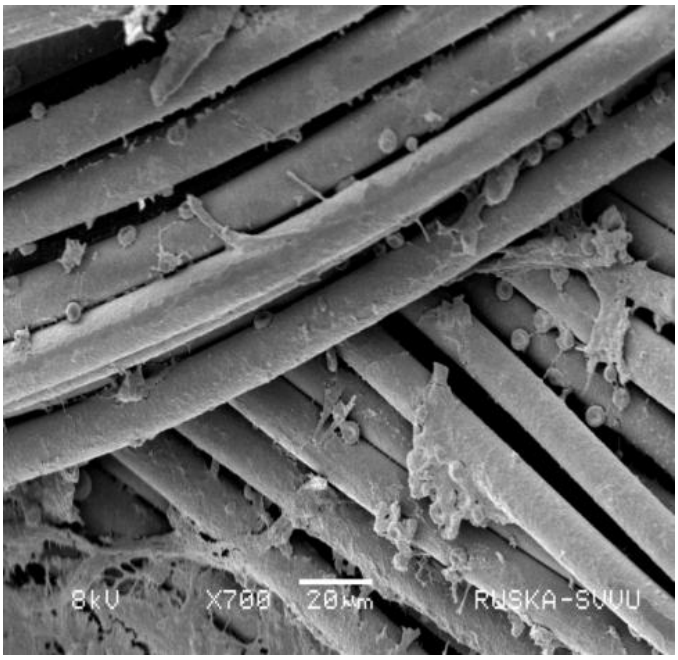


Plate 38: Scanning electron micrograph of polyglactin 910 on 14th day in group III animal showing erythrocytes over and between the suture filament and irregular electron dense material over filament. X700

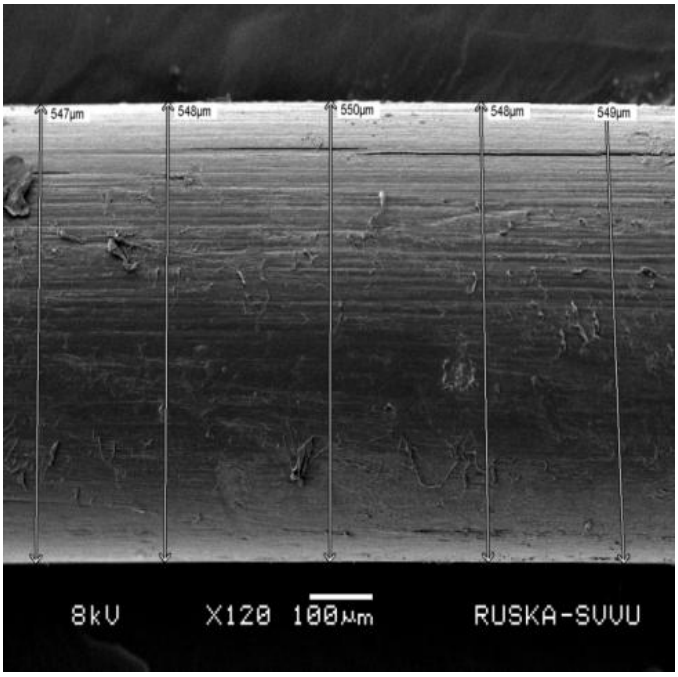


Plate 39: Scanning electron micrograp of polydioxanone on 14th day in group IV animal showing homogenous monofilament suture filament. X120

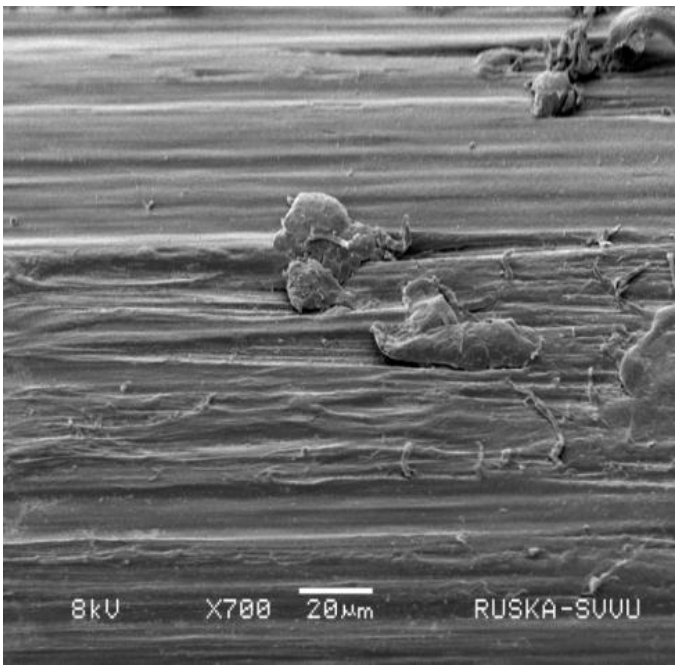


Plate 40: Scanning electron micrograph of polydioxanone on 14th day in group IV animal showing homogenus suture surface. X700

Discussion

V. DISCUSSION

5.1 Physiological parameter

5.1.1 Respiratory Rate (breaths/minute)

The respiratory rate was within normal physiological limit between pre-operative period to different post-operative intervals in group I and group III animals. Group II animal showed higher respiratory rate at 72 hours and 14th day. Whereas, group IV animal showed higher respiratory rate at 24 hours. Elevated respiratory rate could be attributed to pyrexia and pain in animal (Kelly, 1984; Coles, 1986 and Moitra, 1997).

5.1.2 Heart Rate (beats/minute)

The heart rate was within normal physiological limits between pre-operative period to different post-operative intervals in group II and group III. Whereas, animals of group I and group IV had higher heart rate at 72 hours and on 14th day in addition animals of group IV had significantly higher heart rate in compare to pre-operative level. Elevated heart rate could be attributed to pain and pyrexia in animal (Kelly, 1984 and Moitra, 1997).

5.1.3 Rectal Temperature (°F)

The rectal temperature was higher at 48 hours and 72 hours post-operatively when compared to pre-operative level in all the groups of animals, in addition group II animals showed higher rectal temperature at 24 hours when compared to 0 hours. The elevated rectal temperature could be due to pain and inflammatory conditions during and after surgery (Coles, 1986 and Moitra, 1997).

5.2 Haematological parameters

5.2.1 Total Erythrocyte Count (million cells/microlitre)

There was no significant change in total erythrocyte count between pre-operative period to different post-operative intervals in group I, group II, group III and group IV animals.

5.2.2 Haemoglobin (%)

In all the groups of animals there was decreased haemoglobin level at all the post-operative interval of study when compared to pre-operative level. This could be attributed to minor blood loss during surgery or fluid retention and haemodilution post-operatively (Millis *et al,* 1992).

5.2.3 Packed Cell Volume (%)

In all the groups of animals there was decreased Packed Cell Volume level at all the post-operative interval of study when compared to pre-operative level. This could be attributed to minor blood loss during surgery or fluid retention and haemodilution post-operatively (Millis *et al,* 1992).

5.2.4 Total Leukocyte Count (thousand cells/micro litre)

The total leukocyte was statistically significant at 24 hours when compared to 0 hours in group I and group II animals. Whereas, group III and group IV animal showed that there was severe leukocytosis between 24 to 72 hours when compared to pre-operative level. This could be attributed to surgical stress and tissue damage (Coles, 1986 and Benjamin, 2001). These results were in accordance with Schmidt and Booker (1982).

Dharmaceelan *et al*. (2000) also found significant increase in total leukocyte count on post-operative day.

5.2.5 Neutrophils (%)

There was neutrophilia in the group I, group II, group III and group IV animals between 24 to 72 hours. The neutrophil level by 14^{th} day has reached normal in all the groups of animals. This could be attributed to response of body to surgical trauma, tissue manipulation and inflammation (Coles, 1986 and Benjamin, 2001). Millis *et al*. (1992) found neutrophilia at 24 hours following ovariohysterectomy in bitches. However, Dharmaceelan *et al.* (2000) reported no significant change in neutrophil count after operation.

5.2.6 Eosinophils (%)

The eosinophil count of group I, group II, group III and group IV animals remained within normal physiological limits.

5.2.7 Lymphocyte (%)

There was lymphocytopenia in group I, group II, group III and group IV animals between 24 to 72 hours. This could be attributed to body response to systemic stress (Benjamin 2001). Millis *et al.* (1992) found lymphocytopenia at 24 hours after ovariohysterectomy in bitches. Dharmaceelan *et al.* (2000) observed lymphocytopenia during surgery and lymphocytosis during post-operative day.

5.2.8 Monocyte (%)

The monocyte count in all the groups of animal remained within normal physiological limits.

5.3 Biochemical Parameter

5.3.1 Aspertate Aminotransferase

There was increase in aspertate aminotransferase level in all the groups of animal on 14^{th} day when compared to pre-operative period (0 hours). The increased level was within the physiological limits in group II and group IV animals. Group I and group III animal had higher aspertate aminotransferase level than the normal physiological limit. The increased aspertate aminotransferase lavel might be due to increased in body temperature due to stress in animals as reported by Deswal and chohan (1981).

5.3.2 Lactate Dehydrogenase

There was increased lactate dehydrogenase level in group I, group II, group III and group IV animals when compared to pre-operative level. However, the level of lactate dehydrogenase remained within normal physiological limits in all the groups of animals.. The increased lactate dehydrogenase level might be due to increased body temperature (Spur 1972), The increased lactate dehydrogenase level was also due to temperature stress in animals (Deswal and Chohan 1981).

5.3.3 Creatine Phosphokinase

There was increased Creatine Phosphokinase level in group I, group II, group III and group IV animals when compared to pre-operative level. However, the level of

creatine phosphokinase remained within normal physiological limits in all the groups of animals. Cretine Kinase was sensitive and specific indicator of muscle damage in dogs and horse (Gerber 1964). Creatine kinase was not a predictable indicator of surgical stress (Austain *et al.,* 2003).

5.4 Clinical evaluation

In the present study none of animal showed skin wound oedema, infection and muscular tissue infection.

5.5 Histopathology

Histopathological studies were conducted by collecting Obliques abdominus externus muscle biopsy in group I, group II, group III and group IV animals before suturing (0 day) and on 14^{th} day after suturing.

The chromic catgut elicited maximum inflammatory response followed by polyglycolic acid, polyglactin 910 and polydioxanone.

The polydioxanone and polyglactin 910 elicited very minimal inflammatory reaction. These results are in agreement with several earlier worker (Edlich *et al.,* 1973; Tabuer *et al.,* 1975; Urdhal, 1975; Cassie, 1977; Pavan, 1979; Varma *et al.,* 1981; Freeman *et al.,* 1987; Sanz e*t al.,* 1988; Shuhaiber *et al.,* 1989; Smeak *et al*., 1989; Sharma *et al.,* 1991; Vipond *et al.,* 1991; Greenwald *et al*., 1994; Bennet *et al.* 1997; Wainstein *et al*., 1997; Molea *et al.,* 2000; Nary fiho *et al.* 2002; Thiede *et al*., 2002; John *et al*., 2004 and Kosan *et al*., 2008).

5.6 Electron microscopic study

Scanning electron microscopic observation of chromic catgut suture material surface on 14th day showed distruption of fibers these results are in agreement with the finding that of Stone *et al*., (1985). Whereas, polyglcolic acid, polyglactin 910 showed intact filament without disruption and these result are in agreement with the finding that of chu *et al.,* (1982). Polydioxanone showed uniform intact surface.

Summary

VI. SUMMARY

A study was conducted to evaluate the effect of four suture materials Chromic catgut, polyglycolic acid, Polyglactin 910 and Polydioxanone absorbable suture materials for canine laparotomy wound closure in order to assess haematological, biochemical and physiological changes before and after surgery thereby monitoring the inertness of suture material for canine laparotomy. Histopathological, electron microscopic assessment of suture material reaction to the tissue were made before and after surgery in order to assess the tissue reaction caused by the suture material.

The four groups consisted of six animals each (group I, group II, group III and group IV) for laparotomy wound closure in canine. Chromic catgut in group I, polyglycolic acid in group II, polyglactin 910 in group III and polydioxanoe in group IV was used during the entire period of observation none of the animals developed any complications like post-operative infection or mortality.

The physiolgical parameters viz., respiratory rate was within normal physiological limit between pre-operative period to different post-operative intervals in group I and group III animals. Group II animal showed higher respiratory rate at 72 hours and on 14^{th} day. Whereas, group IV animal showed higher respiratory rate at 24 hours. The heart rate was within normal physiological limits between pre-operative period to different post-operative intervals in group II and group III. Whereas, animals of group I and group IV had higher heart rate at 72 hours and 14^{th} day in addition animals of group IV had significantly higher heart rate in compare to pre-operative level. The rectal temperature was higher at 48 hours and 72 hours post-operatively when compared to pre-operative

level in all the groups of animals, in addition group II animals showed higher rectal temperature at 24 hours when compared to 0 hours.

Haematological parameter, total erythrocyte count (million cells/microlitre) in group I, group II, group III and group IV animals there was no significant change in total erythrocyte count between pre-operative period to different post-operative intervals. In all the groups of animals there was decreased haemoglobin level at all the post-operative interval of study when compared to pre-operative level. In all the groups of animals there was decreased packed cell volume level at all the post-operative interval of study when compared to pre-operative level. The total leukocytosis was statistically significant at 24 hours when compared to 0 hours in group I and group II animals. Whereas, group III and group IV animal showed that there was severe leukocytosis between 24 to 72 hours when compared to pre-operative level. The neutrophils level in the group I, group II, group III and group IV animals there was neutrophilia between 24 to 72 hours. The neutrophil level by 14^{th} day has reached normal in all the groups of animals. The eosinophil count in all the groups of animal remained within normal physiological limits.

There was lymphocytopenia in group I, group II, group III and group IV animals between 24 to 72 hours. The monocyte count in all the groups of animal remained within normal physiological limits.

In biochemical parameter, there was increase in aspertate aminotransferase level in all the groups of animal on 14^{th} day when compared to pre-operative period (0 hours). The increased level was within the physiological limits in group II and group IV animals.

Group I and group III animal had higher aspertate aminotransferase level than the normal physiological limit.

There was increased lactate dehydrogenase level in group I, group II, group III and group IV animals when compared to pre-operative level. However, the level of lactate dehydrogenase remained within normal physiological limits in all the groups of animals.

There was increased Creatine Phosphokinase level in group I, group II, group III and group IV animals when compared to pre-operative level. However, the level of Creatine Phosphokinase remained within normal physiological limits in all the groups of animals.

Histopatholgical studies were conducted by collecting Obliques abdominus externus muscle biopsy in group I, group II, group III and group IV animals before suturing (0 day) and on 14^{th} day after suturing. Group I animal showed extensive fibroblast proliferation neovascularisatin and collagen deposition, surrounding the suture area with severe mononuclear cell infiltration on 14^{th} day. In group II animal showed cellular infiltrate predominated neutrophils surrounding the suture and group III animal showed very mild inflammatory reaction surrounding the suture material with collagen deposition wheras, group IV animal showed very rare inflammatory cell and granulation tissue with new blood vessels .Collagen fibers took pink colour and muscle fibers with blue colour in PTAH staining.

Scanning electron microscopic observation of suture material surface on 14th day showed disruption of suture in group I animal. Whereas, multifilament suture material on 14th day in group II and group III showed intact suture filament without any disruption. The monofilament suture material on 14th day in group IV animal showed uniform surface.

Based on above findings following conclusion were made

1. Suture materials of present study have got no effect on physiological, haematological and biochemical parameter changes.

2. Polydioxanone caused less inflammatory reaction, followed by polyglactin 910, polygyolic acid and chromic catgut.

3. Under scanning electron microscope there was no surface erosion on 14th day in polyglycolic acid, polyglactin 910 and polydioxanone suture.

4. Appropriate suture for laparotomy wound closure in order of their merit were polydioxanone, polyglactin 910 and polyglycolic acid.

5. Chromic catgut suture can be preffered where adhesions are desired in laparotomy.

6. Under scanning electron microscope there was surface erosion on 14th day in chromic catgut suture.

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Abstract

COMPARATIVE EVALUATION OF CHROMIC CATGUT POLYGLYCOLIC ACID POLYGLACTIN 910 AND POLYDIOXANONE SUTURES FOR LAPAROTOMY WOUND CLOSURE IN CANINES

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ABSTRACT

The study was conducted on 24 clinical cases of dogs to evaluate the effect of four suture materials using chromic catgut, polyglycolic acid, polyglactin 910 and polydioxanone in group I, group II, group III and group IV respectively and to assess haematological, biochemical and physiological changes before and after surgery in order to assess the inertness of suture material for canine laparotomy. Histopathological, electron microscopic assessment of suture material reaction to the tissue were made before and after surgery in order to assess the tissue reaction caused by the suture material. The physiological, haematological and biochemical parameter were within in the normal physiological limit in all the group of animal. Chromic catgut showed extensive fibroblast proliferation, neovascularisation and collagen deposition surrounding the sutured area with sever mononuclear cell infiltration. Polyglycolic acid showed predominated neutrophil surrounding the sutured area whereas, polyglactin 910 and polydioxanone caused less inflammatory reaction. By scanning electron microscopic study on 14^{th} day revealed there was surface change in chromic catgut whereas in polyglcolic acid, polyglactin 910 and polydioxanone there was no surface destruction. Histopathological and scanning electron microscopic studies on laparotomy wound healing were indicative that Polydioxanone can be preferred absorbable suture material in long term wound healing over polyglactin 910, polyglycolic acid and chromic catgut suture.