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TISSUE REACTION TO SURGICAL SUTURES  
IN INFECTED WOUNDS

Submitted by

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In partial fulfillment of the requirements  
for the Degree of Doctor of Philosophy

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WE HEREBY RECOMMEND THAT THE THESIS PREPARED UNDER OUR SUPERVISION BY SUDARSHAN VARMA ENTITLED TISSUE REACTION TO SURGICAL SUTURES IN INFECTED WOUNDS BE ACCEPTED AS FULFILLING IN PART REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY.

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## ABSTRACT OF THESIS

### TISSUE REACTION TO SURGICAL SUTURES IN INFECTED WOUNDS

A comparison was made between Dexon (braided polyglycolic acid), plain catgut, chromic catgut, multifilament steel, monofilament nylon, braided silk and Mersilene (braided dacron) in infected wounds.

Four groups of 15 dogs were used. Three identical rows of 27 subcutaneous pockets were created in each dog. Knots of 3/0 suture were embedded in the pockets. Each suture pocket in the first row was inoculated with 0.1 milliliter of a standard culture of Staphylococcus aureus, containing  $74 \times 10^6$  organisms. The other two rows received similar treatment with ten-fold and hundred-fold dilutions respectively. Each row had a control pocket inoculated with culture but no suture material, and a second pocket with only the culture medium.

Groups of dogs were sacrificed at 6, 10, 20 and 40 days respectively. The diameter of the lesion around each suture was measured grossly. Microscopic measurements of lesions and cellular infiltration were used to rate the tissue reactions subjectively.

Pockets containing suture materials had significantly more reaction than control pockets. The three dilutions of culture

produced significantly different reactions. In the acute stage of infection (6 days), steel and nylon elicited the least reaction and catgut produced the most. Silk, Mersilene and Dexon were intermediate. In more chronic stages (10, 20 and 40 days), relative reactions to Mersilene and silk increased. Although Dexon showed intense tissue reaction at 6 days, it elicited relatively little in chronic implantations. Steel and nylon remained relatively inert at all stages. At 20 and 40 days silk evoked the most intense reaction.

Microscopic measurements of lesions correlated well with gross measurements. Neutrophils were the predominant cells in the acute stage. Later, macrophages and fibroblasts predominated. Silk, Mersilene and the catguts attracted large numbers of neutrophils in chronic implantations, suggesting persistence of infection. The Splendor-Hoepli phenomenon was demonstrated around silk sutures at 40 days.

Suture materials play an important role in postoperative wound infections, and different suture materials elicit different degrees of reaction in contaminated wounds. In chronic implantations, the reaction to various suture materials differs from that observed in the acute stages of infection.

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## CHAPTER I

### INTRODUCTION

The modern surgeon is offered an ever widening array of absorbable and non-absorbable suture materials, with a wide range of physical configurations and chemical structures. The surgeon's choice of suture material has been based on various considerations, including the biologic interactions of suture materials and tissues. These interactions can alter the mechanical properties of the wound and must be understood to evaluate sutures for use in various tissues. Many attempts have been made to find the "ideal" suture but as yet no material has been developed that meets all the established criteria. The generally accepted characteristics of an ideal suture material include superior handling quality, good knot security, adequate tensile strength, weak allergenic properties, minimal tissue reaction and no adverse effect on a wound in the presence of infection.

The goal in surgical treatment of wounds is to restrict loss of tissue function to a minimum. The decisive factor in choosing a suture material should be to secure approximation of wound edges and to interfere minimally with the healing process. Unfortunately, many of the distressing complications of surgery, such as infection, wound disruption and chronic sinus formation occur in sutured wounds. The

problem of subcutaneous wound infection and "spitting" sutures has plagued the surgeon for years. Suture materials in a wound may affect local conditions unfavorably and lead to infection. It has been suggested that the degree of enhancement of infection by a suture material roughly parallels the inflammatory tissue reaction elicited by it.

Surgeons would welcome objective biologic data upon which they could base their choice of suture materials in both clean and infected wounds. An investigation of the relative merits of both conventional and newer suture materials in the presence of acute infection was conducted by the author in an earlier study (Varma, 1973). A new synthetic absorbable suture, Dexon,<sup>1</sup> was compared with six conventional sutures in infected wounds. The results of the short-term study were at variance with claims that Dexon is a relatively inert suture material. In light of this finding, a further investigation of sutures in infected wounds in a long-term study was felt desirable.

This study was conducted in four groups of dogs to quantitatively determine the amount of tissue reaction elicited by seven suture materials, at various stages of implantation, in a field contaminated with Staphylococcus aureus, a common pathogen of postoperative wound infections. Tissue reaction in comparatively acute stages of infection was studied in one group of dogs which were sacrificed 6

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<sup>1</sup>Dexon, Davis & Geck, American Cyanamid Co., New York, New York.

days after surgery. This group would compare with the time limits of the author's previous work. Reaction to more chronic implantations was evaluated in three other groups, sacrificed at 10, 20 and 40 days, to provide a more complete spectrum of observations.

## CHAPTER II

### REVIEW OF LITERATURE

#### Clinical Evaluation of the Newer Suture Materials

In most surgical operations suture materials are required for reconstruction of incised or damaged tissues and for ligation of major blood vessels (Everett, 1974). The choice of a suture material is, for most surgeons, largely a matter of habit and tradition (Everett, 1970). Each surgical specialty can document clinical and experimental evidence to emphasize the individuality of a suture material (Ulin, 1971). In light of recent progress, particularly since introduction of synthetic sutures, the relative merits of both conventional and newer suture materials have been documented by objective clinical data (Van Winkle and Hastings, 1972). A review of clinical evaluation of suture materials used in various surgical fields (in human patients, unless otherwise stated) is presented below.

Since the advent of polyglycolic acid (PGA), a new synthetic absorbable suture, Dexon, has been developed and used clinically. It has been shown to cause minimal tissue reaction when used in the skin or subcutaneous tissues (Herron, 1974; Jones and Shorry, 1975; Morgan, 1969; Summer-Smith and Dingwall, 1972). Clough and Alexander-Williams (1975) performed a randomized trial comparing

PGA subcuticular skin closure with interrupted silk skin closure in 152 patients. They illustrated the lack of tissue reaction to PGA in contrast to the inflammatory reaction to silk. Cocke (1972) also reported minimal tissue reaction to PGA in 20 patients undergoing plastic surgery. He claimed that PGA had adequate tensile strength to hold skin edges together during the critical stages of healing, confirming an earlier report (Morgan, 1969).

The above reports are at variance with the findings of other workers. Rose (1975) reported that while the immediate healing of wounds sutured with PGA was very good, the long-term cosmetic effect was poor. In 20 percent of upper abdominal wounds the scars gradually become raised, broad and hypertrophic in appearance, not unlike a keloid. These findings are similar to those of Foster (1975) and Watts (1975). In a retrospective study of paramedian wounds, the incidence of hypertrophic scars approached 50 percent with PGA compared to 15 percent in wounds closed with nylon or silk sutures (Cox and Simpson, 1975).

Miller (1973) has reported the use of polypropylene (Prolene) sutures in nearly 300 patients over a four year period. The scars formed after wound closure with this material have been esthetically pleasing due to minimal inflammatory reaction. Polypropylene has, therefore, been recommended for plastic surgery.

Horton et al. (1974) evaluated a new synthetic absorbable suture, polyglactin 910 (Vicryl), in 100 clinical cases in a variety of surgical operations. The suture was generally used in the skin, subcuticularly or subcutaneously. There was no appreciable tissue reaction around the suture and no late extrusions of suture material occurred.

General surgical wound closure with PGA has shown it to cause minimal tissue reaction (Anscombe, 1972; Anscombe et al., 1970; Bonke et al., 1973; Dardik et al., 1971; Delpín et al., 1970; Herrmann et al., 1970; Kelly, 1969; Liu and Hennessy, 1970; Morgan, 1969; Myllärnieme, 1971; Turner et al., 1972). Polyglycolic acid sutures elicited less tissue reaction than catgut (Anscombe et al., 1970; Borthwick, 1973; Miln et al., 1972) and other suture materials (Bonke et al., 1973; Myllärnieme, 1971). The in vivo tensile strength was found to be greater than that of catgut during the critical stages of wound healing (Anscombe et al., 1970; Liu and Hennessy, 1970; McCarthy, 1970). Postoperative wound complications have been minimal using PGA (Bonke et al., 1973; Delpín et al., 1970; Herrmann et al., 1970; Kelly, 1969; Liu and Hennessy, 1970; McCarthy, 1970). Dardik et al. (1971) reported an incidence of 3.9 percent wound infection using PGA while the infection rate was 9 percent in a concomitant series of operations in which a variety of

conventional sutures (catgut, silk, nylon, dacron and wire) were used. McCarthy (1970) reported 5.8 percent wound infection using PGA.

In a study of 296 closures of abdominal incisions, teflon-dacron sutures were compared with PGA in regard to the incidence of wound dehiscence, wound infections and subsequent development of incisional hernias (Gallitano and Kondi, 1973). The two types of sutures were equal in regard to wound dehiscence and wound infection. Both showed certain characteristics making them preferable to steel, catgut or retention sutures of any type. Polyglycolic acid had a definite superiority in the prevention of incisional hernias and superficial wound infections were easier to manage. Turner et al. (1972) encountered 11 percent wound complications with both PGA and conventional sutures.

Polyglycolic acid was compared with chromic catgut in 198 alternating patients in an open study of closure of operative wounds following excision of hemorrhoidal tissue (Khubchandani et al., 1974). Polyglycolic acid sutures were found to be clinically superior as evidenced by minimal edema and minimal tissue reaction.

Only one report has appeared in the literature evaluating PGA in clinical veterinary surgery (Borthwick, 1973). Polyglycolic acid sutures were used in 54 canine and 5 feline patients in a variety of different operations involving the abdomen, urinary bladder, vagina and skin. Uncomplicated healing occurred in approximately 90

percent of dogs and all cats. Factors of infection or tumor were culpable in the remaining 10 percent of the dogs.

Polyglactin 910, as another specific alternative to catgut, was evaluated subjectively in 72 unselected patients in both elective and emergency surgical treatments (Martyn, 1975). Follow-up observations for up to 30 days proved it to be an inert absorbable suture material and it maintained adequate tensile strength even in two cases of wound infection. Similar findings were reported by Horton et al. (1974), who used polyglactin 910 to suture muscle, fascia and mucous membranes.

Dennis and Aka (1973) described a method of through-and-through abdominal wound closure with monofilament suture materials for poor-risk wounds. This method was free of postclosure evisceration and uncomplicated by significant suppuration. They felt that success appeared attributable to minimization of foreign suture material in the line of approximation. Monofilament closures were strikingly effective in reducing the infection rate as compared to closure with braided sutures of silk or dacron.

During the past several years the use of monofilament nylon has seemed attractive because of its relatively complete inertness to tissues. Unfortunately, nylon knots tend to untie and, as a substitute, non-absorbable monofilament polypropylene suture was introduced (Sanders, 1970). It was used in surgical operations in 151 patients



and was found superior to currently used materials such as silk, wire and nylon. An overall incidence of wound infection was 2.6 percent. Hermann (1974) evaluated polypropylene in 250 patients in whom a variety of abdominal incisions were closed. He concluded that the material was inert with minimal tissue reaction, resisted bacterial contamination or infection and was well tolerated by patients in the late postoperative period. Miller (1973) also found polypropylene to be relatively inert, evoking minimal inflammatory reaction. He reported an overall wound dehiscence rate in the acceptable range of 3 percent.

Parkash (1973) compared tissue compatibility of silk (Permahand) and dacron (Dacrofil) in 4000 abdominal operations. Statistically significant differences were noted in the two groups. There were far less complications with dacron (2.5 to 4 times less tissue reaction).

The newer synthetic suture materials have been evaluated in various specialized surgical operations. Favorable results with the use of PGA, in comparison with conventional sutures, were reported in oral surgery (Lilly et al., 1973; Nordenram and Bang, 1972; Wallace et al., 1970), gastro-intestinal anastomoses (Clark et al., 1972; McCarthy, 1970; Miln et al., 1972), thoracic surgery (Takita, 1970), urogenital surgery (Brannan et al., 1973; Holmlund, 1973; Long and Derrick, 1972; McGinn, 1973; Miller Jr., 1973; Mouzas

and Pompa, 1974), obstetrics and gynecology (Beard et al., 1974; Daume, 1972; Livingstone et al., 1974; Rahman and Way, 1972; Tompkins and Lea, 1972), orthopedic surgery (Allman, 1973) and ophthalmic surgery (Blau et al., 1975; Furgiuele, 1974; Merrit et al., 1974; O'Donoghue, 1970; Sugar et al., 1974; Walker, 1974; White and Parks, 1974).

An undesirable finding with the use of PGA in circumcision was that 25 percent of the sutures were still present at 28 days, a major disadvantage for this particular operation (McGinn, 1973). In ligation of large parametrial stumps or as running sutures in Cesarean sections the rough surface and cutting action were also found to be disadvantageous (Daume et al., 1972). A disadvantage noted in ophthalmic operations was that the braided nature of PGA tended to draw adventitia into the suture tract (Walker, 1974). Substitution of PGA sutures for chromic catgut in closure of knee joint capsules was associated with a 30 percent incidence of wound disruption (Scholz et al., 1972). It was hypothesized that the presence of a mild amount of postoperative reaction, as elicited by catgut, may be helpful in promoting more rapid wound healing.

Polyglactin 910, another synthetic absorbable suture, has also been used favorably in oral (Horton et al., 1974), orthopedic (Martyn, 1975) and ophthalmic surgery (Munton et al., 1974).

Pouliquen and Offret (1972) have suggested that infection is the most important consideration in choosing suture materials for corneal wounds. Synthetic monofilament sutures, such as polyamide fibers (nylon and perlon), have been found to be the least reactive of all materials used in the cornea (Pouliquen and Offret, 1972; Schmitt and Doden, 1972).

According to a poll of ophthalmologists, the majority prefer plain or chromic catgut for strabismus surgery (Dunlap, 1972). A recent increased incidence of reaction to collagen materials was also reported. A new synthetic non-absorbable suture material, braided polyester coated with polytetrafluoroethylene (Ethiflex), was evaluated for squint surgery and found to cause less tissue reaction than collagen (Landers, 1972).

### Inflammatory Tissue Reaction to Surgical Sutures

The goal of surgical treatment of wounds should be to restrict to a minimum the loss of function by providing ideal conditions for healing. Decisive factors in the choice of suture materials should be that they secure approximation of wound edges during the necessary time and have minimal influence on the healing process (Brunis and Zederfeldt, 1970). Some of the complications following surgery may be directly attributable to the suture material itself (Everett, 1974). A certain degree of inflammatory reaction is a prerequisite for repair but a marked inflammatory reaction results in impaired

healing. Sutures are foreign bodies which induce and prolong tissue reaction in the wound area (Brunius and Zederfeldt, 1970). Experimental and clinical studies have demonstrated that the more intense and prolonged the tissue reaction to suture material is, the greater is the risk of inadequate maintenance of wound approximation and disturbed healing (Brunius and Zederfeldt, 1970).

Different suture materials evoke various degrees of inflammatory reaction in tissues and with the use of complicated grading systems, differences among the reactions to suture materials can be detected (Van Winkle and Hastings, 1972). As a research tool, the technique of histopathologic grading of tissue reactions by Sewell et al. (1955) has proven valuable and, with slight modifications, is the basis of comparison of suture reactions in most experimental studies. This microscopic evaluation is based on the diameter of the inflammatory response, the overall cellular density and an estimate of the number of each of the cell types (mononucleated phagocytes, neutrophils, eosinophils, lymphocytes, fibroblasts and giant cells) per oil immersion field. Empiric grades and weighing factors are assigned to each parameter, and a single value is obtained for each section examined, by multiplying the respective grades assigned by their corresponding weighing factor. The final figure determines the degree of tissue reaction, the higher numbers indicating more severe or acute reactions.

Several studies have been made to evaluate biologic changes in a sutured wound and the interaction of sutures and various tissues.

Sewell et al. (1955) studied suture implants obtained from rats, rabbits and dogs, to compare tissue reactions caused by sutures of beef intestine serosa with sutures of sheep intestine submucosa. Bovine and ovine catgut did not differ significantly in any given species in the degree of tissue reaction occasioned by their implantation. Plain catgut produced slightly greater inflammatory response than chromic catgut.

Postlethwait (1968) made an extensive study of histologic reaction to various surgical sutures. He suggested that an exact description of tissue reaction to catgut is difficult to provide as the response at any specific time is dependent on the stage and rapidity of absorption. The reaction to plain catgut was mainly granulomatous, consisting of fibroblasts, histiocytes\* and giant cells and a few neutrophils. Chromic catgut had essentially a similar reaction but was generally a little less acute and slightly more prolonged. At the height of the absorption reaction the response was predominantly neutrophilic or histiocytic. With silk the predominant cells nearest the suture were histiocytes. More typically the fibrous tissue encapsulation surrounded a granulomatous type reaction consisting of

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\*The literature appears to be inconsistent in the use of the term histiocyte, but it is assumed reference is being made to mononuclear phagocytes or macrophages.

fibroblasts, histiocytes, giant cells, lymphocytes and a few capillaries. Neutrophils could usually be found but were not a major feature of the reaction. Cotton, although of a different chemical composition, elicited a similar reaction to that of silk, the major difference being earlier separation of cotton fibers with cellular invasion inbetween. Tissue response to metals was impressively bland. Early acute reaction occurred, followed mainly by fibroblastic and histiocytic response. Nylon, dacron and teflon elicited cellular reactions similar to that of silk but invariably less intense and rapidly replaced by a mature fibrous tissue capsule. The synthetic sutures also had little tendency to be separated by histiocytic or fibroblastic invasion. Multifilament dacron coated with teflon had a similar reaction to that of plain dacron, but at times the teflon appeared to be shed and incited some reaction in the vicinity of the suture.

In another long-term study by the same author (Postlethwait, 1970a), silk, cotton, dacron, nylon and polypropylene were implanted in the abdominal wall muscles of rabbits and tissue reaction graded at intervals up to two years. Monofilament sutures produced much less tissue reaction than the natural fibers, cotton and silk. Nylon evoked the least tissue reaction while dacron and polypropylene followed in that order but almost as a pair. The teflon-coated dacron sutures again shed the teflon and elicited more inflammatory reaction.

Tissue reaction to sutures was studied in 666 specimens obtained from human patients at a second operation or at autopsy from one day to 23 years after operation (Postlethwait et al., 1975). It was again emphasized that reaction to catgut depended on the stage of absorption and was mainly histiocytic in type. Non-absorbable sutures were encapsulated by a rim of connective tissue, while near the suture, histiocytes, giant cells and lymphocytes were found. This was most marked with silk and cotton, less so with dacron and least with nylon and wire. Tissue infiltration into the suture interstices was variable. It was felt that although a precise comparison could not be made, the reaction in humans generally was similar to, but less intense, than that seen in experimental animals. Transfer of the findings in animal studies, therefore, appeared to be acceptable with only moderate limitations.

Kovács and Somogyvári (1969) implanted polyester (dacron), catgut and silk sutures in subcutaneous connective tissue and muscles of horses and dogs, and the nature of tissue reaction was followed over a period of 120 days. Comparative histological examination yielded best results with polyester. The local inflammatory reaction subsided soon after operation and the absorption phenomenon was very mild. After 45 days angio-fibroblastic tissue showing a marked fibrous transformation was seen creeping into the texture of the multifilament, dividing it into bundles and monofilaments. The

traditional suture materials, silk and catgut, were encapsulated by connective tissue. In the internal layer of the connective tissue capsule an intense cellular activity was apparent.

In another study (Dynowski, 1975), nine various kinds of surgical sutures were investigated in rabbits and guinea pigs. In the group of non-absorbable sutures, steel caused the least reaction and silk elicited the most intense reaction. Linen thread caused reaction of medium intensity.

With the advent of PGA, several studies have been conducted to compare inflammatory reaction elicited by it and other conventional sutures (Bergman et al., 1971; Echeverría and Jiménez, 1970; Eilert et al., 1971; Frazza and Schmitt, 1971; Hermann et al., 1970; Holm-Jensen and Agner, 1974; Katz and Turner, 1970; Kelly, 1969; Livingstone et al., 1974; Postlethwait, 1970b, 1974; Urdahl, 1975; Wandall et al., 1972). Minimal inflammatory response using PGA was repeatedly stressed, and it was found to evoke less tissue reaction than catgut and compared favorably with the synthetic non-absorbable materials.

Postlethwait (1970b) described the typical cellular response elicited by PGA. The initial reaction due to the passage of needle and suture was the same as with other materials. After the first seven days, less and less peripheral cellular reaction was noticed. Fibroblasts and histiocytes were the predominant cell types. Neutrophils



and lymphocytes were less frequent but giant cells were more common. Later, most of the reaction was between the filaments. When absorption was complete a cluster of phagocytic cells remained. The author noticed that while reaction to catgut was marked during the period of absorption, this did not occur with PGA which showed decreased reaction as filaments disappeared.

Absorption studies of PGA have shown that the suture material is completely absorbed by hydrolysis in 50 to 90 days (Eilert et al., 1971; Katz and Turner, 1970; Urdahl, 1975). Disintegration of fibers and poor suture recovery was evident as early as 20 days (Eilert et al., 1971; Holm-Jensen and Agner, 1974; Postlethwait, 1974), giving rise to concern that the material may not have adequate tensile strength to maintain wound edges during the important stages of healing. Lichtenstein (1970) has reported that healing wounds in rabbits were less than 30 percent of original tensile strength within two weeks and even after two months the wound was only about 41 percent as strong as normal tissue. He suggested that any suture material which loses most of its strength within two weeks should never be used to close supportive structures. Holm-Jensen and Agner (1974) have indicated that the rate of absorption of PGA is too short and for an absorbable suture material they still prefer catgut because of its longer absorption time.

Cutright et al. (1971) used rats in comparative studies to determine soft-tissue reactions and speeds of biodegradation of polylactic acid (PLA) and polyglycolic acid (PGA) sutures. The initial inflammatory reaction to PGA was greater, comparable with the reaction to braided silk suture, but both materials demonstrated a progressively milder response at later stages. Foreign body giant cells were evident throughout the study in all suture bearing areas, but this reaction remained essentially localized. The PGA strands composing the suture remained more oriented in comparison to PLA, where strands became confluent and somewhat dispersed after 11 postoperative days.

Conn et al. (1974) showed that polyglactin 910 sutures elicited minimal acute or chronic inflammatory response in the muscles of rabbits. The early acute reaction was similar to that of silk and plain or chromic catgut. After 30 days there was diminished response to polyglactin 910 sutures, and by 60 days they were completely absorbed without any evidence of chronic inflammatory reaction.

Munton et al. (1974) also reported that polyglactin 910, when implanted in animal tissue, evoked a mild tissue response. Absorption, occurring by hydrolysis, was complete by 80 days. In another study (Craig et al., 1975) polyglactin 910 was compared to PGA in implantation studies in rats. Based on histologic examination, both suture types elicited minimal tissue reaction. Virtually all remnants

of polyglactin 910 were absorbed by 90 days, while considerable quantities of PGA persisted at 120 days.

Miller (1970, 1973) reported preclinical surgical evaluation of non-absorbable polypropylene in rats, rabbits and dogs. The early, acute inflammatory reaction, limited in degree and dominated by neutrophils, was seen after implantation. Implants then appeared essentially inert. There was early and rapid proliferation of connective tissue, which encapsulated the suture after several days. A comparative study was performed in rats using polypropylene, cotton, silk, stainless steel, braided dacron, teflon-coated dacron and nylon. At seven days postimplantation the most pronounced tissue reaction was observed with cotton. Although nylon and polypropylene evoked a mild tissue reaction, the response to the latter was minimal by comparison.

Kukuruz et al. (1972) tested a bactericidal plastic suture (Lietilan) in muscles of rabbits. At 24 hours leukocytic infiltration was more intense around silk than around the bactericidal sutures. After seven days both sutures were surrounded by granulation tissue consisting of mononuclear cells and budding-off vessels. Later, lymphoid cells and mononuclear macrophages appeared. More neutrophils and foreign body giant cells could be found around silk than Lietilan.

Nathan (1972) evaluated freshly prepared autograft collagenous fibers from tendons and aponeuroses in various tissues in rabbits. Histologically, a more intense cellular infiltration was always observed around silk and catgut than around autograft fibers.

Lilly et al. (1969) studied the reaction of the oral mucosa of dogs to various suture materials. Catgut, monofilament sutures, polypropylene and nylon caused less reaction than multifilament materials, dacron, teflon-coated dacron, silk and siliconized silk. Use of PGA in oral tissues showed it to evoke mild tissue reaction, comparable to that of monofilament sutures (Lilly et al., 1972) and less intense than that evoked by catgut or multifilament sutures (Lilly et al., 1972; Wallace et al., 1970). In another histological investigation (Nordenram and Bang, 1972) PGA evoked somewhat more tissue reaction in comparison with catgut in guinea pig oral mucosa. More giant cells and neutrophils were seen with PGA.

In experimental gastrointestinal anastomoses minimal tissue reaction was observed with PGA when compared with other conventional sutures (Aird and Matory, 1974; Bergman et al., 1971; Delpín, 1975; Delpín et al., 1970; Echeverría and Jiménez, 1970; Herrmann et al., 1970; Kelly, 1969). Hastings et al. (1975a) studied wound healing in the stomach and colon of mongrel dogs to evaluate effects of various suture materials. Polypropylene and plain and chromic catgut produced the least and silk the greatest

cellular reaction. Shuster (1973) reported a case of hemorrhagic gastritis due to silk suture. There was a chronic ulcerative gastritis and a foreign body reaction to silk suture at the site of anastomosis.

Polyglycolic acid sutures were found to produce the least inflammatory response of all commonly used sutures in the urinary tract (Adams et al., 1975; Bergman et al., 1971; Brannan et al., 1973; Morrow et al., 1974). Hastings et al. (1975b) examined the response of urinary bladder to various suture materials and found silk and braided dacron to produce the greatest tissue reaction. Plain catgut produced the least tissue reaction but both plain and chromic catgut were irregularly absorbed. Polyglactin 910 sutures produced tissue reaction comparable with chromic catgut, but were regularly absorbed between 28 and 70 days.

Condon (1970) compared the effects of plain and chromic collagen, nylon and virgin silk in corneal wounds of rabbits. Histologically, nylon showed the least reaction. Chandler et al. (1974) also demonstrated that eyes sutured with monofilament nylon were much less irritated and inflamed. Sugar et al. (1974) showed that corneal tissue reaction to PGA was comparable to that of silk or catgut.

The fate of PGA and rat-tail tendon sutures (RTTS) in the cornea was studied in rabbits by electron microscopy (Henriquez et al., 1974). A brisk granulomatous reaction accompanied by

vascularization was incited by PGA whereas RTTS integrated well into the stroma without significant inflammation.

Nathan (1972) implanted freshly prepared collagenous fibers, obtained from rabbit tendons or aponeuroses, in the cornea of the same animal. There was a zone of marked opacity due to infiltration and edema around silk and catgut while the cornea was clear around the autograft fibers. Histologically, dense infiltrations of mononuclear cells and occasional foreign body giant cells were seen around the silk and catgut sutures. In contrast the autograft fibers remained homogeneous, without infiltration of cells.

Polyglycolic acid sutures have been evaluated and compared with other sutures for use in vascular anastomoses. Alvarez-Cordero et al. (1973) reported that in the first days PGA elicited an inflammatory response equal to that of other suture materials. At 30 days PGA produced a poor inflammatory response. Silk and braided dacron elicited a fibroblastic reaction, leading to the formation of thick collagen bundles. The mild inflammatory response to PGA in vascular anastomoses has been reported previously by Dardik et al. (1970) and Delpín et al. (1970), the latter showing that PGA was superior to catgut but did not differ from silk.

A histopathologic study was made of the reaction of flexor tendons in dogs to 10 different suture materials (Srugi and Adamson, 1972). Nylon seemed to evoke the least reaction. Stainless steel

wire and other synthetic sutures were similar in their behavior. Silk and silicone-treated silk were the least favorable.

Biodegradable polylactic acid (PLA) polymers were investigated as possible replacement for non-degradable stainless steel in the repair of mandibular and maxillary fractures of dogs and for blow-out fractures of the floor of the orbit in monkeys (Kulkarni et al., 1971). In vivo studies of PLA implanted intramuscularly as sutures indicated that the tissue response to the polymer was similar to stainless steel and dacron.

In another trial (Toranto et al., 1974), small amounts of biodegradable substances consisting of presently available absorbable sutures were used with silicone or clear dental acrylic to develop a composite alloplastic implant. In the model under investigation the prosthesis of chromic catgut and silicone established superior fixation.

#### Sutures as Niduses for Concretions and Thrombi

The urinary and biliary tracts present unique problems in the choice of suture materials because the presence of any foreign body acts as a nidus for stone formation (Brunius and Zederfeldt, 1970; Delpin, 1975; Peacock and Van Winkle, 1970; Van Winkle and Hastings, 1972).

Silvennoinen (1970) compared silk and catgut placed in the gall bladders of rabbits and found concretions on silk sutures beginning at

approximately 28 days postoperatively. By 150 days, nearly 70 percent of the silk sutures formed a nidus for stones, while no concretions were found around chromic catgut sutures at any time period.

Cholelithiasis has been observed due to stones formed around non-absorbable sutures (Gunn, 1972; Mackie et al., 1973; Silvennoinen, 1970). Silvennoinen (1970) studied 77 patients who required a second operation of the biliary tract. In these patients, 10 percent had stones which contained either silk or polyester suture material. No stones containing catgut were found. Gunn (1972) reported a case of a common bile-duct calculus which had formed around a thread (cotton) cystic duct ligature performed six years previously. Mackie et al. (1973) reported three cases in which non-absorbable suture material used to ligate the cystic duct was subsequently found within the lumen of extrahepatic bile-ducts. In two cases a gall-stone formed de novo around a nucleus of either silk or nylon, while in the third case two nylon sutures, encrusted with calcareous material, were removed from the stump of the cystic duct.

It has been suggested that any commonly used suture left exposed to urine might act as a nidus for deposition of crystalline substances (Morrow et al., 1974). Obstruction in the ureter, when a silk suture formed the nidus for a calcified cast, illustrated the risk of recurrent stone formation when residual suture material is left within the urinary tract (Silber and Thornbury, 1973).



In an experimental study on rabbits, PGA was compared with catgut for bladder closure (Bergman et al., 1971). In a few animals concretions were found on the suture line closed with PGA but none with catgut. Further studies by Bergman and Holmund (1973) demonstrated complete absence of calculogenetic potential of both catgut and PGA. From this study it could not be precluded that urinary salts had a special tendency to precipitate on PGA fibers. Bartone and Gardner (1973), however, reported a higher incidence (4%) of calculi formation with PGA than with catgut in cystotomies performed in guinea pigs. Cystotomies were performed in rats and sutured with silk, catgut and PGA, using different suturing techniques (Baur and Seuter, 1973). A definite correlation was shown between suture technique and calculi formation but there was no significant difference between suture materials used, although the absorbable sutures (catgut and PGA) formed much smaller calculi than silk. Brannan et al. (1973) performed ureterotomies and cystotomies in dogs using chromic catgut and PGA for closures. Wound healing was excellent in both groups, without evidence of calculus formation. Their experience in clinical cases of urologic surgery also showed PGA to be non-calculogenetic. Morrow et al. (1974) performed genitourinary surgical procedures in mongrel dogs, wounds being closed with silk, chromic catgut and PGA sutures. By three to six weeks all PGA sutures had sloughed the intraluminal portion, the suture material

showing least clinical calculogenetic potential. By eight weeks one catgut and all silk sutures had crystalline excrescences.

Based on in vitro experiments, Van Winkle and Hastings (1972) suggested it is possible that stone formation is more dependent on the multifilament or monofilament character of the suture than on the material itself. Later Hastings et al. (1975b) studied formation of bladder-stones with various suture materials exposed in the bladder lumen of dogs. Calculus formation was observed with braided sutures, silk and dacron, but not with catgut or monofilament polypropylene.

The increasing use of prosthetic implants and microvascular anastomoses in cardiovascular surgery has generated an interest in the relative thrombogenicity of suture materials (Salzman, 1971). A suitable in vivo test in dogs was developed by Lixfeld and MacGregor (1974), providing an index of suture thrombogenicity. Monofilament polypropylene appeared to be significantly less thrombogenic than monofilament nylon or any of the braided sutures tested (braided polyester, teflon-coated braided polyester and braided silk). Wandall (1974) also recommended monofilament polypropylene or polyesters for use in prosthesis fixation in cardiovascular surgery. Risk of thrombus formation was reported to be highest with catgut and silk while stainless steel had low thrombogenic properties.

In vascular anastomoses in adult dogs, less than 10 percent of the PGA-sutured vessels became thrombosed, compared with 23 percent for silk-sutured anastomoses in control animals (Dardik et al., 1970), indicating that PGA was accompanied by a satisfactorily low incidence of thrombosis at suture line.

### Allergic Reaction to Sutures

Postoperative complications frequently encountered in surgical patients include rejection of sutures from the operative wounds. These disorders of healing are probably related to antigenicity of or delayed hypersensitivity to surgical sutures (Cochrane, 1969; Getzen and Jansen, 1966; Lord, 1973; Feacock and Van Winkle, 1970; Umińska et al., 1974).

Capperauld (1972) failed to demonstrate catgut hypersensitivity when he performed an analysis of plain and chromic catgut sutures coupled with immunological testing in rabbits. However, Umińska et al. (1974) illustrated that repeated implantation of surgical sutures seems to play a certain role in the hypersensitivity phenomenon. A study on catgut sutures by the regional lymph node test showed that catgut is an antigen for rabbits. Increased blast cells and plasma-cytes in the lymph node smears were demonstrated.

Getzen and Jansen (1966) observed that allergy to suture material may play a contributing role in the development of post-operative wound infections. They showed that 83 percent of the

patients with wound infections around silk and chromic catgut suture had a positive allergic response to the suture material. In patients in whom there was no evidence of wound infection only 5 percent demonstrated an allergic reaction to the suture material used. Synthetic materials like nylon, dacron and wire did not demonstrate any allergenic properties. They suggested that for routine usage and in patients who have demonstrated previous wound infection, synthetic non-allergenic materials such as nylon, dacron and wire should help reduce an overall incidence of wound infection. Lord (1973) also demonstrated that in patients with an abnormal reaction to sutures there is a statistically significant incidence of wound infection and dehiscence. He suggested this to be a delayed-type hypersensitivity reaction. Cochrane (1969) suggested that a certain proportion of surgical wounds, which break down and extrude stitches with local reaction, are not in fact due to infection, but due to the patient's sensitivity to and intolerance of the suture material itself.

Roe (1975) reported a case of sterile calcified granuloma on the external surface of a ventriculectomy, identified four years after closure of a ventricular septal defect. Sensitivity to silk suture material was suggested as the cause. The patient also had a history of multiple silk suture sinuses developing in the uninfected original thoracotomy incision. A report has appeared in *Texas Medicine* (\_\_\_\_\_, 1975) where a postoperative wound infection with subsequent drainage

after spinal surgery was thought to be caused by allergic reaction to black silk.

Liber and Choi (1973) observed an eosinophilic deposit around multifilament silk sutures in surgical and autopsy specimens obtained between 18 days and 3 years after operation. Its structure and staining characteristics were closely similar to the Splendore-Hoeppli phenomenon (SHP) which occurs around certain fungi and helminthic parasites. No SHP was found about catgut, nylon or cotton sutures or around teflon (Polytef), dacron or polyethylene prostheses. It was suggested that SHP represents an antigen-antibody reaction and that the formation of deposit is related to the biochemical and immunological nature of silk. There was a varying degree of suppuration present around 20 percent of silk sutures showing SHP.

No immediate or late extrusions of suture material were encountered when synthetic, absorbable polyglactin 910 was evaluated in 100 clinical cases (Horton et al., 1974), suggesting the material has non-allergenic properties. Similar experiences were reported with use of PGA sutures in general surgery (Miln et al., 1972) and in ophthalmic surgery (Furgiuele, 1974). However, Wandall et al. (1972) showed that repeated implantations of PGA at six to seven week intervals causes a change in the cellular response and a massive infiltration of eosinophilic cells suggestive of a hypersensitivity response. Similar findings were not observed with catgut.

Nathan (1972) described a technique for preparing fresh, collagenous fibers from tendons or aponeuroses of the body for use as surgical suture material in the same individual without alteration of natural immunological properties. A marked reduction of foreign body reaction was observed when autograft suture was tested in corneas of rabbits. In this study it was shown that autograft fibers eliminated the allergic reactions described with catgut and other absorbable suture materials.

#### Suture-Tissue Interreaction in Infected Wounds

The problem of subcutaneous wound infection has plagued the surgeon for years. It is evident that all surgical wounds are contaminated and bacteria can be cultured from a high percentage of so called "clean" wounds (Everett, 1970). However, the incidence of postoperative wound infection is only about 5 percent. The actual development of wound infection depends on the dose and virulence of the contaminating organisms, the resistance of the host and the local conditions of the wound.

It has long been recognized that suture materials can lead to development or persistence of local infection (Elek and Conen, 1957; James and MacLeod, 1961). These reports have given evidence to suggest that inoculation of bacteria into a wound does not necessarily dictate the development of an infection, but that the presence of suture

material in the wound may affect the local conditions unfavorably and lead to infection.

Elek and Conen (1957) demonstrated that the number of staphylococci necessary to produce an infection in the human skin was  $7.5 \times 10^6$  viable organisms. If a silk stitch was inserted into the wound as few as  $3 \times 10^2$  bacteria produced an infection.

James and MacLeod (1961) performed a more complete study, implanting small pieces of contaminated suture material subcutaneously into mice. They found that of the sutures used, silk, silicone-treated silk, cotton and dacron were most effective in potentiating infection. Nylon and human hair were less effective in that they required larger inocula of staphylococci. They concluded that the degree of enhancement of infection by the suture material roughly paralleled the inflammatory reaction caused by the material itself. They also studied the ability of different suture materials to pick up organisms from a staphylococcal culture. It was found that braided materials picked up about ten times as many organisms per unit length as monofilament nylon.

Carpendale and Sereda (1965) showed a greatly increased infection rate in experimentally contaminated skin wounds sutured with silk, compared with identical wounds closed with tape. They emphasized that percutaneous sutures provided an avenue for bacteria to enter the wound and to produce stitch abscesses. These findings have

been further illustrated by other workers. In a reproducible experimental wound infection model Edlich et al. (1968) demonstrated that there was an increased likelihood of infection attending primary closure of contaminated wounds, giving strong support to the thesis that percutaneous sutures should be avoided in closure of contaminated wounds. Schauerhamer et al. (1971) found that only 7.7 percent of taped wounds developed gross infection after contamination with Escherichia coli. Sutured wounds contaminated with either Staphylococcus aureus or E. coli during the first 48 hours after closure were susceptible to development of gross infection. In another experimental model, suture closure of dead space was found to enhance the incidence of wound infection (De Holl et al., 1974). The deleterious effects of suture closure of dead space appeared to be secondary to the foreign body in the wound.

Everett (1970) has stated that sutures impair the wound's ability to resist infection. The mechanisms by which sutures potentiate infection in wounds are multiple (Edlich et al., 1968; Everett, 1970). Sutures are foreign bodies which increase and prolong the inflammatory reaction in wounds and seem to be a predisposing factor for the occurrence of infection (Brunius and Zederfeldt, 1970; De Holl et al., 1974). An exudative foreign body reaction and local tissue autolysis may facilitate bacterial growth (Everett, 1970).



As early as 1867, Lister conceived that harmful bacteria lie within the interstices of silk sutures and if they could be killed, a ligature could be left in the body (MacKenzie, 1973). The theory of bacteria-harboring properties of braided suture materials has further been advanced by several workers (Alexander et al., 1967; Clough and Alexander-Williams, 1975; Delpin, 1975; Dennis and Aka, 1973; Edlich et al., 1968; Everett, 1974; Myers, 1971; Peacock and Van Winkle, 1970; Van Winkle and Hastings, 1972; Varma, 1973; Wandall, 1974; Watts, 1975). It has been suggested that the interstices between multifilament sutures through which leukocytes penetrate only with difficulty, serve as a refuge for devitalized tissue, blood and bacteria (Peacock and Van Winkle, 1970) and provide a site for bacterial multiplication (Van Winkle and Hastings, 1972). Under such circumstances contamination could be converted to infection. Thus, it is likely that the critical factor in converting a contaminated wound to an infected one is the physical construction of the suture rather than the material used (Van Winkle and Hastings, 1972; Wandall, 1974).

Van Winkle and Hastings (1972) observed the resistance that wounds closed with monofilament suture materials have to infection and recommended that in the presence of potential contamination, multifilament sutures and plain catgut should be avoided. In such situations they have preferred to use monofilament non-absorbable

suture materials such as steel, polypropylene or nylon, or if an absorbable suture is desired, chromic catgut. Other workers have also recommended use of monofilament sutures in preference to multifilament sutures or catgut in infected wounds (Alexander et al., 1967; De Holl et al., 1974; Delpín, 1975; Dennis and Aka, 1973; Everett, 1974; Hermann, 1974; Kratzer and Onsanit, 1974; Varma et al., 1974; Wandall, 1974).

Elkin (1940) noted that the number of infections was less in wounds sutured with silk as compared to those in which catgut was employed. In a comparative study with silk and catgut, infections occurred in 2.1 percent of clean wounds sutured with silk as compared to 9.4 percent with catgut. In potentially infected cases there was likewise a marked difference. The incidence of infection was 7.9 percent with silk compared to 21.4 percent with catgut. Myers (1971) also observed that infection often accelerates absorption of catgut and leads to the development of incisional hernia. Peacock and Van Winkle (1970) have, however, shown preference for absorbable sutures where contamination cannot be satisfactorily eliminated.

The incidence of corneal infection has been shown to be less with monofilament nylon than with braided materials (Pouliquen and Offret, 1972; Schmitt and Doden, 1972). However, Norn (1972) observed that the incidence of pus around sutures seems to be

independent of the suture material used (silk and collagen) and the postoperative interval.

Chronic discharging sinuses and "spitting" sutures are common in infected wounds (Everett, 1970, 1974; Myers, 1971). A local abscess around non-absorbable braided sutures, setting up a chronic infection with persistent drainage, clears only when the suture is extruded spontaneously or removed surgically (Myers, 1971; Everett, 1970, 1974; - 1975). It was reported that approximately 80 percent of infected wounds developed sinuses when braided silk was used as a suture material, compared to only 6 percent with monofilament sutures (Everett, 1974). "Spitting" has been reported to be rare with catgut (Myers, 1971).

With the advent of PGA, a new suture material was made available for clinical use. Using PGA clinically, Ferguson (1972) felt that the new material was much less likely to form stitch abscesses in subcutaneous tissue than catgut. Several other clinical reports have appeared recently, evaluating tissue reaction elicited by PGA in infected wounds.

Dardik et al. (1971) experienced an overall wound infection of 3.9 percent using PGA compared to 9 percent using a variety of conventional sutures (catgut, silk, nylon, dacron and wire). They felt that PGA was well tolerated not only in clean cases but also in situations where gross infections were present. Rahman and Way (1972)

reported similar findings in gynecologic surgery, the incidence of infection using PGA being 0.5 percent, which was four times less than that with conventional sutures. In another clinical trial, incidence of infection was considerably less with PGA (2.7%) than with catgut (26%) when the two sutures were used for circumcision (Mouzas and Pompa, 1974).

Gallitano and Kondi (1973) found PGA to be similar to teflon-dacron sutures in regard to wound dehiscence and infection. Both sutures showed a superiority in prevention of incisional hernias when compared to steel, catgut or retention sutures of any type. No significant difference in the incidence of wound infection was observed when PGA was compared with silk for skin closure (Clough and Alexander-Williams, 1975). Foster (1975) did not notice a high incidence of infection with PGA although a proportion of wounds proceeded to hypertrophic scar formation.

Clough and Alexander-Williams (1975) felt that by virtue of the braided nature of PGA, such a suture might "seal in" infection in contaminated wounds. Watts (1975) has reverted to use of catgut because in his experience where any infection occurred in a wound sutured with PGA there was a hazard of latent infection from bacteria being trapped in the interstices of the material. It was found necessary on occasions to totally excise wounds containing PGA.

Experimental investigations comparing sutures in infected wounds have been limited. With the advent of newer synthetic materials, biological data have gradually become available, upon which the modern surgeon could base his choice of suture material in contaminated or infected wounds.

Eilert et al. (1971) investigated the stability of PGA and chromic catgut in subcutaneous wounds in rabbits. The wounds were infected with S. aureus organisms and were evaluated at 5, 10, 15, 30 and 60 days. Their study demonstrated no detectable differences in the tensile strength or healing of wounds closed with PGA or chromic catgut sutures.

Wandall et al. (1972) studied the differences between the biological behavior of PGA and catgut with regard to tissue response, loss of tensile strength and the disappearance rate in infected wounds. It was found that catgut lost one-third to one-half of its tensile strength within the first two days, but after this only a slight decline occurred with one-third of its initial strength remaining at 60 days. Polyglycolic acid retained tensile strength with only 10 percent loss within the first six days of implantation, but from there on a rather rapid decrease took place and no strength could be measured at 20 to 21 days. The findings were similar in infected and non-infected wounds.

Herrmann et al. (1970) implanted suture loops of dacron, PGA, chromic catgut and plain catgut subcutaneously in rats and contaminated the loops with a 1:10 suspension of rat cecal contents in saline, in an effort to determine the resistance of various suture materials to infection. At seven days the loops were removed and tested in a tensiometer. Plain catgut showed a marked loss of tensile strength and dacron retained its original strength despite infection being present. Chromic catgut and PGA had loss of tensile strength intermediate between the two.

Alexander et al. (1967) investigated the problem of whether some suture materials are more likely to potentiate infection than others. They implanted different suture materials into subcutaneous pockets in rabbits. Each pocket was inoculated with S. aureus culture of varying dilution and the degree of infection assessed by the diameter of the abscesses around the suture. In their study, the non-absorbable, monofilament materials (nylon, flexon wire and polyethylene) resisted infection better than catgut or multifilament materials. Chromic catgut caused less infection than plain catgut or the braided materials. It was also found that iodized catgut withstood contamination better than chromicized or plain catgut. They concluded from their results that the use of monofilament material of practically any type was preferable to the use of multifilament suture materials in contaminated wounds.

A similar study was conducted in dogs by Varma (1973) comparing PGA sutures with monofilament steel and nylon, braided silk, dacron and catgut (plain and chromic) in subcutaneous wounds contaminated with varying dilutions of S. aureus. The findings were reported by Varma et al. (1974). It was clearly shown that the degree of infection induced was directly related to the infectious dose of S. aureus. The degree of infection increased in the presence of suture materials, confirming earlier reports of Elek and Conen (1957) and James and MacLeod (1961). In this study different suture materials elicited different degrees of reaction in contaminated wounds. Monofilament nylon and steel produced the least reaction and catgut the most. Braided sutures of silk, dacron and PGA were intermediate between the two groups. Although not statistically significant, chromic catgut produced slightly more reaction than plain catgut, contrary to the findings of Alexander et al. (1967) and Herrmann et al. (1970). It was concluded that the physical nature of the suture material played an important role in potentiating wound infection. A hypothesis was advanced that if PGA could be made available as a monofilament suture it would compare favorably with steel and nylon in infected wounds.

The experimental findings of Edlich et al. (1973, 1974) were at variance with those of Alexander et al. (1967) and Varma (1973). They felt that the physical configuration of the suture played a

relatively unimportant role and advanced a hypothesis that the chemical structure of the suture may be the most important factor in the development of early surgical infection. They studied various suture materials in infected wounds in mice. Incidence of infection with nylon or polypropylene was less than with other non-absorbable sutures. Dacron was next in reactivity. Stainless steel was more reactive than dacron but significantly less than multifilament sutures. Reaction was more pronounced with silk and cotton. Monofilament materials were less reactive than multifilament ones but not significantly so. Among the absorbables, PGA evoked the least reaction, significantly less than catgut. Coating the multifilament sutures with silicone, wax or teflon did not alter the incidence of early infection. They did, however, confirm earlier claims that the presence of even the least reactive suture in contaminated wounds exerts a significant potentiation of infection. It was suggested that the raison d'être for the superiority of nylon and PGA sutures in contaminated wounds may be related to the antibacterial activity of the degradation products of these sutures.

Ludewig et al. (1971) implanted iodized and chromic catgut sutures subcutaneously in mice and inoculated  $17 \times 10^6$  S. aureus organisms along the suture tracts. A statistically significant reduction in infection was noted in mice in which iodized catgut sutures were used, agreeing with the findings reported by Alexander et al. (1967).



Kukuruz et al. (1971) studied a plastic surgical suture 'Lietilam' for its bactericidal properties when subjected to various pathogenic organisms in rabbits. It was found that tissue reaction was more intense around silk than around the plastic sutures.

In an in vitro study, Gravens et al. (1973) demonstrated the antibacterial effect of treating sutures with silver. Catgut, dacron and silk were treated with a silver-zinc-allantoin complex. These sutures were contaminated with S. aureus and in the case of silk with P. aeruginosa also. Silk and dacron sutures treated with the silver complex and contaminated with S. aureus showed bacterial reduction over controls of 88.2 percent and 99.0 percent respectively. Bacterial recovery from silk sutures treated with the silver complex and contaminated with P. aeruginosa was reduced 99.9 percent.

Lilly et al. (1973) undertook an in vitro study, using a specially designed bacteriologic culture chamber, to determine if PGA suture inhibited transmission of bacteria. In this laboratory study, steel and catgut exhibited no evidence of bacterial transmission at 24 hours, while 25 percent of PGA and 74.1 percent of silk sutures exhibited bacterial transmission. They felt that this was a major factor in the decreased oral tissue reaction to PGA as compared to multifilament non-absorbable sutures.

In a recent series of in vitro tests, the time dependent alteration of catgut and PGA sutures was examined (Sebeseri et al., 1975).

In a sterile urine environment PGA dissolved on day 6 and in infected urine on day 3. Catgut did not dissolve at all in the sterile urine and in infected urine it dissolved only on day 8. In view of these results the use of PGA in the urine environment may cause concern especially if an infection exists. These workers have indicated that more laboratory studies on in vivo behavior of PGA in infected animal models are needed before it can be given preferential endorsement.

## CHAPTER III

### MATERIALS AND METHODS

#### Experimental Animals

Sixty animals were selected for this study from a pool of mongrel dogs. On acquisition each dog was assigned a number and given a physical examination. Only healthy dogs were chosen for continued study. Two milliliters of blood were withdrawn from the cephalic vein into a bottle containing ethylenediaminetetraacetic acid (EDTA). Routine hematology was performed. Hemoglobin (Hb) values were determined using a hemoglobinometer<sup>1</sup>. Packed cell volume (PCV) was obtained using the microhematocrit method (Schalm et al., 1975). A refractometer<sup>2</sup> was used to determine the total protein (TP). A Coulter counter<sup>3</sup> was employed to obtain the the total red blood cell count (RBC) and the total white blood cell count (WBC). Differential WBC counts were made from a blood slide (Schalm et al., 1975).

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<sup>1</sup>Hemoglobinometer, Coulter Electronics Inc., Hialeah, Fla.

<sup>2</sup>Refractometer, Atago, Japan.

<sup>3</sup>Coulter counter, Model Z<sub>B</sub>, Coulter Electronics Inc., Hialeah, Fla.

A fecal sample was taken from each dog and a parasite egg count was done using the McMaster egg counting technique (Soulsby, 1968). Results appear in the Appendix Table A1. Irrespective of the fecal egg count, all dogs were wormed with thenium compound tablets.<sup>4</sup> The dogs were vaccinated against canine distemper and canine contagious hepatitis using a combined tissue-culture-adapted (living) vaccine<sup>5</sup> and against leptospirosis using a combined Leptospira canicola and Leptospira icterohemorrhagiae vaccine.<sup>6</sup> After this initial treatment, the dogs were randomly housed in kennels in groups of three. They were fed daily and water was provided ad libitum.

After a period of not less than two weeks a second blood sample was taken for hematology. On obtaining a normal hemogram (Schalm et al., 1975), the dog was accepted for the experiment. Any dog whose blood values were not within normal limits had periodic blood samples taken until the hemogram was acceptable. Selected dogs were randomly allocated to groups A (6-day survival), B (10-day survival), C (20-day survival), and D (40-day survival), each group receiving one dog at a time. Thus 15 dogs were eventually allocated

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<sup>4</sup> Ancaris, Burroughs Wellcome and Co., London, England.

<sup>5</sup> Epivas T. C. plus, Wellcome Research Laboratories, Beckenham, England.

<sup>6</sup> Leptovax plus, Wellcome Research Laboratories, Beckenham, England.

Table 1. Experimental Animals. Total Number of Dogs = 60 (29M, 31F)\*. Avg. Weight ( $\bar{x}$ ) = 14.5 kg.

Group A			Group B			Group C			Group D		
Dog Number	Sex	Weight (kg.)	Dog Number	Sex	Weight (kg.)	Dog Number	Sex	Weight (kg.)	Dog Number	Sex	Weight (kg.)
2	M	17	8	M	10	12	F	11	10	F	15
16	M	15	18	F	13	20	M	20	17	F	11
29	M	11	25	M	18	6	F	16	15	F	16
32	M	15	31	M	14	38	M	18	23	F	12
27	F	11	35	M	10	34	F	10	14	F	10
4	F	13	42	F	15	47	M	15	52	M	20
26	F	11	40	F	11	53	F	13	55	F	13
56	F	15	44	M	18	36	M	16	37	M	10
63	M	14	33	M	18	54	F	18	41	F	15
13	F	10	64	M	16	65	F	11	61	F	16
62	M	18	11	F	15	59	M	11	66	F	14
70	F	13	5	F	11	69	M	18	67	M	14
78	F	14	68	M	15	74	M	18	77	F	11
72	M	20	51	F	10	71	M	15	82	M	13
88	F	20	76	F	13	81	M	17	83	M	17
$\bar{x} = 14.5$ kg. (7 M, 8 F)			$\bar{x} = 13.8$ kg. (8 M, 7 F)			$\bar{x} = 15.1$ kg. (9 M, 6 F)			$\bar{x} = 14.5$ kg. (5 M, 10 F)		

\* (M = Male, F = Female)

to each group. They were comprised of both males and females and weighed from 10 to 20 kg (Table 1). Final hematologic values of PCV, TP, WBC and differential WBC count for each group are given in the Appendix (Tables A2, A3, A4 and A5 respectively). Criteria used for selection were a PCV of not less than 35 percent, TP of not less than 6 grams per 100 milliliters and WBC of not more than 18,000 per cubic millimeter. Table 2 shows means and standard deviations of above parameters for each group.

Although considered ideal to operate on one dog from each group at one session, it was not found feasible in this experiment due to limited surgical facilities and kennel space. Only three dogs, representing different groups, were operated on at any given session, using the same bacterial culture preparation. The groups were rotated uniformly throughout the experiment, thus eliminating differences between groups due to differences in bacterial cultures or experimental conditions.

#### Infective Material and Preparation of Bacterial Culture

The infective material was a pure culture of Staphylococcus aureus which had been used in a previous study by the author (Varma, 1973). It was obtained from a human postoperative wound infection. The organism was hemolytic, coagulase positive and a mannitol salt fermenter. It remained pathogenic for dogs.

Table 2. Means ( $\bar{x}$ ) and Standard Deviations (s. d.) of Hematologic Values for the Four Groups.

Group	PCV		TP		WBC		Differential Count (%)							
							TN		L		M		E	
	$\bar{x}$	s. d.	$\bar{x}$	s. d.	$\bar{x}$	s. d.	$\bar{x}$	s. d.	$\bar{x}$	s. d.	$\bar{x}$	s. d.	$\bar{x}$	s. d.
A	42.0	4.8	7.8	0.9	13.7	2.9	53.6	8.2	37.2	7.1	0.9	1.4	8.3	3.6
B	44.9	4.7	7.7	0.8	13.7	2.3	61.2	8.8	31.8	8.5	0.9	1.9	6.1	3.2
C	41.1	3.5	7.7	0.8	12.9	3.1	60.8	7.0	31.5	7.6	0.9	1.4	6.8	4.4
D	41.5	5.7	7.1	0.7	13.9	3.3	54.5	8.0	37.3	7.2	0.3	0.6	7.9	5.8

PCV Packed Cell Volume (Hematocrit) in %.

TP Total Protein in grams per 100 milliliters.

WBC White Blood Cell Count in thousands per cubic millimeter.

TN Total Neutrophils - expressed in % of total WBC.

L Lymphocytes - expressed in % of total WBC.

M Monocytes - expressed in % of total WBC.

E Eosinophils - expressed in % of total WBC.

Pure culture of the S. aureus was grown on a blood agar plate. Using a sterile bacteriological loop, an isolated 24-hour colony of the organism was inoculated into sterile tryptose broth (hereafter referred to as broth) in a screw-capped test tube. This was incubated at 37°C for 24 hours, the cap being applied loosely to allow for aerobic growth of the organism. In this way a heavy growth of the organism was obtained.

A spectrophotometer<sup>7</sup> was used to standardize the percentage light transmittance and hence the concentration of the cultures to be used for animal inoculation. The spectrophotometer was zeroed (100% light transmittance) with a clean cuvette containing sterile broth. Reading were taken at a wavelength of 500 Angstrom Units (A°), which was kept constant throughout the experiment.

Using aseptic technique, 5 milliliters of the concentrated culture was taken from the test tube with a pipette and transferred into a sterile cuvette. The cuvette containing the broth was removed from the spectrophotometer and replaced by the cuvette containing the culture. The percentage light transmittance was read from the scale. In earlier trials, it had been established that the density of the culture giving discrete individual abscesses in dogs corresponded to 29 percent light transmittance. Thus the spectrophotometer reading of 29 percent was arbitrarily chosen for the density of the

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<sup>7</sup>Spectronic 20, Bosch and Lomb, Rochester, New York.



least diluted culture solution to be used for animal inoculation. The cuvette containing the concentrated culture was carefully diluted with sterile broth from a pipette until a scale reading of 29 percent was obtained. The tube was constantly shaken using a whirl mixer<sup>8</sup> during the dilution process, in order to obtain a homogeneous suspension and thus record an accurate reading. The resulting culture was designated as 1:1 dilution.

Six test tubes with screw caps, each containing 4.5 milliliters of sterile broth were placed in a test-tube rack. Using aseptic technique, 0.5 milliliter of the 1:1 dilution was pipetted into the first tube containing 4.5 milliliters of broth. This resulted in a ten-fold dilution of the standardized 1:1 dilution and was then designated 1:10 dilution. This suspension was shaken on the whirl mixer and, using aseptic technique, 0.5 milliliter from this tube was pipetted into the second tube containing 4.5 milliliters of broth. This resulted in a further ten-fold dilution of the culture, or a hundred-fold dilution of the standard 1:1 dilution. The hundred-fold dilution was designated 1:100 ( $1:10^2$ ) dilution. Using this technique serial ten-fold dilutions were continued until a dilution of  $1:10^6$  was obtained.

The 1:10 dilution was transferred into a sterile cuvette, the tube placed in the spectrophotometer and a reading obtained. This procedure was repeated with the 1:100 dilution. The

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<sup>8</sup> Whirlmixer, Fison's Scientific Apparatus, Loughborough, Leicestershire, England.

spectrophotometer was constantly zeroed with broth in a clean cuvette, making sure that the wavelength remained at 500 A°. The readings obtained for 1:10 and 1:100 were found to be 82 percent  $\pm$  2 light transmittance and 96 percent  $\pm$  2 light transmittance respectively. After obtaining the readings the cultures were transferred back to their respective test tubes.

The 1:10<sup>6</sup> dilution was shaken and 0.1 milliliter of this suspension was pipetted into a sterile blood agar plate. A right-angled glass rod was dipped in 70 percent alcohol and flamed. Using the glass rod, the culture dropped on the blood agar plate was spread over the entire surface of the agar. The plate was incubated for 24 hours to verify bacterial numbers by a colony count.

The above procedure was repeated for each set of dogs to be operated on in a day, starting with a fresh S. aureus culture on a blood agar plate.

Bacterial colony counts were obtained for each set of dilutions using 0.1 milliliter of 1:10<sup>6</sup> dilution as described. A 24-hour plate gave discrete individual colonies that could be accurately counted. The counts were consistent throughout the course of the experiment. The average colony count obtained on the plates was 74 (range 68-82 colonies). It was thus estimated that the 1:1, 1:10 and 1:100 dilution of the cultures contained  $74 \times 10^7$  organisms/milliliter,  $74 \times 10^6$  organisms/milliliter and  $74 \times 10^5$  organisms/milliliter respectively (Table 3).

Table 3. Standardized Dilutions of Staphylococcus aureus Culture.

Dilution	Percentage Light Transmittance*	Organisms/Milliliter
1:1	29	$(74 \pm 8) \times 10^7$
1:10	$82 \pm 2$	$(74 \pm 8) \times 10^6$
1:100	$96 \pm 2$	$(74 \pm 8) \times 10^5$

\* Spectrophotometer readings obtained at a wavelength of 500 A°.

Periodically throughout the experiment coagulase and mannitol salt fermentation tests were performed to make sure that the organism retained its original properties.

### Suture Materials

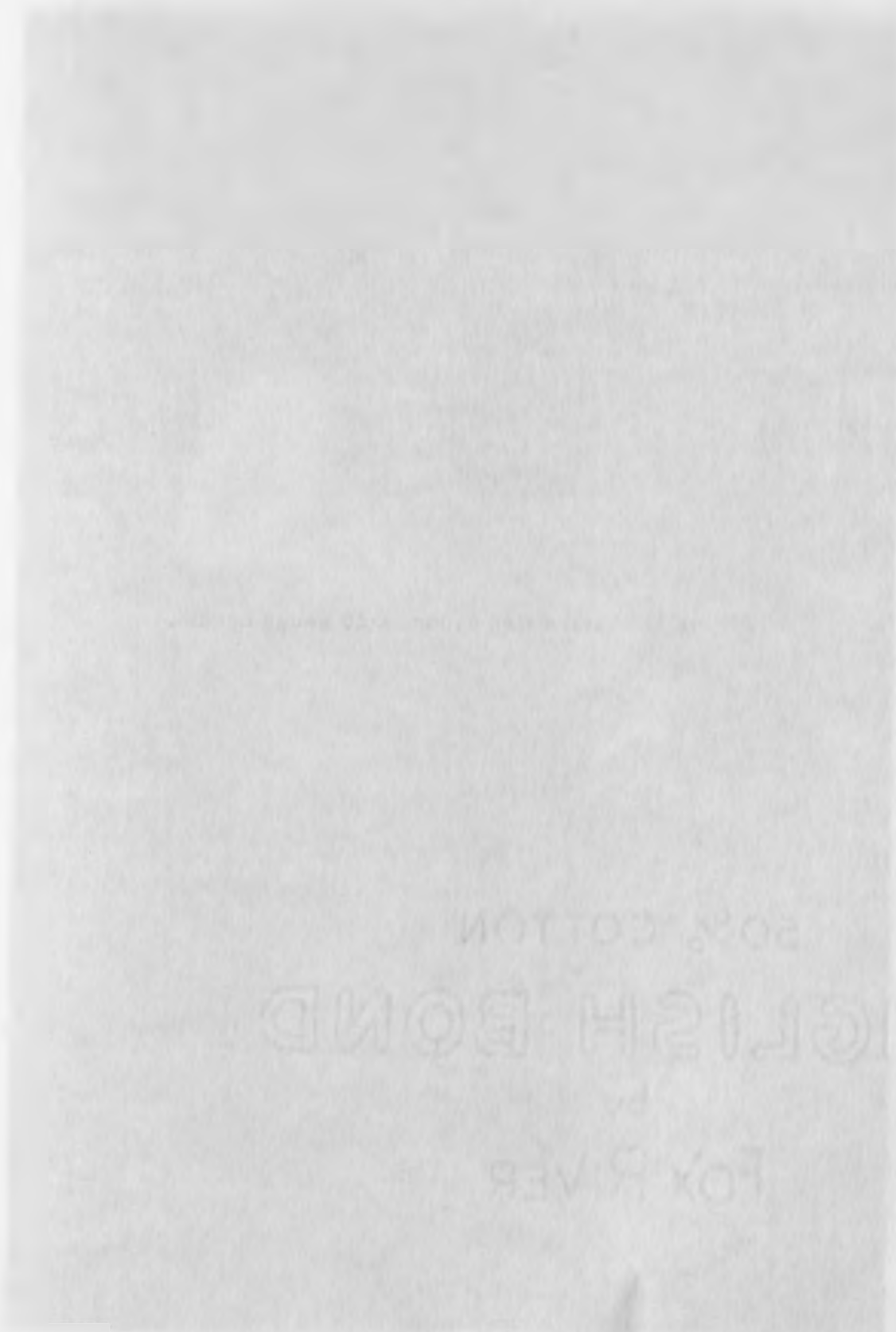
Seven different suture materials were studied in this project. They were braided polyglycolic acid (Dexon),<sup>9</sup> multifilament stainless steel, monofilament nylon, black braided silk, braided dacron (Mersilene), plain surgical catgut and chromic surgical catgut.<sup>10</sup> All suture materials were of size 3/0. The standard suture to be implanted consisted of two square knots tied around the shank of a 1 inch, 20 gauge (outside diameter 0.038 inch or 1 millimeter) disposable hypodermic needle<sup>11</sup> (Figure 1). All knots were hand tied just prior to surgery. The excessive suture ends were cut, leaving 3-millimeter free ends on the knot. The knot was slipped off the needle and placed in a sterile container.

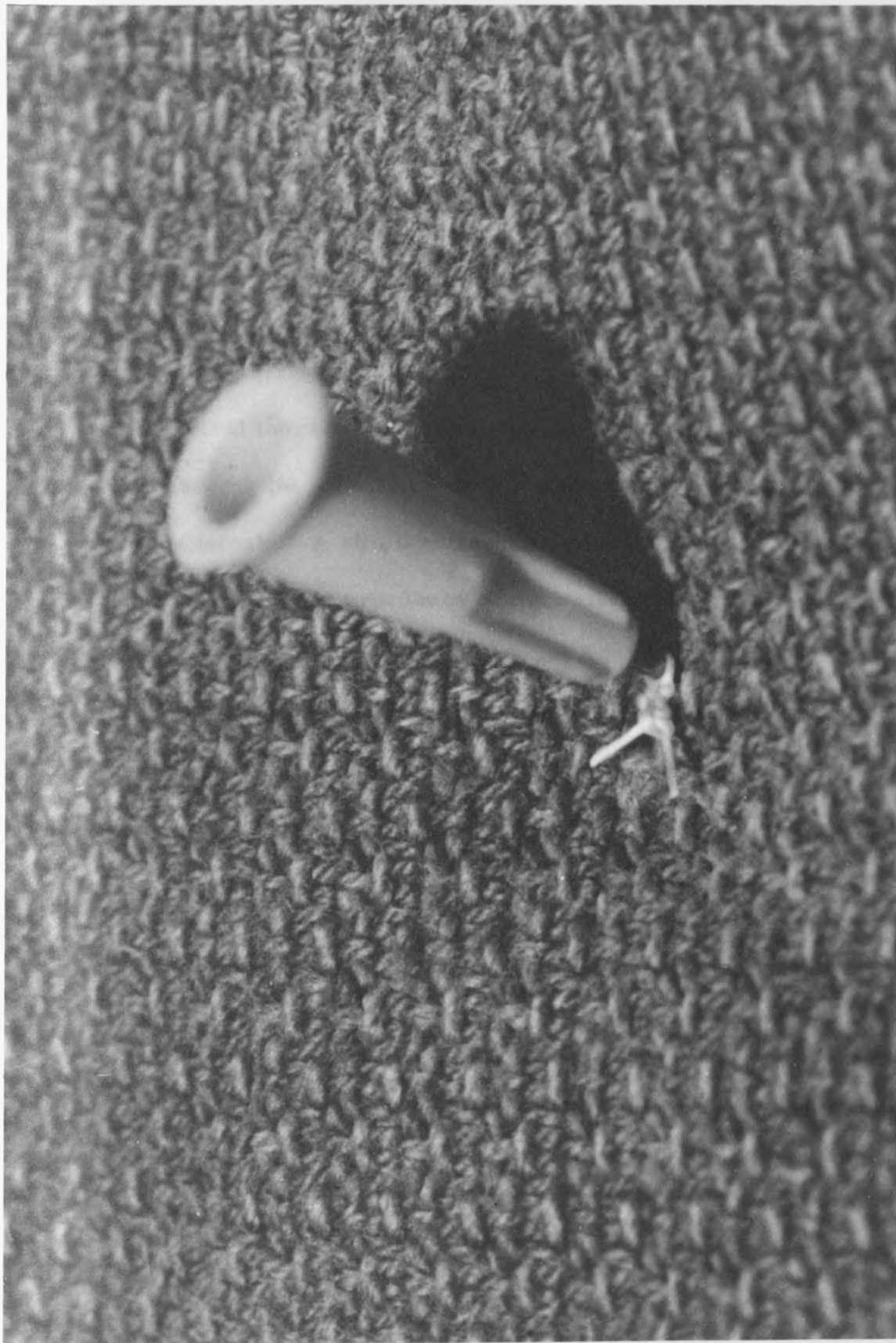
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<sup>9</sup>Dexon, Davis & Geck, American Cyanamid Co., New York, New York.

<sup>10</sup>Ethicon<sup>R</sup> Inc., Somerville, N. J.

<sup>11</sup>Monoject, Sherwood Medical Industries Inc., Deland, Fla.





### Implantation of Suture Materials and Inoculation of Suture Pockets

Each dog was given a physical examination prior to induction of anesthesia. Atropine sulfate<sup>12</sup> was given subcutaneously at the dosage rate of 0.04 mg/kg of body weight, as a preanesthetic agent. Anesthesia was induced and maintained with sodium pentobarbital<sup>13</sup> given intravenously. An endotracheal catheter was inserted to maintain a patent airway.

The ventral thoraco-abdominal region was clipped from the sixth sternebra to the preputial opening in the male and to the pubic region in the female. A similar area was clipped on the back, just lateral to the transverse processes of the vertebrae on one side extending from the seventh thoracic to the sixth lumbar vertebra. As a means of landmarking for future reference, nine transverse superficial scratch marks, 5 centimeters long and 4 centimeters apart, were made on the ventral midline with a one and one-half inch, 19 gauge hypodermic needle.<sup>14</sup> They were then painted with India ink. Similar marks 2.5 centimeters long were made on the clipped region on the back. This provided sufficient space to implant two identical

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<sup>12</sup>1% injectable atropine sulfate, E. T. Monks & Co. Ltd., Nairobi, Kenya.

<sup>13</sup>Sagatal, 6% Pentobarbitone Sodium Solution, May & Baker Ltd., Dagenham, England.

<sup>14</sup>Terumo, Jintan Terumo Co. Ltd., Tokyo, Japan.

rows of sutures on the ventral region and one on the back. The excess ink was removed with 70 percent alcohol sponges.

The surgical sites received standard surgical preparation with a povidone-iodine surgical scrub<sup>15</sup> and 0.5 percent chlorhexidine<sup>16</sup> in 70 percent alcohol. Preparation was completed on the back first. The area was covered with sterile towels and the dog placed in dorsal recumbency. Preparation was then completed on the ventral region and covered with a sterile towel.

Just prior to surgery the ventral surgical site was given a final preparation with 70 percent alcohol and the area allowed to dry. The area was then toweled and draped.

To make sure that all suture materials and the control pockets were randomly distributed over the area of implantation in the 15 dogs in each group, random sample tables were employed (Tables 4, 5, 6 and 7) (Ostle, 1963). All three rows in each dog had the suture materials placed in the same sequence. Two pockets in each row had no suture material, one containing only sterile tryptose broth and the other containing only culture suspension of the same dilution as the rest of the pockets in that row. The culture suspension acted as a control pocket and broth was inoculated to see if it elicited any tissue reaction. The three dilutions of culture were randomly assigned to

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<sup>15</sup> Disadine, Avlex Ltd., Wigan, Lancashire, England.

<sup>16</sup> Hibitane Gluconate, Imperial Chemical Industries Ltd., Cheshire, England.



Table 4. Random Distribution of Sutures in the 15 Dogs in Group A.\*

Dog Number	Pockets								
	1	2	3	4	5	6	7	8	9
2	Mersilene	Silk	Dexon	Plain Catgut	Broth	Steel	Nylon	Chromic Catgut	Culture
16	Chromic Catgut	Culture	Steel	Silk	Nylon	Plain Catgut	Mersilene	Broth	Dexon
29	Steel	Mersilene	Dexon	Nylon	Silk	Culture	Broth	Chromic Catgut	Plain Catgut
32	Plain Catgut	Broth	Steel	Nylon	Culture	Chromic Catgut	Mersilene	Dexon	Silk
27	Silk	Culture	Steel	Mersilene	Chromic Catgut	Broth	Dexon	Nylon	Plain Catgut
4	Dexon	Mersilene	Chromic Catgut	Silk	Broth	Nylon	Plain Catgut	Steel	Culture
26	Nylon	Broth	Culture	Chromic Catgut	Steel	Plain Catgut	Dexon	Mersilene	Silk
56	Broth	Dexon	Silk	Mersilene	Culture	Steel	Chromic Catgut	Nylon	Plain Catgut
63	Nylon	Chromic Catgut	Broth	Culture	Mersilene	Plain Catgut	Dexon	Silk	Steel
13	Chromic Catgut	Nylon	Dexon	Silk	Steel	Culture	Mersilene	Broth	Plain Catgut
62	Culture	Broth	Chromic Catgut	Mersilene	Silk	Steel	Plain Catgut	Nylon	Dexon
70	Culture	Steel	Silk	Broth	Plain Catgut	Mersilene	Dexon	Nylon	Chromic Catgut
78	Broth	Plain Catgut	Culture	Chromic Catgut	Dexon	Nylon	Mersilene	Silk	Steel
72	Culture	Steel	Nylon	Broth	Chromic Catgut	Dexon	Plain Catgut	Silk	Mersilene
88	Chromic Catgut	Broth	Culture	Nylon	Mersilene	Silk	Plain Catgut	Dexon	Steel

\*Suture materials were inserted in the same sequence in all three rows of pockets in each dog.

Table 5. Random Distribution of Sutures in the 15 Dogs in Group B.\*

Dog Number	Pockets								
	1	2	3	4	5	6	7	8	9
8	Plain Catgut	Silk	Dexon	Chromic Catgut	Broth	Steel	Nylon	Mersilene	Culture
18	Dexon	Steel	Nylon	Chromic Catgut	Broth	Silk	Plain Catgut	Mersilene	Culture
25	Plain Catgut	Nylon	Mersilene	Silk	Culture	Steel	Chromic Catgut	Broth	Dexon
31	Chromic Catgut	Dexon	Steel	Nylon	Plain Catgut	Culture	Silk	Broth	Mersilene
35	Mersilene	Plain Catgut	Silk	Culture	Steel	Nylon	Chromic Catgut	Broth	Dexon
42	Broth	Culture	Mersilene	Steel	Nylon	Plain Catgut	Silk	Dexon	Chromic Catgut
40	Chromic Catgut	Broth	Nylon	Silk	Mersilene	Dexon	Culture	Plain Catgut	Steel
44	Dexon	Silk	Mersilene	Culture	Chromic Catgut	Broth	Plain Catgut	Nylon	Steel
33	Dexon	Broth	Steel	Silk	Chromic Catgut	Culture	Mersilene	Nylon	Plain Catgut
64	Silk	Chromic Catgut	Culture	Mersilene	Plain Catgut	Dexon	Steel	Nylon	Broth
11	Plain Catgut	Silk	Broth	Steel	Dexon	Culture	Nylon	Chromic Catgut	Mersilene
5	Dexon	Chromic Catgut	Culture	Mersilene	Plain Catgut	Nylon	Silk	Steel	Broth
68	Dexon	Silk	Plain Catgut	Mersilene	Broth	Steel	Culture	Nylon	Chromic Catgut
51	Broth	Chromic Catgut	Plain Catgut	Dexon	Culture	Silk	Mersilene	Steel	Nylon
76	Mersilene	Nylon	Steel	Culture	Chromic Catgut	Dexon	Plain Catgut	Broth	Silk

\*Suture materials were inserted in the same sequence in all three rows of pockets in each dog.

Table 6. Random Distribution of Sutures in the 15 Dogs in Group C. \*

Dog Number	Pockets								
	1	2	3	4	5	6	7	8	9
12	Chromic Catgut	Silk	Plain Catgut	Broth	Dexon	Nylon	Culture	Mersilene	Steel
20	Plain Catgut	Mersilene	Steel	Culture	Silk	Nylon	Chromic Catgut	Broth	Dexon
6	Steel	Plain Catgut	Silk	Culture	Dexon	Broth	Chromic Catgut	Mersilene	Nylon
38	Plain Catgut	Broth	Silk	Chromic Catgut	Culture	Mersilene	Nylon	Steel	Dexon
34	Steel	Culture	Nylon	Dexon	Plain Catgut	Chromic Catgut	Broth	Mersilene	Silk
47	Silk	Plain Catgut	Nylon	Mersilene	Broth	Chromic Catgut	Dexon	Culture	Steel
53	Plain Catgut	Nylon	Mersilene	Chromic Catgut	Culture	Silk	Broth	Steel	Dexon
36	Mersilene	Plain Catgut	Silk	Steel	Nylon	Chromic Catgut	Broth	Dexon	Culture
54	Culture	Plain Catgut	Nylon	Steel	Dexon	Broth	Mersilene	Silk	Chromic Catgut
65	Steel	Nylon	Broth	Chromic Catgut	Plain Catgut	Dexon	Mersilene	Silk	Culture
59	Chromic Catgut	Silk	Steel	Nylon	Mersilene	Culture	Broth	Dexon	Plain Catgut
69	Mersilene	Plain Catgut	Chromic Catgut	Culture	Broth	Steel	Silk	Nylon	Dexon
74	Plain Catgut	Chromic Catgut	Culture	Broth	Dexon	Steel	Silk	Nylon	Mersilene
71	Plain Catgut	Nylon	Steel	Dexon	Chromic Catgut	Silk	Broth	Mersilene	Culture
81	Chromic Catgut	Plain Catgut	Culture	Silk	Steel	Mersilene	Broth	Dexon	Nylon

\* Suture materials were inserted in the same sequence in all three rows of pockets in each dog.

Table 7. Random Distribution of Sutures in the 15 Dogs in Group D. \*

Dog Number	Pockets								
	1	2	3	4	5	6	7	8	9
10	Broth	Dexon	Plain Catgut	Silk	Nylon	Mersilene	Steel	Chromic Catgut	Culture
17	Plain Catgut	Mersilene	Steel	Silk	Nylon	Broth	Chromic Catgut	Culture	Dexon
15	Broth	Nylon	Plain Catgut	Culture	Mersilene	Dexon	Steel	Chromic Catgut	Silk
23	Steel	Dexon	Silk	Plain Catgut	Mersilene	Chromic Catgut	Nylon	Culture	Broth
14	Silk	Plain Catgut	Culture	Nylon	Chromic Catgut	Mersilene	Broth	Steel	Dexon
52	Dexon	Culture	Steel	Plain Catgut	Broth	Chromic Catgut	Nylon	Silk	Mersilene
55	Culture	Plain Catgut	Nylon	Steel	Dexon	Broth	Mersilene	Silk	Chromic Catgut
37	Mersilene	Dexon	Plain Catgut	Chromic Catgut	Culture	Silk	Nylon	Broth	Steel
41	Broth	Dexon	Culture	Silk	Steel	Nylon	Plain Catgut	Mersilene	Chromic Catgut
61	Broth	Steel	Chromic Catgut	Mersilene	Silk	Nylon	Dexon	Plain Catgut	Culture
66	Steel	Mersilene	Culture	Broth	Silk	Chromic Catgut	Dexon	Nylon	Plain Catgut
67	Steel	Broth	Mersilene	Silk	Culture	Chromic Catgut	Dexon	Nylon	Plain Catgut
77	Steel	Silk	Nylon	Plain Catgut	Culture	Dexon	Broth	Chromic Catgut	Mersilene
82	Dexon	Broth	Nylon	Silk	Steel	Plain Catgut	Culture	Mersilene	Chromic Catgut
83	Plain Catgut	Nylon	Mersilene	Dexon	Silk	Culture	Broth	Chromic Catgut	Steel

\* Suture materials were inserted in the same sequence in all three rows of pockets in each dog.

the three rows in each dog. Thus the row of suture pockets on the back did not receive the same dilution of culture in every dog, eliminating differences in tissue reaction between dilutions due to specific inoculation sites for each dilution.

Using the tip of a number 11 surgical blade,<sup>17</sup> a stab incision in the skin was made at the end of each India ink mark, 2.5 centimeters from midline, on one side of the ventral surgical area (Figure 2). The marks were made in such a way that the incision and the subcutaneous tissue pockets to be created did not involve any mammary tissue. A knot, corresponding to the particular pocket, was placed in the hub of a 12 gauge, 9 centimeters long hypodermic needle<sup>18</sup> with a fine pair of tissue forceps. The tip of the needle was placed in the stab incision (Figure 3), directed laterally, and inserted subcutaneously for about 3 centimeters (Figure 4). The beveled tip of the needle could be felt under the skin. Throughout the procedure, care was taken not to transect or puncture any blood vessels in the area, thus minimizing hemorrhage. Occasionally, however, skin bleeders were encountered which were controlled by pressure application. A one-half inch (1.3 centimeters), 27 gauge disposable needle,<sup>19</sup> through which the pocket was to be inoculated, was inserted

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<sup>17</sup>Paragon, Paragon Razor Company, Sheffield, England.

<sup>18</sup>Berbert Mile-Hi, George Berbert & Sons, Denver, Colo.

<sup>19</sup>Old East Africa Ltd., Nairobi, Kenya.



Figure 2. Stab incision in the skin.

Figure 3. Introduction of the 12 gauge needle into the stab incision.

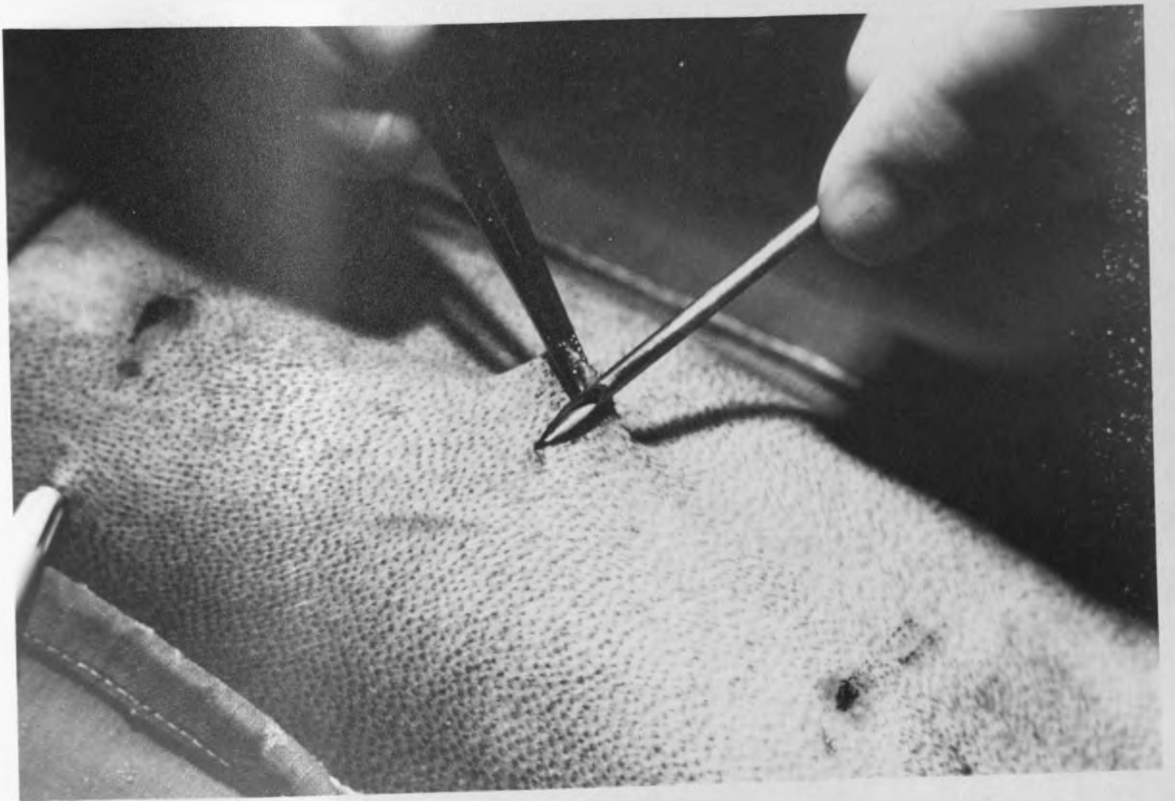
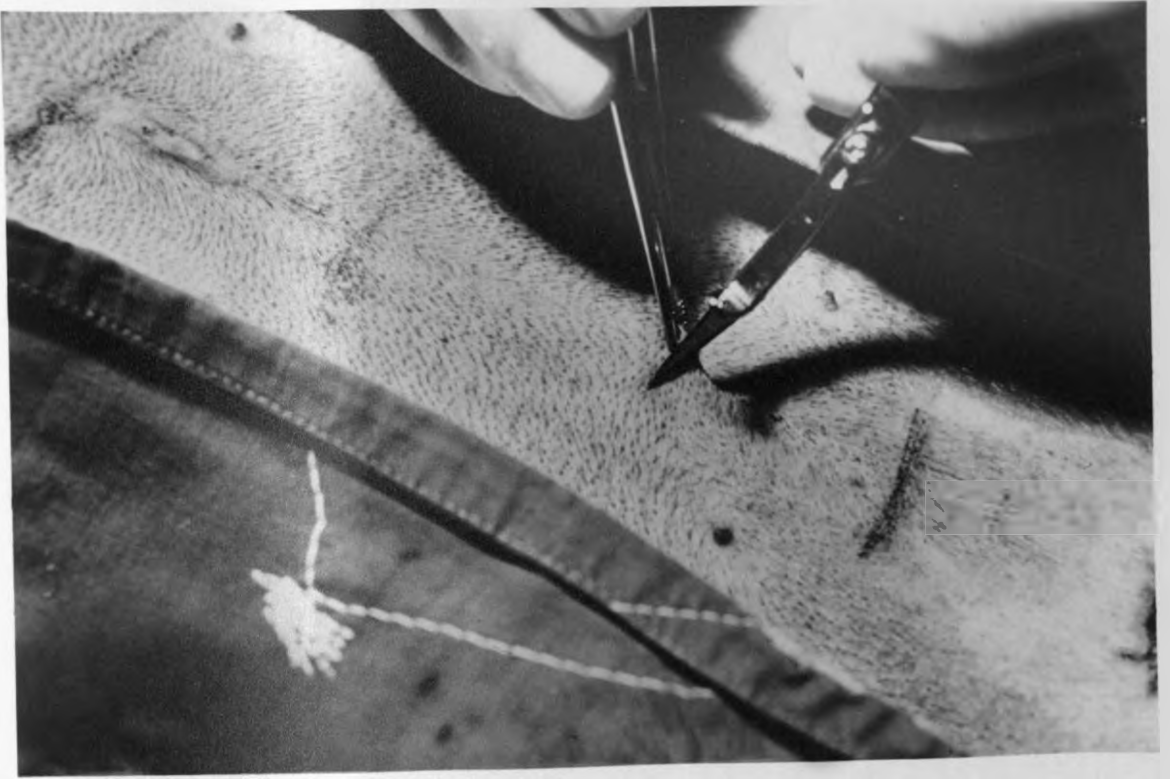
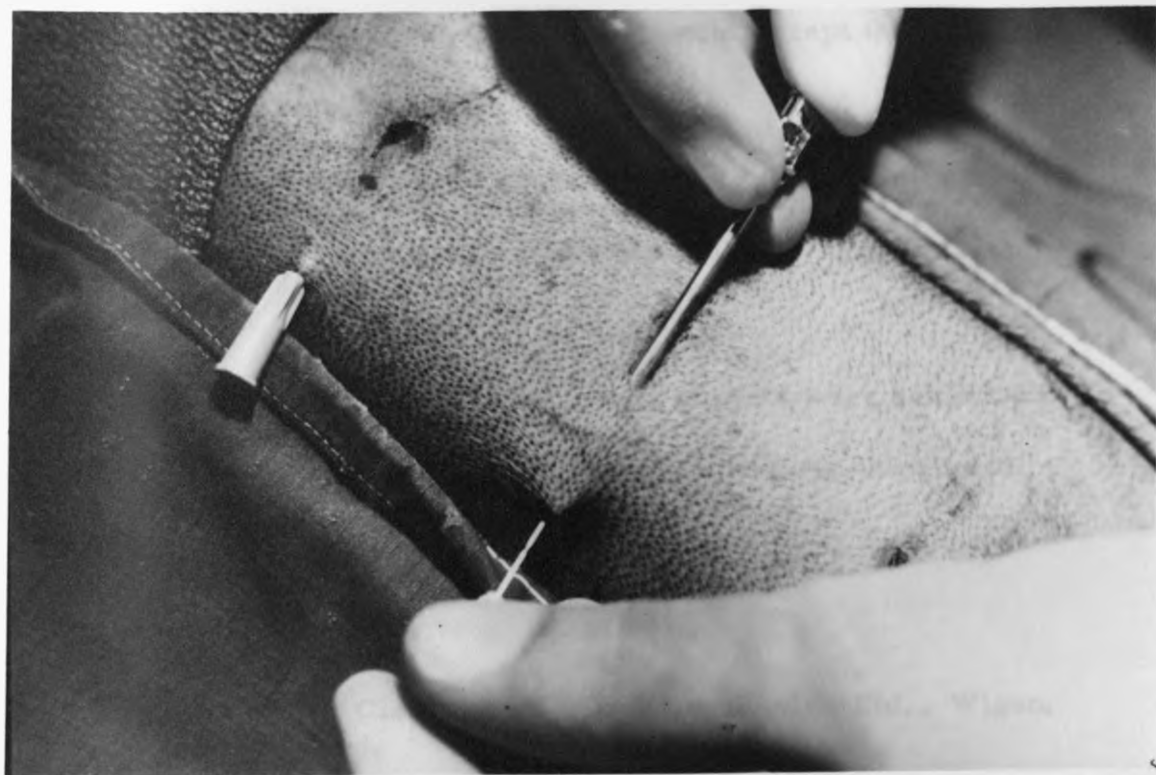
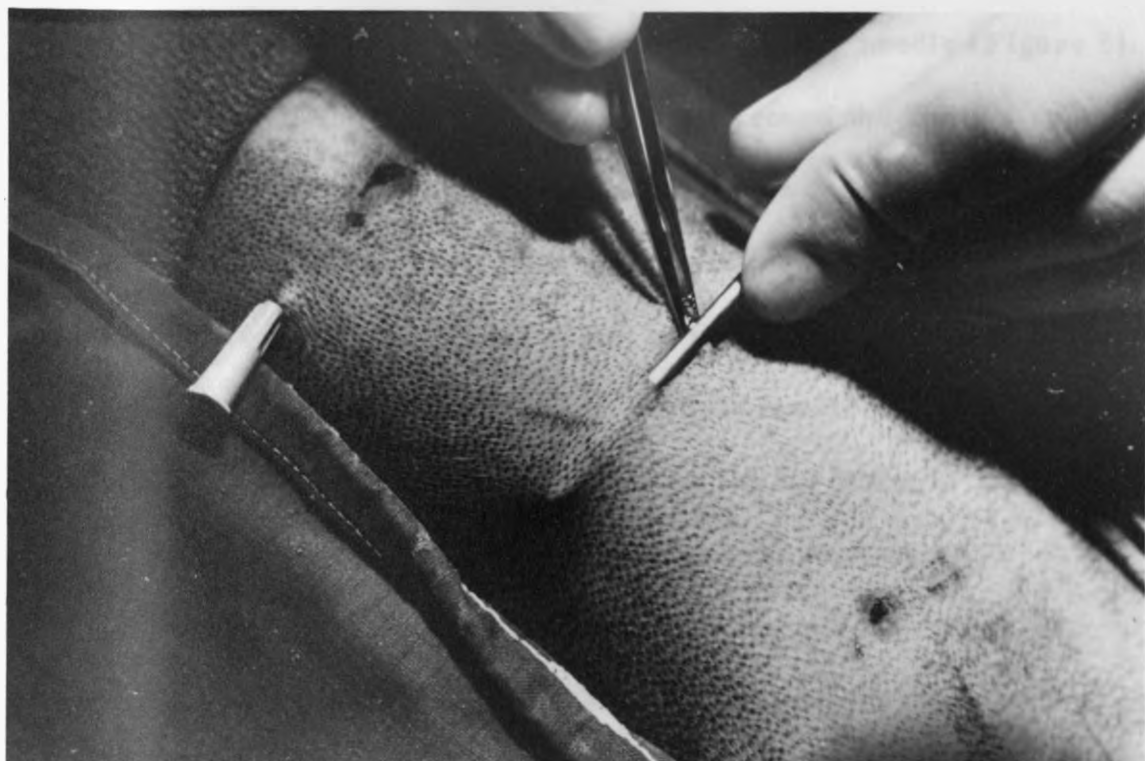




Figure 4. The 12 gauge needle inserted subcutaneously.

Figure 5. Insertion of a 27 gauge needle into the bevel tip of the 12 gauge needle.



through the skin into the beveled tip of the 12 gauge needle (Figure 5). To make sure that the inoculum was placed around the knot, a special stylet that fit into the 12 gauge needle was made from a Steinmann pin. A fine hole 3 millimeters deep was drilled longitudinally into one end of the stylet. The drilled end of the stylet was placed in the hub of the 12 gauge needle and the knot in the needle was inserted until the drilled end of the stylet rested on the tip of the 27 gauge needle (Figures 6 and 7). This ensured that the knot was placed directly over the 27 gauge needle. Leaving the stylet in place, the 12 gauge needle was withdrawn (Figure 8) and then the stylet was removed. The 27 gauge needle was left in position (Figure 9). This was repeated for all of the nine incisions in the row using the corresponding suture knot for each pocket. The pockets to contain only broth or culture received similar treatment except that no knot was placed in the 12 gauge needle.

The same procedure was repeated on the other side of the ventral midline, using a different sterile blade, 12 gauge needle and stylet.

The openings of the subcutaneous pockets were sealed with an adhesive spray of resin in ethyl acetate containing chlorhexidine hydrochloride<sup>20</sup> (Figure 10).

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<sup>20</sup>Hibispray, Clear Plastic Dressing, Avlex Ltd., Wigan, Lancashire, England.



Figure 6. Instruments used for insertion of suture and inoculation of culture.

Stylet (1), 12 gauge needle (2), and 27 gauge needle (3).

Figure 7. Stylet, 12 gauge needle and 27 gauge needle in position in a dog.

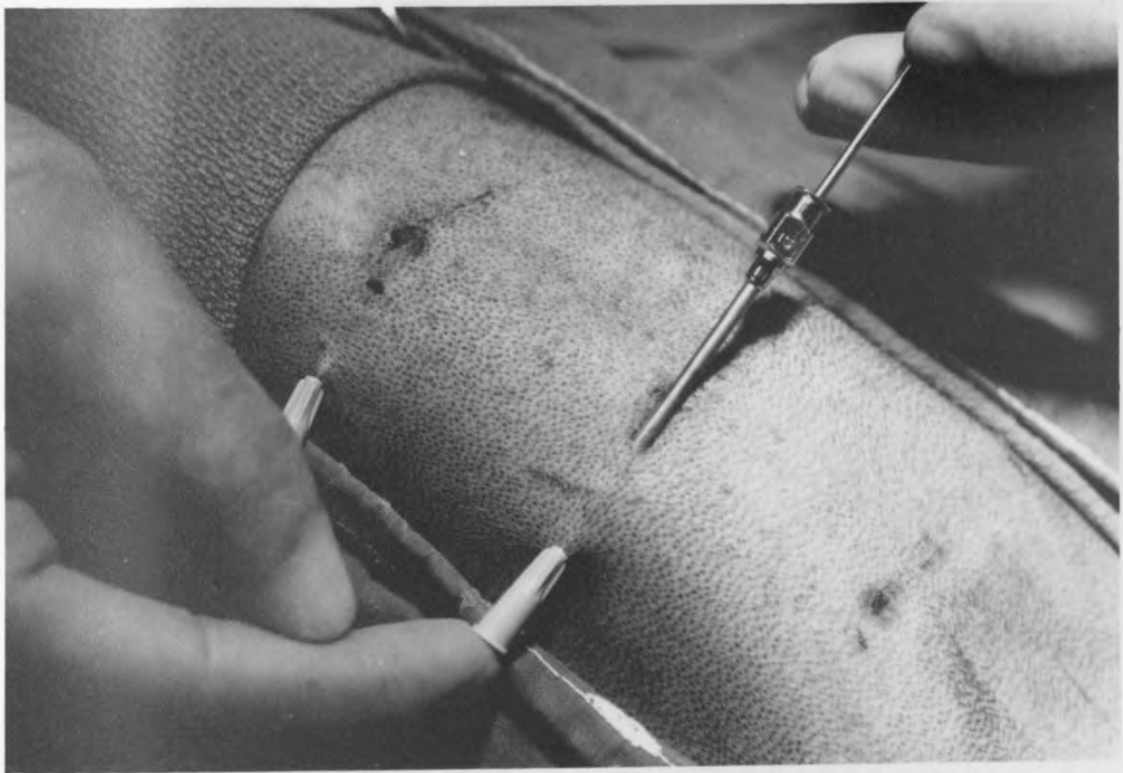
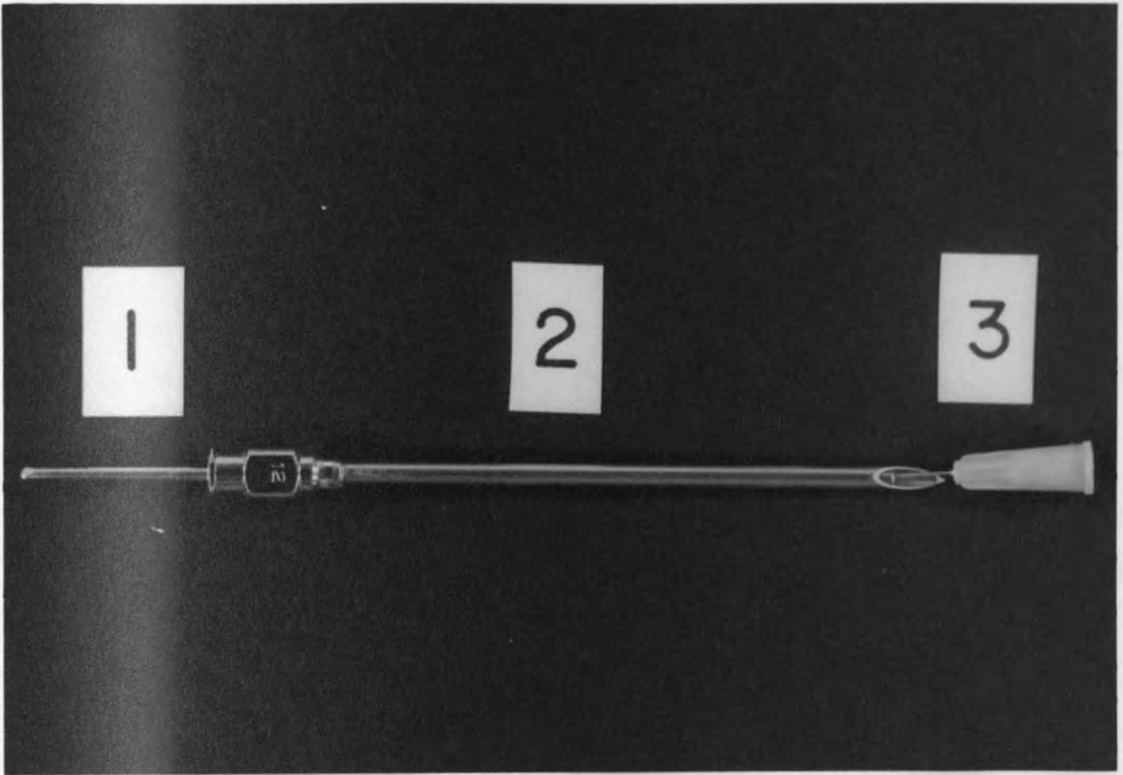


Figure 8. The 12 gauge needle withdrawn, stylet still in place.

Figure 9. Stylet removed, leaving the 27 gauge needle in position.

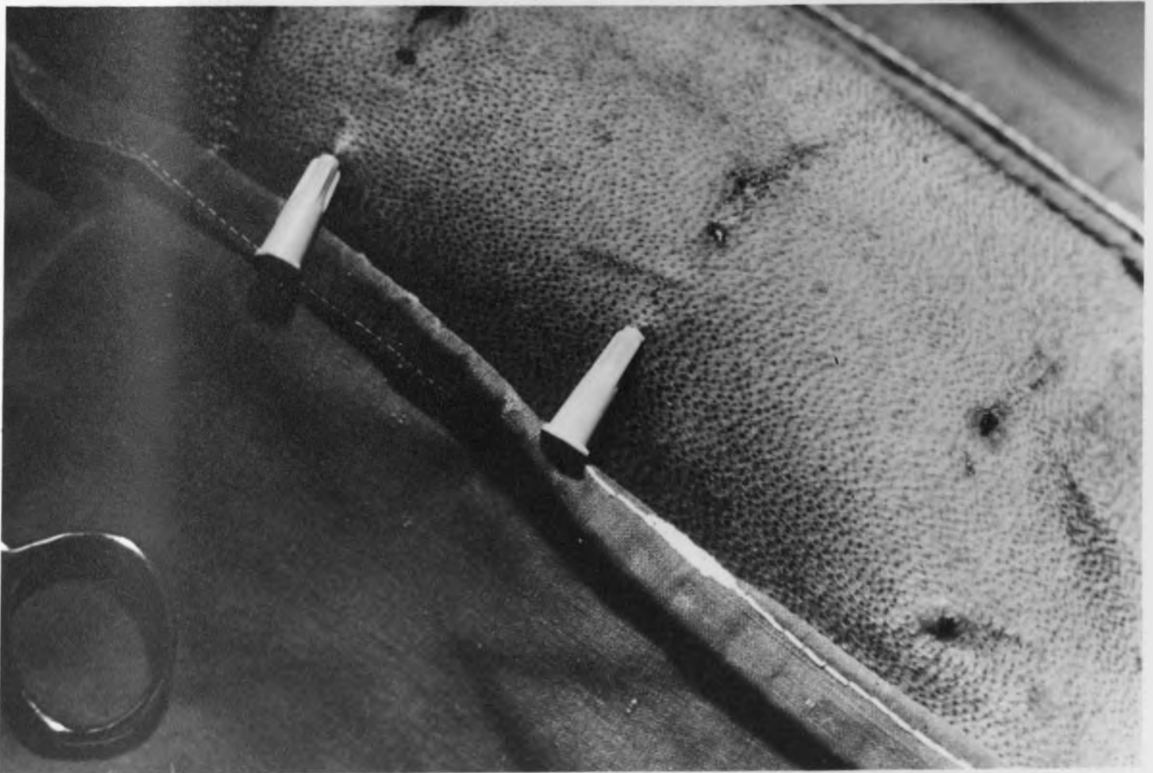
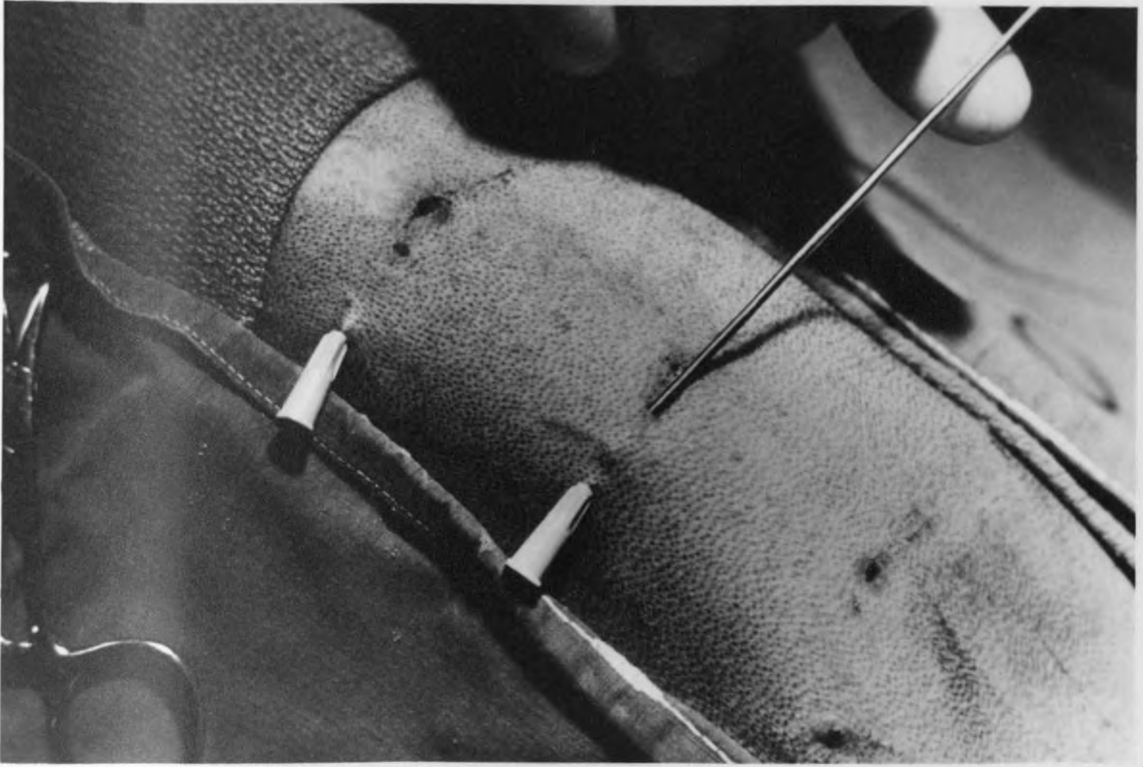
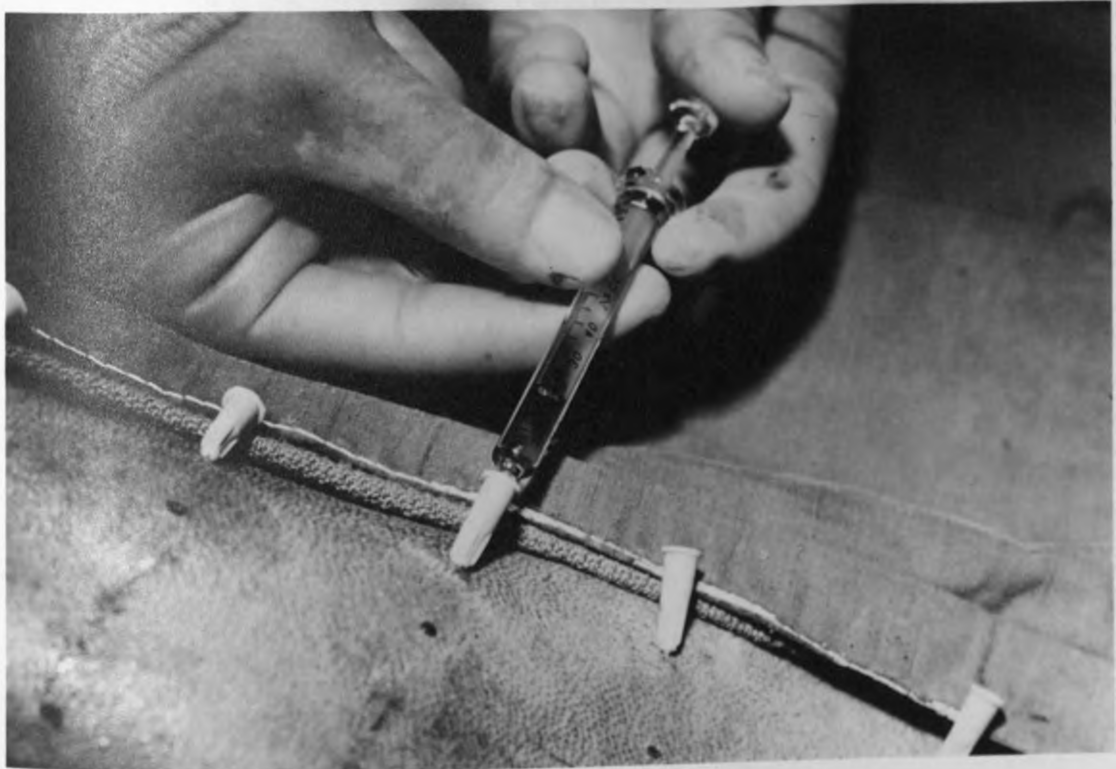
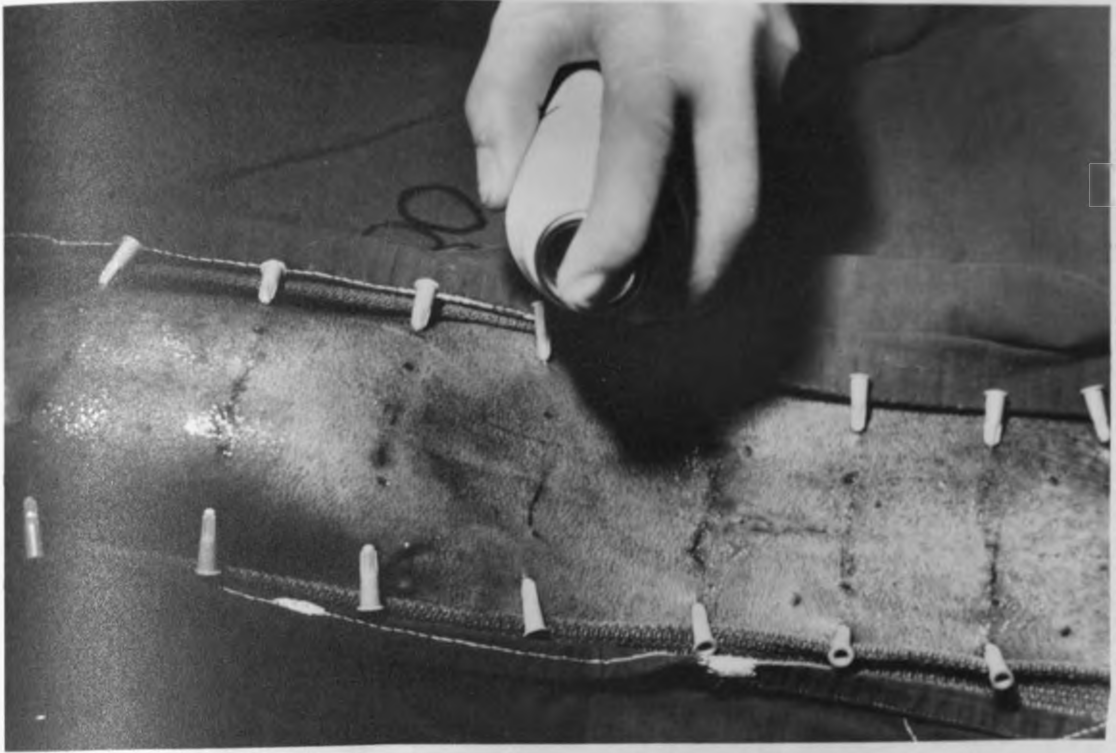




Figure 10. All 27 gauge needles in position and the openings of the pockets being sprayed with the resin adhesive.

Figure 11. Culture injected over the suture knot through the 27 gauge needle.



Three sterile 1-milliliter tuberculin-type disposable syringes<sup>21</sup> and three sterile 16 gauge, 12 centimeters long hypodermic needles,<sup>22</sup> were taken from the instrument tray and placed in a sterile pan. The sterile instrument tray with the third set of knots was removed from the operating table. A second pair of sterile gloves was worn.

The sterile tube of broth was held by the circulating assistant, the tube uncapped, and the neck of the tube flamed. Using a 1-milliliter syringe and a 16 gauge needle, 0.2 milliliter of broth was carefully withdrawn from the tube. The needle was discarded and 0.1 milliliter of broth was injected into the 27 gauge needles corresponding to each broth pocket on both rows. The 27 gauge needles in these two pockets were rotated and withdrawn. The rotation ensured that there was no leakage of broth as the needle was withdrawn. The appropriate dilution of culture to be inoculated in the first row of pockets was shaken to get a homogeneous suspension. The neck of the tube was flamed and, using a different sterile 1-milliliter syringe and 16 gauge needle, the culture was withdrawn into the syringe leaving the needle in the tube. One-tenth milliliter of the culture in the syringe was injected into each of the 27 gauge needles in the proper row (Figure 11). Each needle was rotated and withdrawn immediately after injection. The process was repeated until all eight

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<sup>21</sup>Terumo, Jintan Terumo Co. Ltd., Tokyo, Japan.

<sup>22</sup>Berbert Mile-Hi, George Berbert & Sons, Denver, Colo.

pockets to receive culture were inoculated. The whole procedure was repeated with the appropriate culture for the second row using a different sterile needle and syringe. In some pockets the adhesive seal was not adequate and leakage occurred. These were noted, as this rendered the data from these pockets invalid. When all pockets had been injected, a second layer of resin adhesive was sprayed over the entire area. The drapes and towels were discarded and a sterile towel was placed on the ventral region.

The dog was now placed in dorsolateral recumbency to facilitate working on the back area. The sterile towel was removed and the surgical area was prepared again with 70 percent alcohol and the skin allowed to dry. Gown and gloves were changed for this second site. The area was then toweled and draped.

The entire procedure was repeated on the back. One pocket was injected with broth and the others with the appropriate dilution of culture.

#### Postoperative Management

All dogs were housed individually or in groups of two or three depending on the number of dogs in postoperative phase at any given time. Food was provided daily and water ad libitum until the time of sacrifice. No postoperative treatment was administered.

Rectal temperatures were taken daily and all three suture rows examined for development of lesions and for any drainage. Draining abscesses were noted.

#### Necropsy Procedures and Evaluation of Lesions

The dogs were euthanatized on day 6, 10, 20 or 40 postimplantation depending on the group to which they belonged. A concentrated solution of sodium pentobarbital<sup>23</sup> was given intravenously for euthanasia. All sites of inoculation were examined and all draining abscesses noted.

An abscess in the 1:1 dilution row was chosen at random in each of the dogs in groups A and B. The skin over the abscess was swabbed with alcohol and, using a sterile 20 gauge needle and a syringe, some of the pus from this abscess was aspirated. Two drops of the aspirate were spread on a blood agar plate with a bacteriological loop and the rest of the aspirate was inoculated into a test tube of tryptose broth. The plate and the test tube were incubated at 37°C for 24 hours. The plate was then examined for bacterial growth. The culture was used to inoculate a fresh blood agar plate which was further incubated for 24 hours and examined for bacterial growth. This was done to recover the S. aureus organisms and to determine possible contamination. This was not possible with

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<sup>23</sup>Euthatal, 20% Pentobarbitone Sodium Solution, May & Baker, Dagenham, England.

dogs in groups C and D as frank pus was not obtained from any of the inoculation sites.

Measurement of lesions (abscesses or suture granulomas) from the skin surface was not found satisfactory in any of the four groups. The lesions were, therefore, measured subcutaneously. A skin incision was made on the ventral or dorsal midline and the skin reflected laterally on one side by subcutaneous dissection. Care was taken not to incise any abscess. Measurement of lesions was facilitated by this method. Lesions were measured along their longest axes (diameters) with a pair of vernier calipers<sup>24</sup> to the nearest 0.05 centimeter. Lesions not grossly visible or palpable were assigned a measurement of zero. Nodules were not incised as these were fixed for histopathology.

Specimens for representative histopathologic evaluation were taken from four dogs in each group. These dogs were randomly selected, making sure that they were evenly distributed in the course of the experiment. After measuring the lesions grossly, each row of lesions was removed en bloc from the dog in one skin strip, 4 centimeters wide. Each strip was sutured onto a cardboard length and fixed in 10 percent buffered formalin solution for at least two weeks, after which time a section 2 millimeters thick was cut through the middle of each lesion. Steel sutures were removed from all sections

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<sup>24</sup>Mitutoyo Manufacturing Co. Ltd., Japan.

containing this material. The specimens were paraffin embedded, sectioned at 6 microns and stained with hematoxylin and eosin for microscopic examination. One section was made from each specimen. This provided four histologic specimens of tissue reaction around each suture material at each dilution of culture in all four groups.

To assess the tissue response to the implanted sutures, the weighted average method of Sewell et al. (1955) was used as a guideline. An ocular micrometer<sup>25</sup> (12.5X) was used to measure the diameter (greatest dimension) of the tissue reaction around the suture. A 2.5X objective lens was used, giving a magnification of 31.25X. The microscopic measurements were correlated to the gross measurements obtained at necropsy. Cellular infiltration was examined in greater detail. The number of different cell types (neutrophils, eosinophils, lymphocytes, plasma cells, macrophages, giant cells and fibroblasts) per oil immersion field (1125X) was approximated. Five such fields were examined, starting from the immediate vicinity of the suture material, moving towards the periphery of the reaction. A percentage differential cell count was thus obtained. In this scheme the width of the reaction zone and the cell types were used as criteria for subjective ratings of tissue

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<sup>25</sup>Leitz, Ernst Leitz GMBH, Wetzlar, Germany.

reaction. The degree of suture absorption at various periods of implantation was also assessed.

### Statistical Analyses

The recorded data of gross measurements of lesions were subjected to statistical analyses using program Stat 02V - Analysis of Variance for Factorial Designs of the Statistics Laboratory of Colorado State University.

For each of the groups A, B, C and D a one-way randomized blocks analysis of variance was first performed to compare the eight treatments (one control and seven suture materials) at each level of inoculation. The hypothesis used was that at any stage postimplantation the seven suture materials and the culture inoculation (control) elicited different tissue reactions at a given dilution of S. aureus.

On finding the preliminary analysis of variance test significant, the data were examined in greater detail, testing differences between suture materials at a given dilution. The procedure used was Tukey's studentized range test or range simultaneous test procedure (range-STP), using the critical value of a special statistic for significant testing. The least significant range for treatments,  $Q_t$ , was calculated for each period at each dilution (Sokal and Rohlf, 1969). Using the  $Q_t$  value for a given dilution, comparison could be made between any two suture materials.  $Q_t$  was the least range that had to exist between two means for them to be significantly different.



A two-way factorial (dilutions and treatments) in randomized blocks (dogs) analysis of variance was then performed to get an overall or global view of the eight treatments at all three dilutions at a given postimplantation period. This was done to test the main effects due to dilutions or treatments. On obtaining a differential response, the range-STP procedure was again used to investigate the particular sources of significance. The following aspects of the global view were studied in greater detail:

1. Differences between the three dilutions using  $Q_d$  as the least significant range
2. Overall differences between treatments (suture materials)
3. Treatment-dilution interactions, comparing treatment means (across all groups) at each level of dilution.

A three-way repeated measures analysis of variance was then performed on the pooled data of groups A, B, C and D across the three levels of inoculation and the eight treatments. This provided a global view of:

1. Differences between treatments, irrespective of the period of implantation or the dilution level
2. Differences between dilutions as averaged for all groups and treatments
3. Differences between groups, comparing overall effects of treatments and dilutions. The least significant range here was designated  $Q_g$

4. Two and three-way interaction effects of groups, treatments and dilutions.

RESULTS

The first experiment was designed to determine the effect of the temperature of the medium on the growth of the bacteria. The results are shown in Table I. It is seen that the growth of the bacteria was significantly greater at 37°C than at 22°C. The growth of the bacteria was also significantly greater at 37°C than at 15°C. The growth of the bacteria was not significantly different at 15°C and 22°C.

The second experiment was designed to determine the effect of the dilution of the medium on the growth of the bacteria. The results are shown in Table II. It is seen that the growth of the bacteria was significantly greater at a dilution of 1:10 than at a dilution of 1:100. The growth of the bacteria was also significantly greater at a dilution of 1:10 than at a dilution of 1:1000. The growth of the bacteria was not significantly different at a dilution of 1:100 and a dilution of 1:1000.

The third experiment was designed to determine the effect of the group on the growth of the bacteria. The results are shown in Table III. It is seen that the growth of the bacteria was significantly greater in group A than in group B. The growth of the bacteria was also significantly greater in group A than in group C. The growth of the bacteria was not significantly different in group B and group C.

## CHAPTER IV

### RESULTS

#### General Observations

The mean preoperative rectal temperature of the dogs was 101.2° F. All dogs had an elevation of temperature on the first, second or third postoperative day, ranging between 102.6° F and 104.8° F. Temperatures started subsiding generally on the third postoperative day and were between 99.5° F and 102° F for the rest of the survival period.

Discrete abscesses started appearing after the first postoperative day. Draining abscesses were noted between day 1 and day 10. A total of 22 abscesses drained in the 1:1 dilution rows, 6 in the 1:10 dilution rows and 2 in the 1:100 dilution rows (Table 8). Twenty of the 60 dogs had at least one draining abscess, the greatest number on one dog being three. Pockets containing Dexon drained most frequently (9 of 180).

Broth did not elicit any gross reaction in any pocket, showing that it played no role in formation of the lesions. S. aureus was obtained on culturing a random pus sample from each dog in groups A and B at the time of necropsy. Contaminating organisms were found in only three samples.

Table 8. Draining Abscesses for Each Treatment at Each Dilution of Culture Across the Four Groups (60 Dogs).

Treatments	Dilution of Culture		
	1:1* n (d)	1:10* n (d)	1:100* n (d)
Culture only	0	0	0
Steel	3 (3, 6, 7)	1 (6)	1 (8)
Nylon	2 (3, 3)	0	0
Silk	3 (3, 3, 9)	1 (4)	0
Mersilene	3 (4, 6, 7)	1 (4)	0
Dexon	6 (3, 3, 3, 3, 7, 7)	2 (1, 2)	1 (3)
Plain Catgut	3 (2, 4, 4)	1 (5)	0
Chromic Catgut	2 (7, 10)	0	0

\* 60 pockets injected for each treatment at each dilution.

n Number of draining abscesses.

(d) Postimplantation days when drainage occurred.

Gross Measurements of Lesions and Statistical Tests for Group A  
(6 Days)

Measurements of lesion diameters in centimeters in the 1:1, 1:10 and 1:100 dilution rows in each dog in group A are shown in Tables 9, 10 and 11. Respective mean values were used for the bleeding and leaking pockets in order to use the computer program.

Results for one-way randomized block analysis of variance for each dilution are shown in Table 12. F values were found significant, at the 1 percent level, for treatments at all three dilutions.

Using the range-STP test,  $Q_t$  values were calculated for each dilution. The critical value for the test was obtained from statistical tables (Rohlf and Sokal, 1969) and it was 4.389 at the 5 percent significance level.

Table 13 shows lesion means, in centimeters, with the corresponding  $Q_t$  values for all treatments at each dilution. The range-STP test provided simultaneous pairwise comparisons between all pairs of treatments. Any two means whose differences were less than the  $Q_t$  values were connected by a vertical bar. Thus, any pair of means enclosed by the range of any one bar were not significantly different.

A graphic representation of this is shown in Figure 12 A, B and C. The middle of each bar represents the treatment mean. The range is the  $Q_t$  value. Treatments whose simultaneous confidence intervals overlap are not statistically significant. All differences or

Table 9. Gross Measurements of Lesion Diameters in Centimeters in the 1:1 Dilution Rows of Group A (6 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
2	1.20	1.70	1.30	2.35	1.45	2.35	1.90	1.25
16	0.20	1.15	0.70	1.35	1.40	1.40	2.20	1.55
29	1.40	1.20	2.35	1.80	1.70	2.35	1.50	2.30
32	1.10	1.45	1.60	2.00	2.40	b	1.80	2.75
27	0.20	0.95	0.75	1.35	1.20	1.90	1.30	1.45
4	0.80	0.90	0.85	1.05	1.60	2.10	1.30	1.70
26	1.10	1.20	1.90	1.30	1.80	1.00	1.50	1.85
56	1.20	0.40	0.80	1.00	1.60	1.25	1.85	1.30
63	0.20	2.00	1.30	2.00	2.60	2.20	2.60	2.60
13	1.25	1.35	1.20	1.20	1.35	1.30	1.05	2.00
62	0.20	0.60	0.95	1.45	1.40	1.50	2.70	1.20
70	2.10	1.20	1.15	2.50	1.50	2.25	1.90	1.30
78	0.90	1.05	0.70	1.30	1.40	1.05	2.25	1.60
72	0.90	1.25	1.60	2.90	a	1.80	2.25	2.30
88	1.60	0.70	0.80	1.90	1.80	2.30	1.50	1.95

\* Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^6$  Staphylococcus aureus organisms.

<sup>a</sup>Leaking pocket.

<sup>b</sup>Bleeding pocket.

Table 10. Gross Measurements of Lesion Diameters in Centimeters in the 1:10 Dilution Rows of Group A (6 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
2	0.60	0.95	0.70	0.85	0.80	1.05	1.20	1.30
16	0.20	0.55	0.50	0.50	0.80	1.30	1.10	0.75
29	0.30	0.40	0.30	1.00	0.60	0.90	0.85	1.00
32	0.20	0.40	0.35	0.50	0.50	0.75	0.90	0.60
27	0.40	0.30	0.20	0.60	1.00	0.60	1.10	0.80
4	0.40	0.70	1.50	0.80	0.70	0.60	0.55	0.90
26	0.30	0.20	0.50	1.90	1.45	0.70	0.60	0.70
56	0.45	0.70	0.35	0.45	0.20	0.50	0.75	0.90
63	0.45	0.60	1.00	0.50	0.70	1.10	1.40	1.10
13	0.15	0.50	0.40	0.65	0.45	0.95	0.45	0.55
62	0.20	0.50	0.70	0.70	0.65	1.80	1.10	0.70
70	0.55	0.90	0.90	1.30	1.35	0.40	1.05	0.70
78	0.30	0.55	0.60	0.65	0.65	0.60	0.60	0.80
72	0.25	0.90	1.10	1.20	0.40	0.80	1.25	1.50
88	0.30	0.45	0.20	0.75	1.10	0.70	1.10	0.80

\*Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^5$  Staphylococcus aureus organisms.

Table 11. Gross Measurements of Lesion Diameters in Centimeters in the 1:100 Dilution Rows of Group A (6 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
2	0.10	0.60	0.85	0.80	0.95	0.75	1.15	0.55
16		0.30	0.55	0.75	0.55	0.65	0.50	0.55
29	0.20	0.30	0.30	0.70	0.60	0.70	0.55	0.80
32	0.15	b	0.45	0.60	0.50	0.70	0.90	0.50
27		0.20	0.45	0.40	0.30	0.40	0.95	0.80
4	0.30	0.40	0.70	0.65	0.50	0.60	0.70	0.75
26		0.20	0.25	0.40	0.40	0.55	0.55	0.75
56	0.20	0.70	0.90	0.45	0.55	0.60	0.90	1.00
63	0.40	0.20	0.30	0.60	0.40	0.50	0.50	0.60
13		0.30	0.30	a	0.40	0.75	0.50	0.55
62	0.50	0.35	0.60	0.55	0.70	0.60	0.80	0.65
70	0.50	0.40	0.35	0.85	0.50	0.80	0.60	0.55
78	0.40	0.40	0.40	0.45	0.45	0.60	0.50	0.60
72	0.15	0.50	0.40	0.55	0.50	0.70	0.55	0.65
88		0.45	0.50	0.70	0.60	0.75	0.85	0.70

\* Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^4$  Staphylococcus aureus organisms.

<sup>a</sup> Leaking pocket.

<sup>b</sup> Bleeding pocket.

Blanks indicate a measurement of zero.



Table 12. One-Way Randomized Block Analysis of Variance for Treatment Means at Each Level of Inoculation for Group A (6 Days).

	Source of Variation	Degrees of Freedom	Sums of Squares	Mean Squares	F
1:1 Dilution	Dogs	14	8.92512	0.63751	
	Treatments	7	12.90525	1.84361	10.031***
	Error	98	18.01287	0.18380	
	Total	119			
1:10 Dilution	Dogs	14	2.34167	0.16726	
	Treatments	7	4.14592	0.59227	7.061***
	Error	98	8.22033	0.08388	
	Total	119			
1:100 Dilution	Dogs	14	0.93467	0.06676	
	Treatments	7	3.06465	0.43781	15.509***
	Error	98	2.04067	0.02823	
	Total	119			

\*\*\* Significant (for  $\alpha = 0.01$ ).

Table 13. Treatment Means for Each Level of Inoculation with the Corresponding  $Q_t$  Values for Group A (6 Days).

Treatments	Means*	
		<u>1:1 Dilution</u>
Culture	0.95667	$Q_t = 4.389 \sqrt{\frac{0.18380}{15}}$ $= 0.486$
Steel	1.14000	
Nylon	1.19667	
Mersilene	1.65667	
Silk	1.69667	
Dexon	1.76667	
Chromic Catgut	1.80667	
Plain Catgut	1.84000	
		<u>1:10 Dilution</u>
Culture	0.33667	$Q_t = 4.389 \sqrt{\frac{0.08388}{15}}$ $= 0.328$
Steel	0.57333	
Nylon	0.62000	
Mersilene	0.75667	
Silk	0.82333	
Dexon	0.85000	
Chromic Catgut	0.87333	
Plain Catgut	0.93333	
		<u>1:100 Dilution</u>
Culture	0.19333	$Q_t = 4.389 \sqrt{\frac{0.02823}{15}}$ $= 0.190$
Steel	0.37667	
Nylon	0.48667	
Mersilene	0.52667	
Silk	0.60333	
Dexon	0.64333	
Chromic Catgut	0.66667	
Plain Catgut	0.70000	

\* Lesion measurements in centimeters.

Any two means whose differences are less than the  $Q_t$  value are connected by a vertical bar. Any pair of means enclosed by the range of any one bar is not significantly different. .

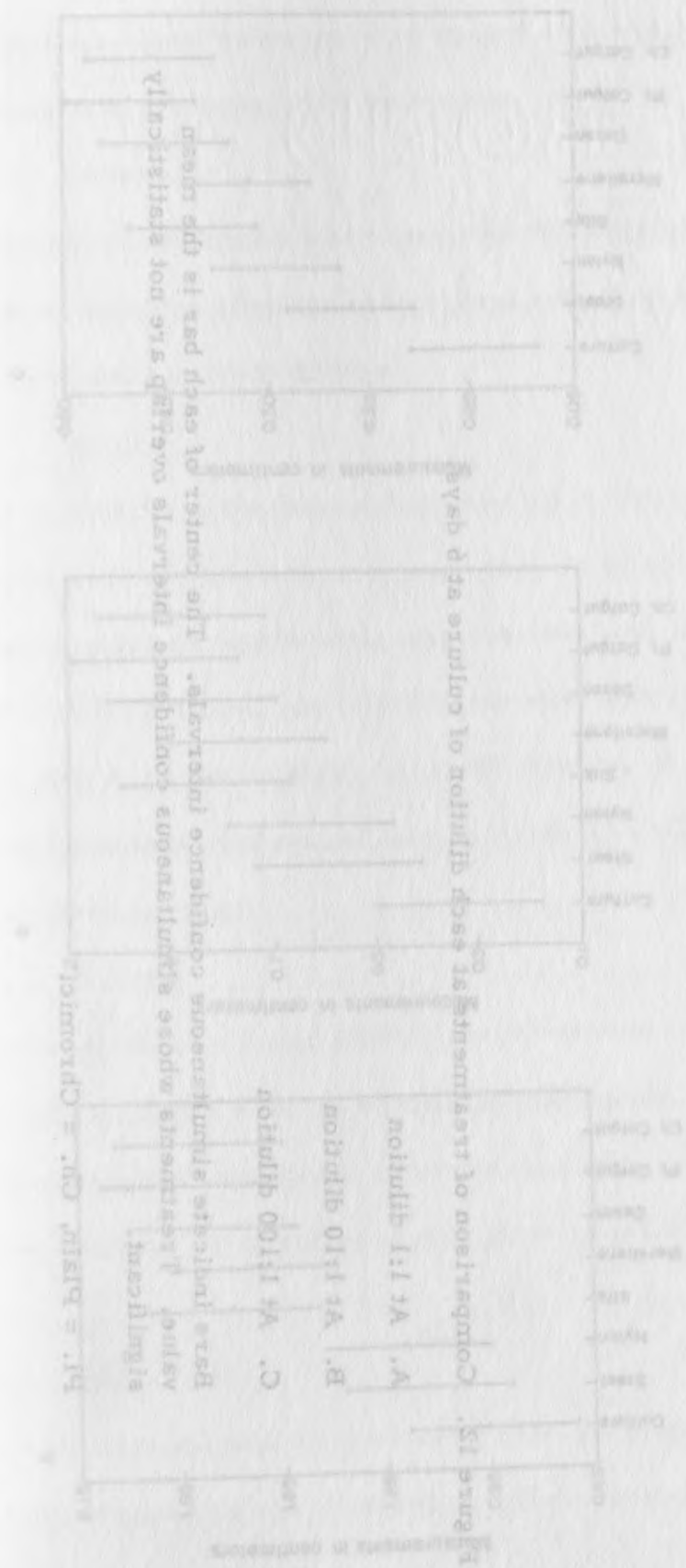


Figure 15: Comparison of treatments at each dilution of caffeine with dsAg.

Y-axis: Response (0 to 1.5)

X-axis: Dilution (C, B, A)

Legend: C: V: 1:100 dilution; B: V: 1:10 dilution; A: V: 1:1 dilution

Significance: Significant difference between C and B.

Figure 15: Comparison of treatments at each dilution of caffeine with dsAg.

Y-axis: Response (0 to 1.5)

X-axis: Dilution (C, B, A)

Legend: C: V: 1:100 dilution; B: V: 1:10 dilution; A: V: 1:1 dilution

Significance: Significant difference between C and B.

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Legend: C: V: 1:100 dilution; B: V: 1:10 dilution; A: V: 1:1 dilution

Significance: Significant difference between C and B.

Figure 12. Comparison of treatments at each dilution of culture at 6 days.

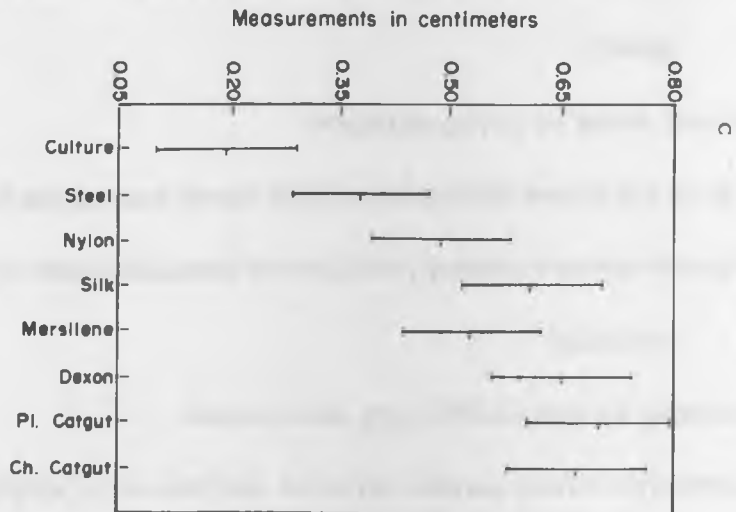
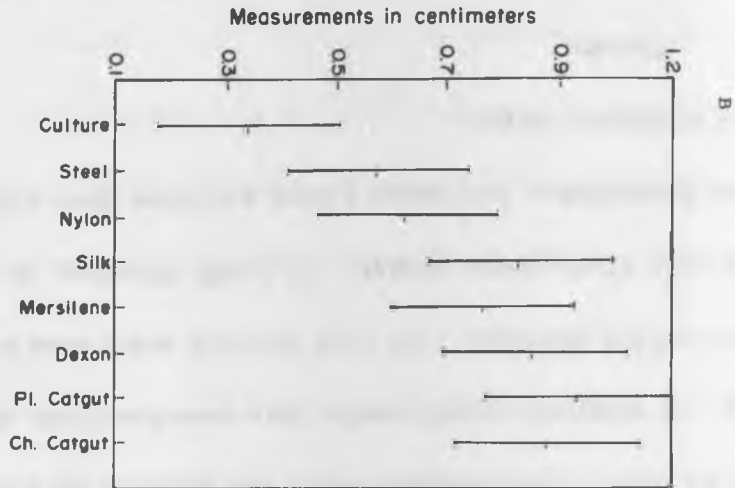
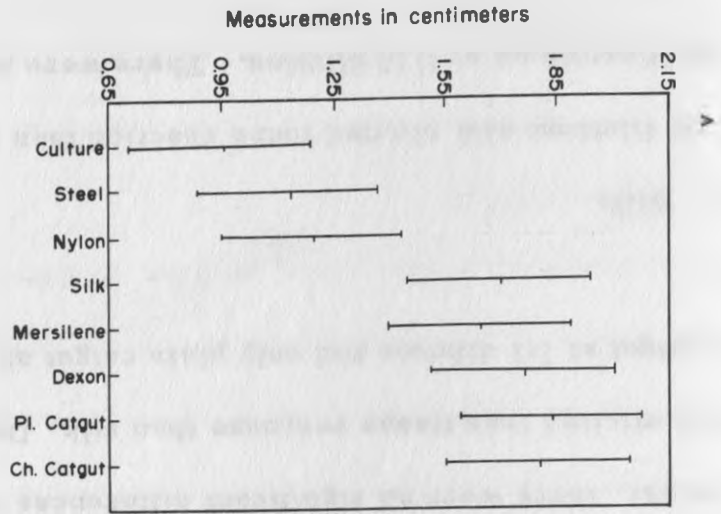
A. At 1:1 dilution

B. At 1:10 dilution

C. At 1:100 dilution

Bars indicate simultaneous confidence intervals. The center of each bar is the mean value. Treatments whose simultaneous confidence intervals overlap are not statistically significant.

Pl. = Plain, Ch. = Chromic.



similarities discussed below refer to statistically significant value as determined by the range-STP procedure.

1. Culture:

Culture alone elicited less tissue reaction than all suture materials at all three dilutions except steel and nylon at 1:1 and 1:10 dilutions and steel at 1:100 dilution.

2. Steel:

Steel stimulated the least reaction of all suture materials. No differences were seen between steel and culture or nylon at all three dilutions. It elicited significantly less reaction than all sutures other than nylon at 1:1 dilution. At 1:10 dilution steel was significantly different only from plain catgut. At 1:100 dilution, it was similar to nylon and Mersilene, but caused less reaction than silk, Dexon and plain and chromic catgut.

3. Nylon:

Although nylon elicited slightly more reaction than steel at all three dilutions, there were no significant differences between the two. Nylon elicited less tissue response than silk, Dexon, chromic and plain catgut at 1:1 dilution and only plain catgut at 1:10 and 1:100 dilutions.

4. Silk:

At all dilutions silk elicited more reaction than steel and nylon, but not significantly so at 1:10 dilution. There were no significant

differences between silk, Mersilene, Dexon, plain and chromic catgut at all three dilutions.

5. Mersilene:

Mersilene was very similar to silk in stimulating reaction although it elicited slightly less reaction than silk at all three dilutions.

6. Dexon:

At all three dilutions Dexon caused slightly more reaction than silk and Mersilene, and less than plain and chromic catgut, but not significantly different from any of these.

7. Catgut (Plain and Chromic):

There were no differences between plain and chromic catgut at all three levels of inoculation. Neither one differed significantly from silk, Mersilene and Dexon. Plain catgut elicited the most intense reaction of all suture materials, but it was not significantly different from chromic catgut.

Figure 13 shows dilution-treatment interaction, comparing treatment means at each of the three dilutions of culture at 6 days. At all three dilutions, tissue response to sutures in ascending order was 1) steel, 2) nylon, 3) Mersilene, 4) silk, 5) Dexon, 6) chromic catgut and 7) plain catgut.

A two-way factorial randomized block analysis of variance for the global view of group A is shown in Table 14. F values were found

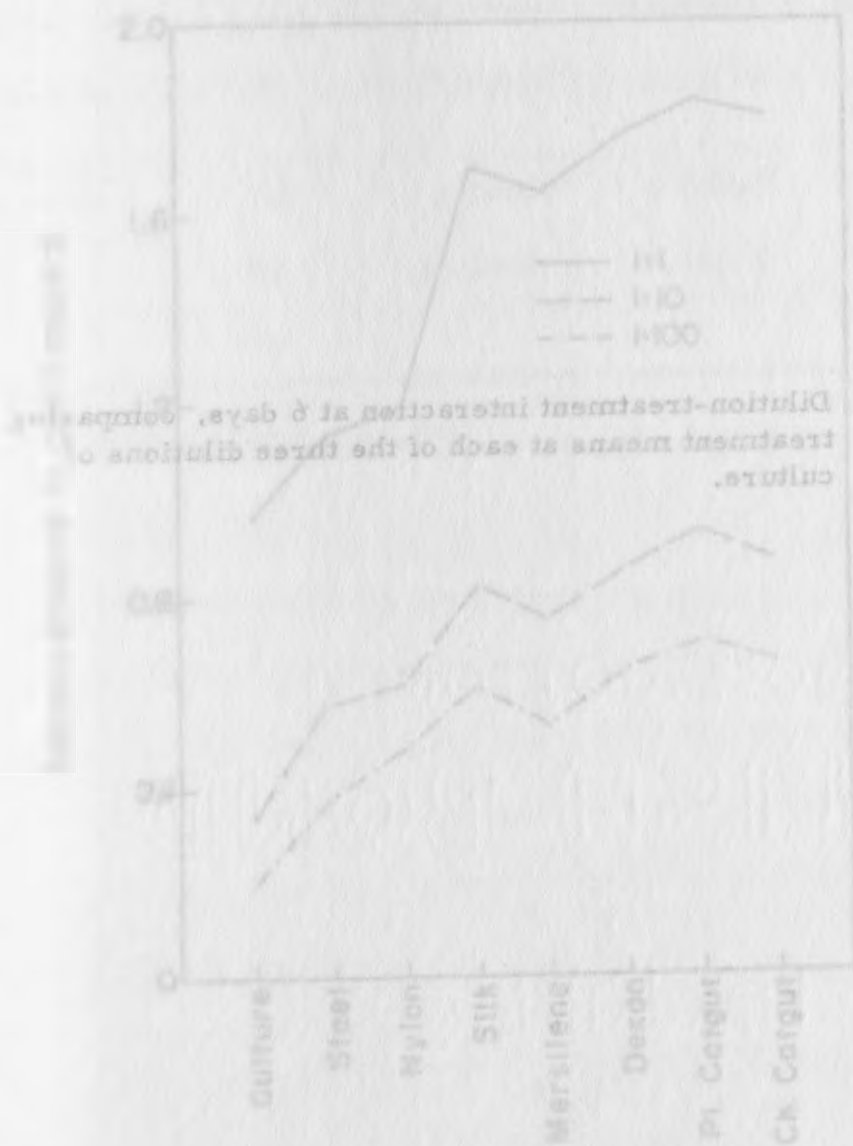




Figure 13. Dilution-treatment interaction at 6 days, comparing treatment means at each of the three dilutions of culture.

Measurements in centimeters

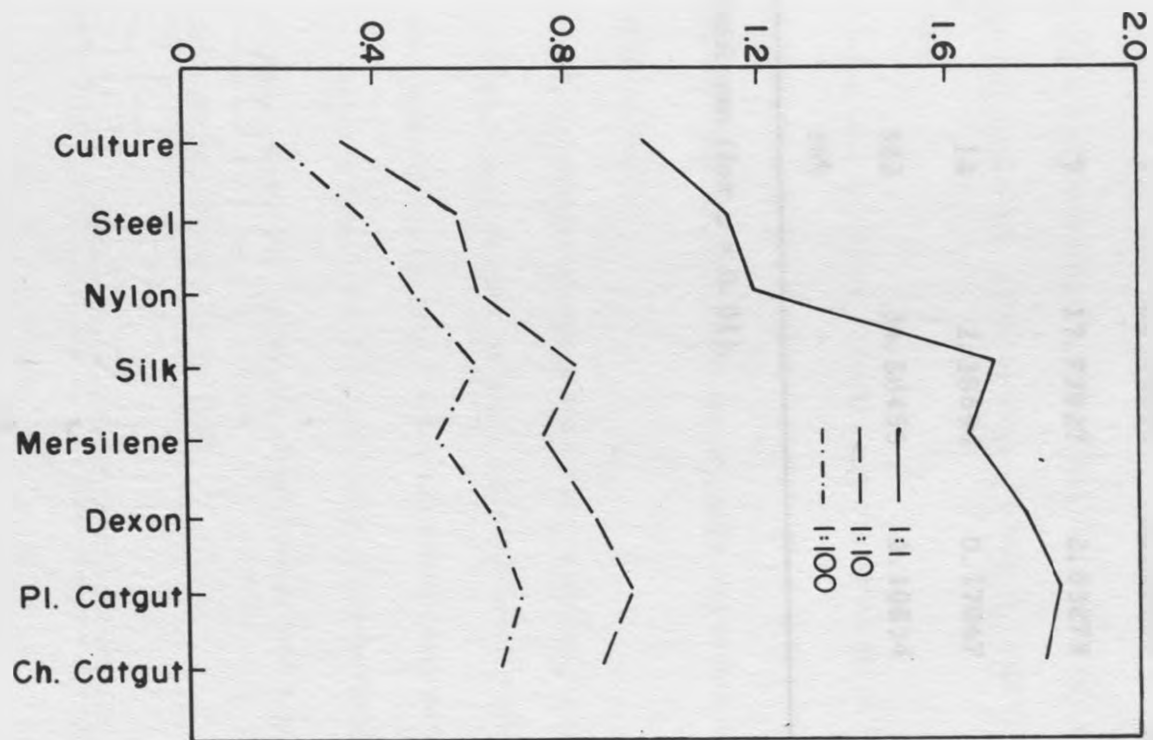


Table 14. Two-Way Factorial Randomized Block Analysis of Variance for the Global View of Group A (6 Days).

Source of Variation	Degrees of Freedom	Sums of Squares	Mean Squares	F
Dogs	14	5.59081	0.39934	
Dilutions	2	64.93935	32.46967	299.710***
Treatments	7	17.72927	2.53275	23.378***
Dilutions x Treatments	14	2.38654	0.17047	1.574
Error	322	34.88453	0.10834	
Total	359			

\*\*\* Significant (for  $\alpha = 0.01$ ).

significant for dilutions and for treatments at the 1 percent level.

The range-STP test was performed on both aspects. The table value of the special statistic, at the 5 percent level, for the three dilutions was 3.32, and for the eight treatments it was 4.286. The mean dilution and treatment values and their pairwise comparisons are shown in Table 15. The corresponding  $Q_d$  and  $Q_t$  values are also shown.

Figure 14 A and B shows graphic representation of dilution means with  $Q_d$  range and the overall treatment means with  $Q_t$  range.

It is clearly seen that there were significant differences between the three dilutions used.

The overall treatment pattern was very similar to that observed for the treatment means at each dilution level separately. Culture elicited significantly less tissue reaction than any suture. Steel and nylon caused less reaction than the other sutures. Silk, Mersilene, Dexon, plain and chromic catgut were not significantly different in stimulating tissue reactions.

#### Gross Measurements of Lesions and Statistical Tests for Group B (10 Days)

Measurements of lesion diameters in centimeters in the 1:1, 1:10 and 1:100 dilution rows in each dog in group B are shown in Tables 16, 17 and 18.

Table 15. Dilution and Treatment Means for the Global View with  $Q_d$  and  $Q_t$  Values for Group A (6 Days).

Means*		
<u>Dilutions</u>		
1:1	1.50750	$Q_d = 3.32 \sqrt{\frac{0.10834}{120}}$ = 0.100
1:10	0.72083	
1:100	0.52458	
<u>Treatments</u>		
Culture	0.49556	$Q_t = 4.286 \sqrt{\frac{0.10834}{45}}$ = 0.210
Steel	0.69667	
Nylon	0.76778	
Mersilene	0.98000	
Silk	1.04111	
Dexon	1.08667	
Chromic Catgut	1.11556	
Plain Catgut	1.15778	

\* Lesion measurements in centimeters.

Any two means whose differences are less than the  $Q_d$  or  $Q_t$  value are connected by a vertical bar. Any pair of means enclosed by the range of any one bar is not significantly different.

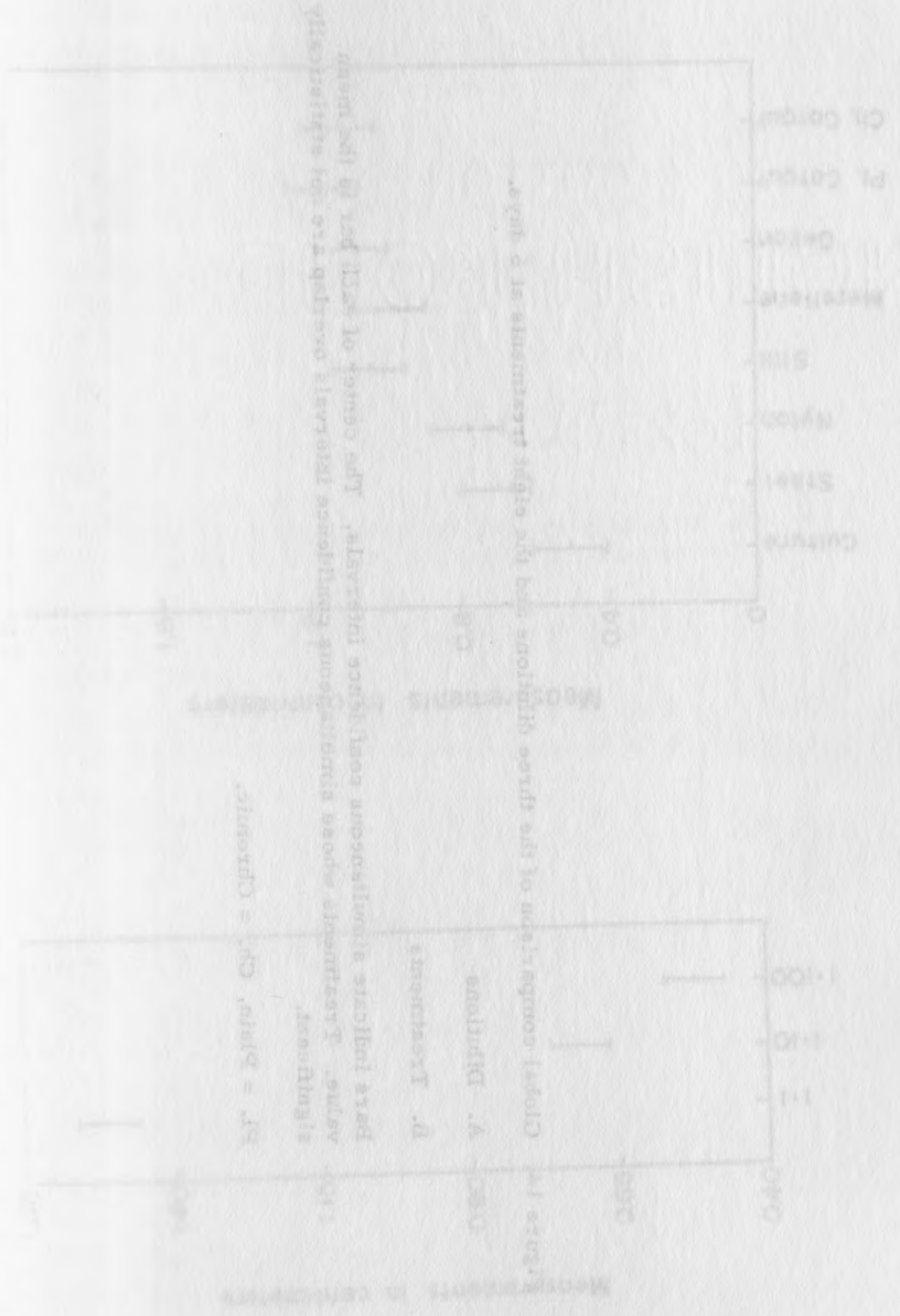


Figure 14. Global comparison of the three dilutions and the eight treatments at 6 days.

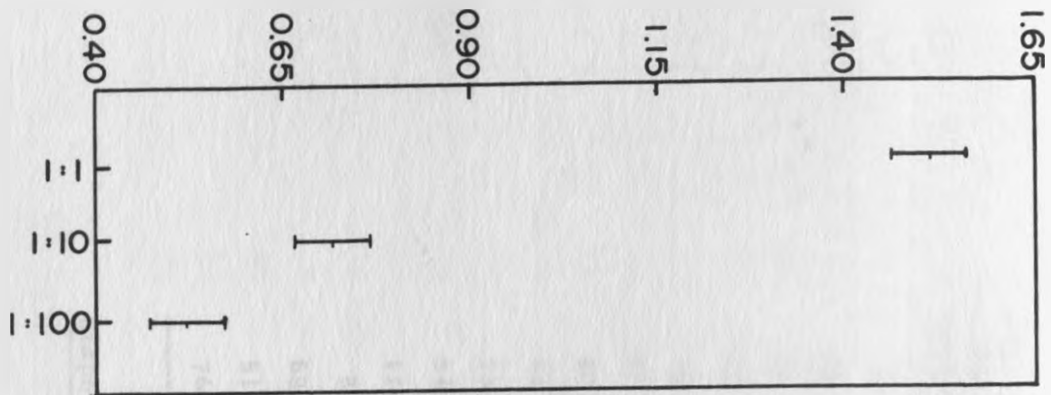
A. Dilutions

B. Treatments

Bars indicate simultaneous confidence intervals. The center of each bar is the mean value. Treatments whose simultaneous confidence intervals overlap are not statistically significant.

Pl. = Plain, Ch. = Chromic.

Measurements in centimeters



Measurements in centimeters

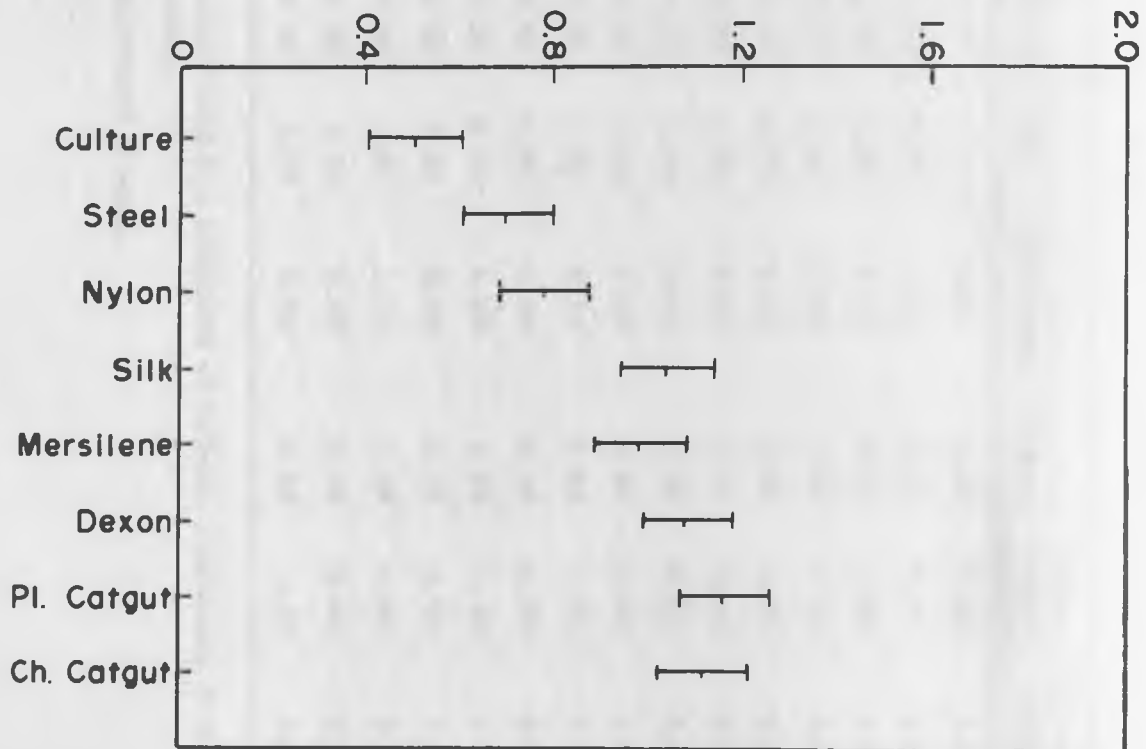




Table 16. Gross Measurements of Lesion Diameters in Centimeters in the 1:1 Dilution Rows of Group B (10 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
8		0.70	0.65	1.10	0.75	0.80	1.40	1.20
18	0.30	0.75	1.15	0.75	1.20	0.65	0.85	1.00
25		1.10	1.00	0.70	0.75	0.65	1.00	1.05
31		0.45	0.50	0.80	0.40	0.60	1.30	0.75
35		0.30	0.10	0.75	0.40	0.55	1.25	0.65
42		1.30	0.65	1.20	0.55	0.75	1.60	0.70
40		0.25	0.45	1.10	0.70	0.30	0.50	0.50
44		0.45	0.50	1.30	1.00	0.90	1.50	1.60
33	0.30	0.45	0.30	1.00	0.85	0.80	1.15	1.15
64		0.40	0.55	0.85	0.40	0.50	0.70	0.75
11		0.75	1.00	0.95	1.00	1.20	1.40	1.50
5		1.60	1.40	1.60	0.45	0.35	0.90	1.10
68	0.40	0.80	0.20	1.80	1.15	1.45	0.50	0.45
51		0.60	0.30	2.20	0.85	0.40	0.60	0.50
76		0.20	0.30	1.15	0.50	0.60	1.25	0.85

\* Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^6$  Staphylococcus aureus organisms.

Blanks indicate a measurement of zero.

Table 17. Gross Measurements of Lesion Diameters in Centimeters in the 1:10 Dilution Rows of Group B (10 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
8		0.60	0.30	1.15	0.60	0.50	0.95	1.00
18		0.30	0.35	0.40	0.85	0.40	0.45	0.70
25	0.45	0.60	0.40	0.85	1.00	0.65	0.80	1.10
31		0.70	0.35	0.50	0.50	0.50	0.95	0.45
35				0.70	0.40	0.50	0.55	0.60
42				0.40			0.60	0.70
40		0.50	0.30	0.55	1.00	0.65	0.70	0.40
44		0.30	0.30	0.80	0.35	0.60	0.45	0.65
33		0.75		0.90	0.55	0.45	0.90	1.45
64						0.60	0.65	0.50
11		0.55	0.40	1.00	0.65	0.60	1.20	0.70
5				0.50	0.60	0.60	0.60	0.70
68	0.70	0.45	0.40	0.50	1.10	0.70	1.10	0.35
51				1.30	0.40	0.60	0.40	1.10
76		0.70		0.45		0.50	0.50	0.70

\*Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^5$  Staphylococcus aureus organisms.

Blanks indicate a measurement of zero.

Table 18. Gross Measurements of Lesion Diameters in Centimeters in the 1:100 Dilution Rows of Group B (10 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
8				0.60	0.30	0.30		0.65
18			0.30	0.40	0.55	0.35	0.30	0.60
25		0.50	0.30	0.50	0.50	0.50	0.65	1.00
31		0.20		0.40	0.40		0.65	a
35		0.20		0.45	0.20	0.30	0.50	0.50
42				0.60	1.30		0.60	
40					0.50		0.70	0.40
44							0.80	
33								
64							0.45	0.40
11		0.45	0.45	0.90	0.60	0.65	0.50	0.65
5					0.50	1.00	0.45	0.70
68	0.45	0.50	0.30	0.60	0.50	0.60		0.40
51				0.80	0.40	0.50	0.70	0.45
76						0.45	0.80	0.45

\* Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^4$  Staphylococcus aureus organisms.

<sup>a</sup>Leaking pocket.

Blanks indicate a measurement of zero.

Results for one-way randomized block analysis of variance for each dilution are shown in Table 19. F values were found significant, at the 1 percent level, for treatments at all three dilutions.

Table 20 shows lesion means and their pairwise comparisons at each dilution. The corresponding  $Q_t$  values are also calculated. Graphic representation of this is shown in Figure 15 A, B and C.

Significant differences are discussed below:

1. Culture:

Culture elicited significantly less tissue reaction than all sutures at 1:1 dilution. At 1:10 dilution, culture was significantly different from all sutures except nylon and at 1:100 dilution there were no differences between culture, steel and nylon.

2. Steel:

There were no significant differences between steel and nylon at all levels of inoculation, although steel elicited slightly more reaction. It elicited less reaction than plain catgut and silk at 1:1 dilution, silk, plain and chromic catgut at 1:10 dilution and only the catguts at 1:100 dilution.

3. Nylon:

Nylon stimulated the least reaction of all suture materials at all levels of inoculation. It showed significantly less reaction than silk and plain catgut at 1:1 dilution, silk, Mersilene, Dexon, plain and chromic catgut at 1:10 dilution, and Mersilene and the catguts at 1:100 dilution of culture.

Table 19. One-Way Randomized Block Analysis of Variance for Treatment Means at Each Level of Inoculation for Group B (10 Days).

	Source of Variation	Degrees of Freedom	Sums of Squares	Mean Squares	F
1:1 Dilution	Dogs	14	2.98875	0.21348	
	Treatments	7	11.69792	1.67113	16.207***
	Error	98	10.10458	0.10311	
	Total	119			
1:10 Dilution	Dogs	14	2.86075	0.20434	
	Treatments	7	6.40481	0.91497	16.715***
	Error	98	5.36425	0.05474	
	Total	119			
1:100 Dilution	Dogs	14	2.32125	0.16580	
	Treatments	7	3.03600	0.43371	7.919***
	Error	98	5.36775	0.05477	
	Total	119			

\*\*\* Significant (for  $\alpha = 0.01$ ).

Table 20. Treatment Means for Each Level of Inoculation with the Corresponding  $Q_t$  Values for Group B (10 Days).

Treatments	Means*	
		<u>1:1 Dilution</u>
Culture	0.06667	
Nylon	0.60333	$Q_t = 4.389 \sqrt{\frac{0.10311}{15}}$ $= 0.364$
Steel	0.67333	
Dexon	0.70000	
Mersilene	0.73000	
Chromic Catgut	0.91667	
Plain Catgut	1.06000	
Silk	1.15000	
Culture	0.07667	
Nylon	0.18667	$Q_t = 4.389 \sqrt{\frac{0.05477}{15}}$ $= 0.265$
Steel	0.36333	
Dexon	0.52333	
Mersilene	0.53333	
Silk	0.66667	
Plain Catgut	0.72000	
Chromic Catgut	0.74000	
Culture	0.03000	
Nylon	0.09000	$Q_t = 4.389 \sqrt{\frac{0.05621}{15}}$ $= 0.269$
Steel	0.12333	
Dexon	0.31000	
Silk	0.35000	
Mersilene	0.38333	
Chromic Catgut	0.41333	
Plain Catgut	0.47000	

\* Lesion measurements in centimeters.

Any two means whose differences are less than the  $Q_t$  value are connected by a vertical bar. Any pair of means enclosed by the range of any one bar is not significantly different.

Unit 3:  $3x^2 - 2x - 4 = 0$  - Quadratic

Mathematics: Completing the square

Step 1: Write the quadratic in the form  $ax^2 + bx + c = 0$ . In this case,  $a = 3$ ,  $b = -2$ , and  $c = -4$ .

Step 2: Divide the entire equation by  $a$  to get  $x^2 + \frac{b}{a}x + \frac{c}{a} = 0$ . Here,  $x^2 - \frac{2}{3}x - \frac{4}{3} = 0$ .

Step 3: Move the constant term  $\frac{c}{a}$  to the right side of the equation:  $x^2 - \frac{2}{3}x = \frac{4}{3}$ .

Step 4: Find the value to complete the square. Take half of the coefficient of  $x$ , which is  $-\frac{1}{3}$ , and square it to get  $\frac{1}{9}$ .

Step 5: Add  $\frac{1}{9}$  to both sides of the equation:  $x^2 - \frac{2}{3}x + \frac{1}{9} = \frac{4}{3} + \frac{1}{9}$ .

Step 6: Write the left side as a perfect square:  $(x - \frac{1}{3})^2 = \frac{13}{9}$ .

Step 7: Take the square root of both sides:  $x - \frac{1}{3} = \pm \sqrt{\frac{13}{9}}$ .

Step 8: Solve for  $x$ :  $x = \frac{1}{3} \pm \frac{\sqrt{13}}{3}$ .

Step 9: Write the solutions:  $x = \frac{1 + \sqrt{13}}{3}$  or  $x = \frac{1 - \sqrt{13}}{3}$ .

Figure 15. Comparison of treatments at each dilution of culture at 10 days.

A. At 1:1 dilution

B. At 1:10 dilution

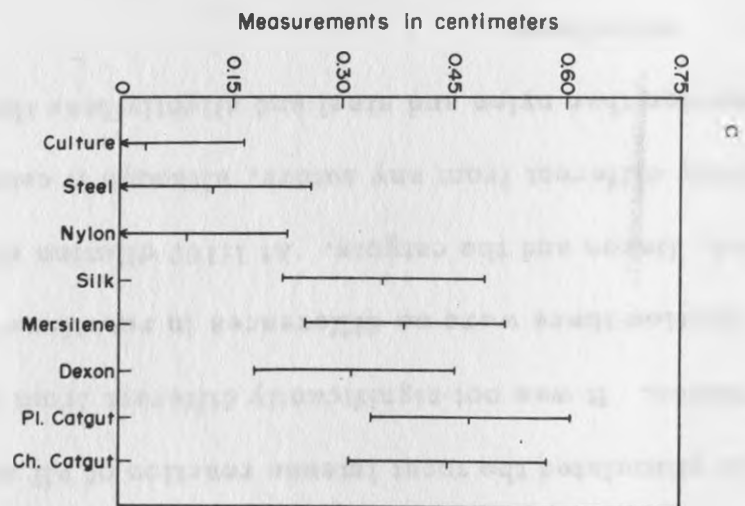
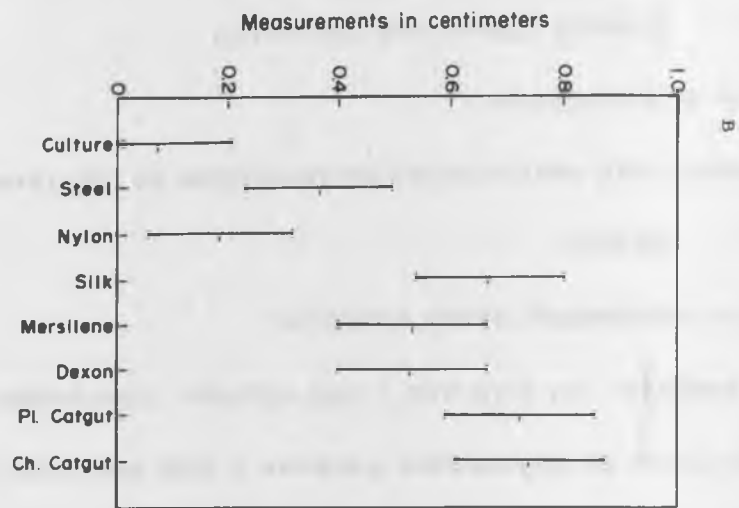
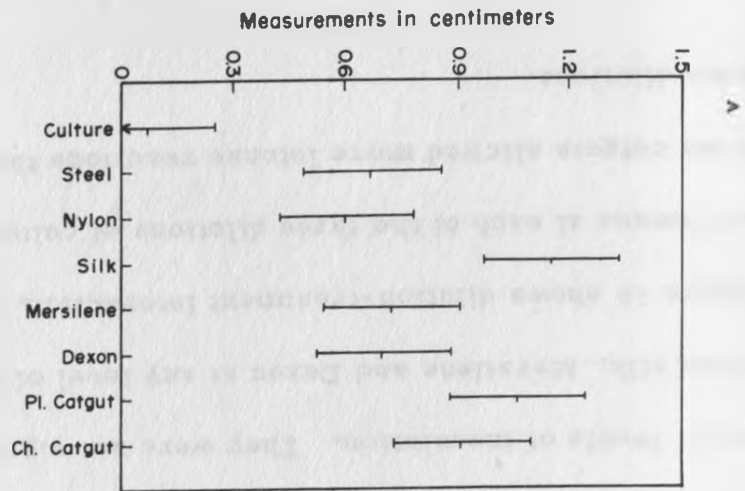
C. At 1:100 dilution

Bars indicate simultaneous confidence intervals. The center of each bar is the mean value. Treatments whose simultaneous confidence intervals overlap are not statistically significant.

Bars that reach the abscissa ( $\downarrow$ ) indicate that the mean value is less than half the simultaneous confidence interval.

Pl. = Plain, Ch. = Chromic.





#### 4. Silk:

Silk stimulated the most intense reaction of all suture materials at 1:1 dilution. It was not significantly different from the catguts. At 1:10 dilution there were no differences in reactions to silk, Mersilene, Dexon and the catguts. At 1:100 dilution silk was not significantly different from any suture, although it caused slightly more reaction than nylon and steel and slightly less than the catguts.

#### 5. Mersilene:

At 1:1 dilution, Mersilene stimulated less reaction than silk but there were no differences between it and steel and nylon or Dexon and the catguts. At 1:10 and 1:100 dilution, Mersilene was similar to silk in stimulating tissue reaction.

#### 6. Dexon:

Dexon was very similar to Mersilene in its tissue response at all levels of inoculation.

#### 7. Catguts (Plain and Chromic):

There were no significant differences between plain and chromic catgut at all levels of inoculation. They were not significantly different from silk, Mersilene and Dexon at any level of inoculation.

Figure 16 shows dilution-treatment interaction, comparing treatment means at each of the three dilutions of culture at 10 days. Silk and the catguts elicited more intense reactions than other sutures at all three dilutions.

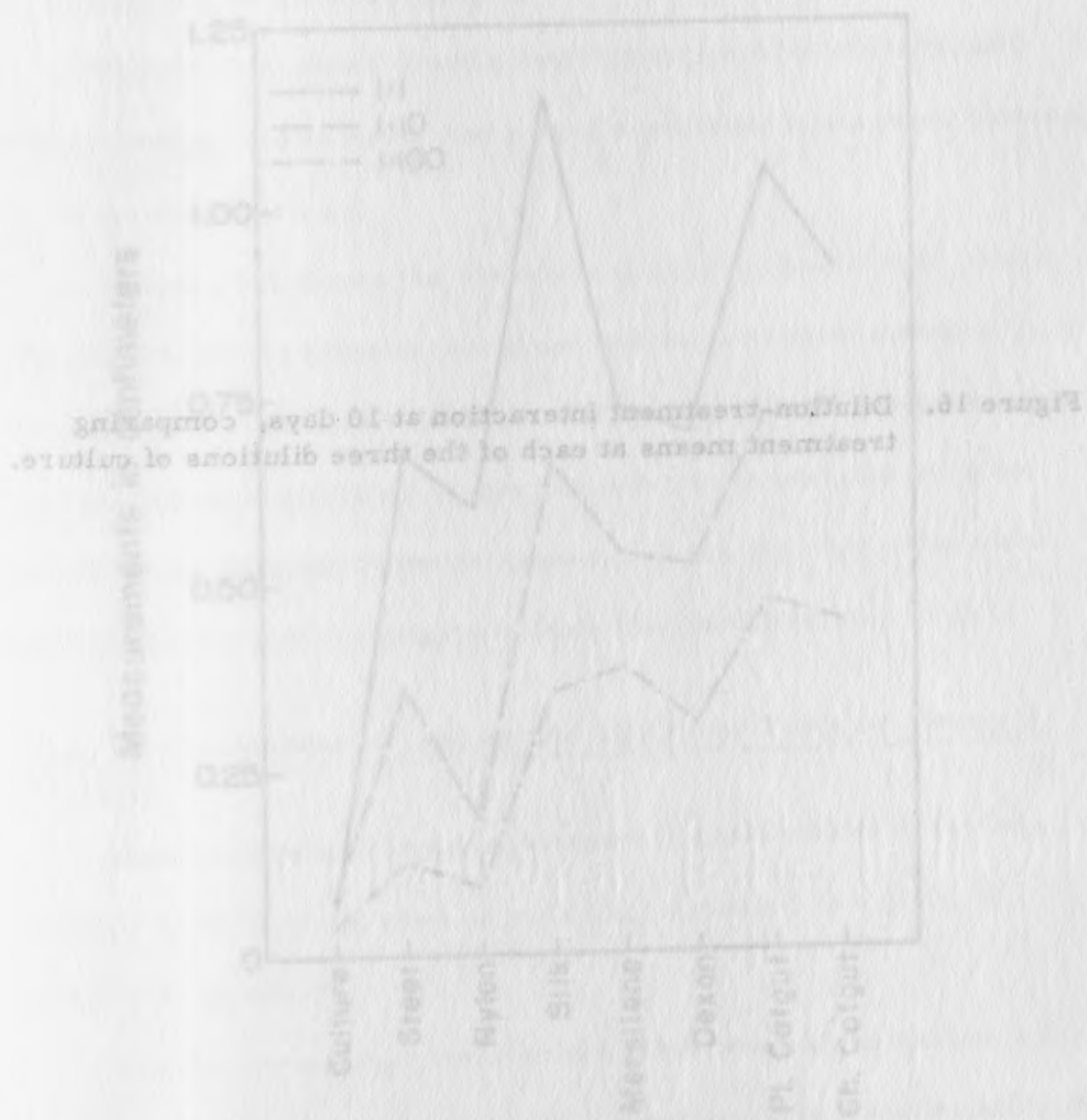
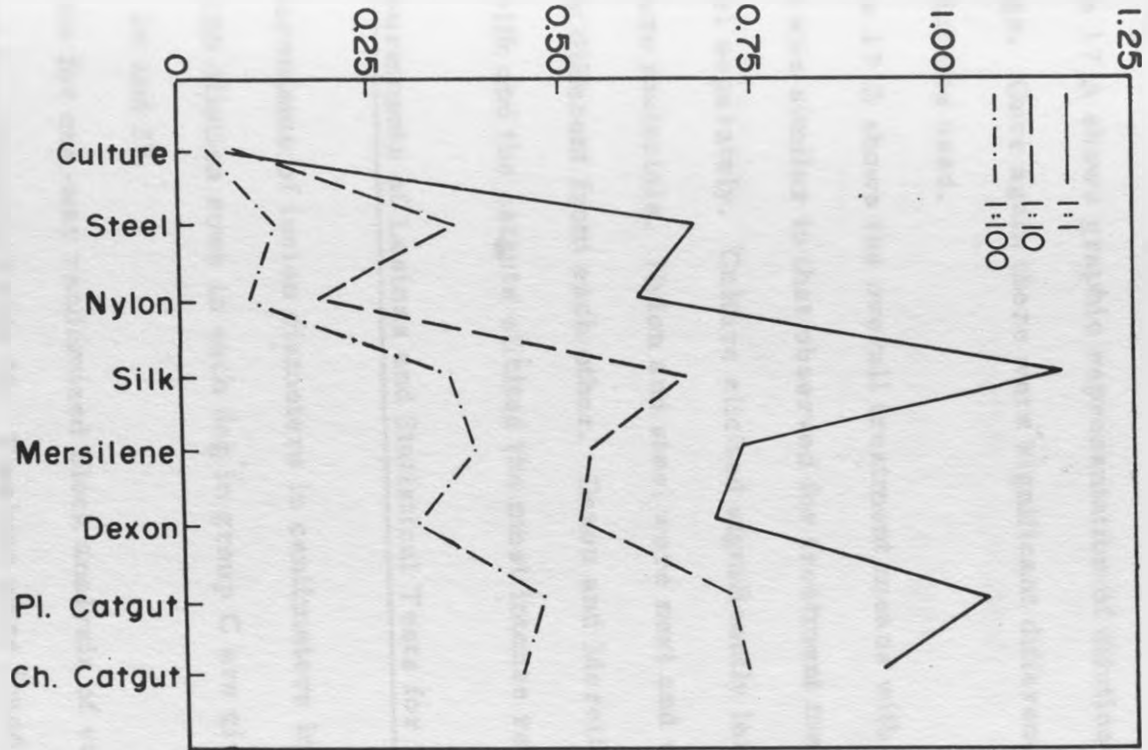


Figure 16. Dilution-treatment interaction at 10 days, comparing treatment means at each of the three dilutions of culture.

Measurements in centimeters



A two-way factorial randomized block analysis of variance for the global view of group B is shown in Table 21. F values were found significant. The mean dilution and treatment values and their pairwise comparisons are shown in Table 22. The corresponding  $Q_d$  and  $Q_t$  values are also given.

Figure 17 A shows graphic representation of dilution means with  $Q_d$  range. Once again there were significant differences between the three dilutions used.

Figure 17 B shows the overall treatment means with  $Q_t$  range. The pattern was similar to that observed for treatment means at each dilution level separately. Culture elicited significantly less reaction than all suture materials. Nylon and steel were next and were not significantly different from each other. Dexon and Mersilene were next while silk and the catguts elicited the most intense reaction.

#### Gross Measurements of Lesions and Statistical Tests for Group C (20 Days)

Measurements of lesion diameters in centimeters in the 1:1, 1:10 and 1:100 dilution rows in each dog in group C are given in Tables 23, 24 and 25.

Results for one-way randomized block analysis of variance for each dilution are shown in Table 26. F values were found significant, at the 1 percent level, for treatments at all three dilutions.

Table 21. Two-Way Factorial Randomized Block Analysis of Variance for the Global View of Group B (10 Days).

Source of Variation	Degrees of Freedom	Sums of Squares	Mean Squares	F
Dogs	14	4.65421	0.33244	
Dilutions	2	12.90637	6.45319	85.325***
Treatments	7	18.24810	2.60687	34.468***
Dilutions x Treatments	14	2.89062	0.20647	2.730***
Error	322	24.35312	0.07563	
Total	359			

\*\*\* Significant (for  $\alpha = 0.01$ ).

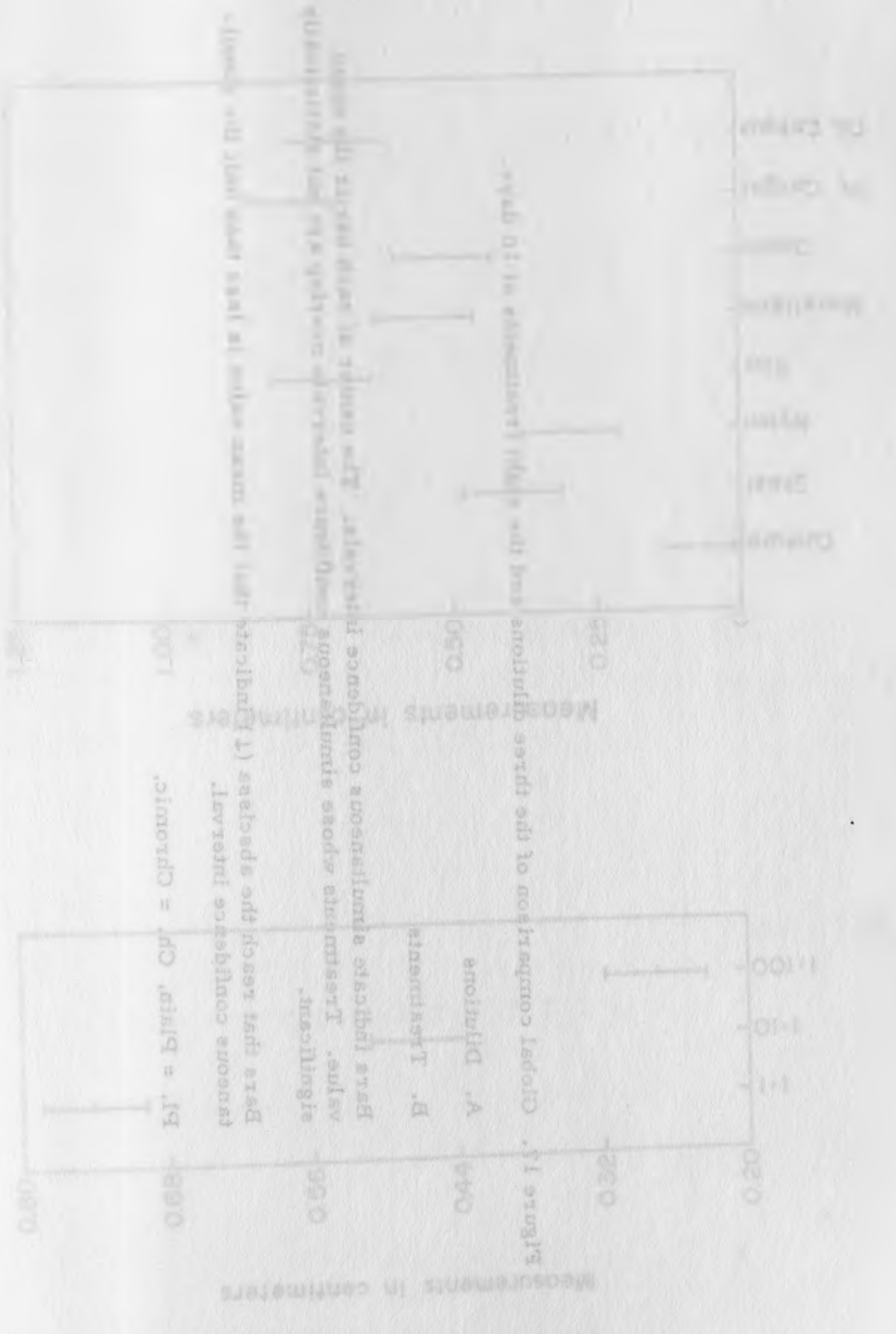
Table 22. Dilution and Treatment Means for the Global View with  $Q_d$  and  $Q_t$  Values for Group B (10 Days).

Means*		
<u>Dilutions</u>		
1:1	0.73750	$Q_d = 3.32 \sqrt{\frac{0.07563}{120}}$ $= 0.083$
1:10	0.47625	
1:100	0.27500	
<u>Treatments</u>		
Culture	0.05778	$Q_t = 4.286 \sqrt{\frac{0.07563}{120}}$ $= 0.176$
Nylon	0.29333	
Steel	0.38667	
Dexon	0.51111	
Mersilene	0.54889	
Chromic Catgut	0.69889	
Silk	0.72222	
Plain Catgut	0.75111	

\* Lesion measurements in centimeters.

Any two means whose differences are less than the  $Q_d$  or  $Q_t$  value are connected by a vertical bar. Any pair of means enclosed by the range of any one bar is not significantly different. .





Measurements in Centimeters

B1 = B2 vs B3, CR = Chi-squared

Measurements in Centimeters

Figure 13. Group comparison of the three conditions

Figure 17. Global comparison of the three dilutions and the eight treatments at 10 days.

A. Dilutions

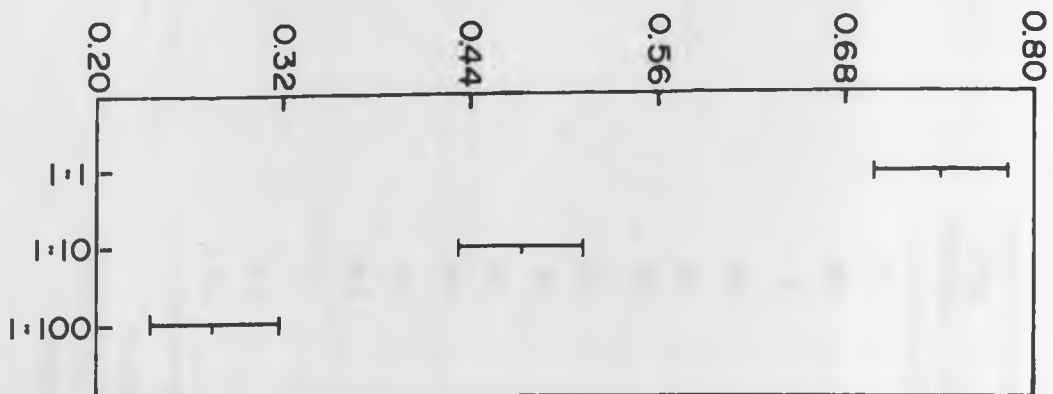
B. Treatments

Bars indicate simultaneous confidence intervals. The center of each bar is the mean value. Treatments whose simultaneous confidence intervals overlap are not statistically significant.

Bars that reach the abscissa ( $\downarrow$ ) indicate that the mean value is less than half the simultaneous confidence interval.

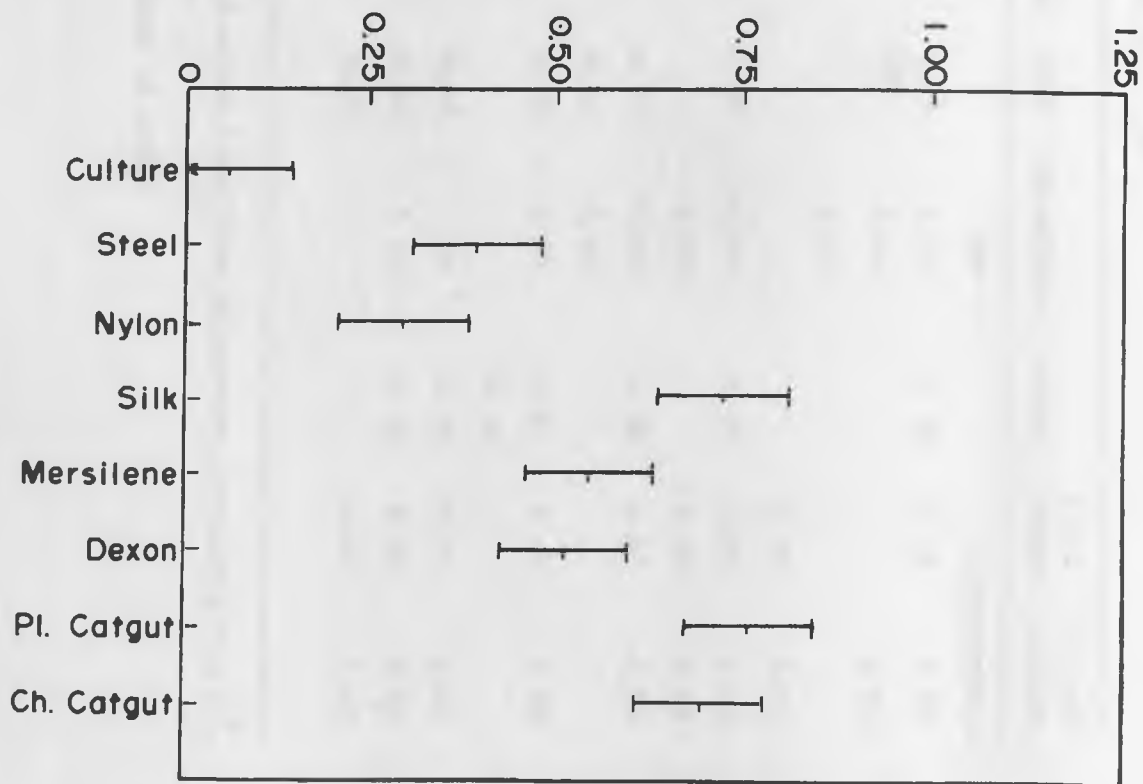
Pl. = Plain, Ch. = Chromic.

Measurements in centimeters



A

Measurements in centimeters



B

Table 23. Gross Measurements of Lesion Diameters in Centimeters in the 1:1 Dilution Rows of Group C (20 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
12					1.00			
20				1.15	0.60	0.30	0.55	0.80
6					0.90			0.40
38		0.40			0.85			
34							0.70	0.55
47				0.65	0.30	0.50	0.60	0.35
53					0.55		0.70	0.45
36		0.25		0.70	0.70	0.20	0.65	0.65
65				0.80	0.70			
59			0.40	0.60	0.70	0.45	0.40	0.40
69						0.35		
74		0.35		0.85	0.70	0.40	0.40	0.80
71				0.85	0.70	0.35	0.50	0.60
81				0.65			0.30	0.45
54								

\* Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^6$  Staphylococcus aureus organisms.

Blanks indicate a measurement of zero.

Table 24. Gross Measurements of Lesion Diameters in Centimeters in the 1:10 Dilution Rows of Group C (20 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
12								
20								
6			b	0.70				0.40
38								
34		0.60			0.60		0.65	0.60
47								
53							0.40	
36	b			0.90				
65								
59	0.20	0.60		0.60			0.40	0.45
69						0.30		0.35
74				0.95		0.50		
71		0.60		0.65	0.80		0.50	0.35
81				0.65				
54								

\* Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^5$  Staphylococcus aureus organisms.

<sup>b</sup> Bleeding pockets.

Blanks indicate a measurement of zero.

Table 25. Gross Measurements of Lesion Diameters in Centimeters in the 1:100 Dilution Rows of Group C (20 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
12								
20								
6				0.50				
38								
34							0.70	
47								
53								
36				0.50			0.50	0.70
65					a			
59				0.35	0.50	0.45		
69							0.50	0.55
74				0.80				0.80
71		0.60		0.80		0.25	0.35	0.40
81								
54					a		0.50	

\* Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^4$  *Staphylococcus aureus* organisms.

<sup>a</sup> Leaking pockets.

Blanks indicate a measurement of zero.

Table 26. One-Way Randomized Block Analysis of Variance for Treatment Means at Each Level of Inoculation for Group C (20 Days).

	Source of Variation	Degrees of Freedom	Sums of Squares	Mean Squares	F
1:1 Dilution	Dogs	14	2.17544	0.15540	10.116***
	Treatments	7	3.98031	0.56862	
	Error	98	5.50812	0.05621	
	Total	119			
1:10 Dilution	Dogs	14	1.63512	0.11679	3.125***
	Treatments	7	0.92465	0.13209	
	Error	98	4.14254	0.04227	
	Total	119			
1:100 Dilution	Dogs	14	1.08188	0.07728	3.076***
	Treatments	7	0.69498	0.09928	
	Error	98	3.16346	0.03228	
	Total	119			

\*\*\* Significant (for  $\alpha = 0.01$ ).

Table 27 shows lesion means and their pairwise comparisons at each dilution. The corresponding  $Q_t$  values are also given.

Graphic representation of this is shown in Figure 18 A, B and C.

Significant differences are discussed below:

1. Culture:

The mean value for culture was zero at 1:1 and 1:100 dilutions and only 0.013 centimeter at 1:10 dilution, showing that at 20 days the effect of culture alone is insignificant.

2. Steel:

Steel elicited significantly less tissue reaction than silk, Mersilene and chromic catgut at 1:1 dilution but there were no significant differences between steel and other sutures at 1:10 and 1:100 dilutions. Although stimulating slightly more reaction than nylon, the two were not significantly different from each other.

3. Nylon:

Nylon elicited the least reaction of all sutures at all three dilutions.

4. Silk:

At 1:1 dilution, silk caused slightly less reaction than Mersilene, although not significantly so. It elicited significantly more reaction than steel and nylon. At 1:10 and 1:100 dilutions silk enhanced the most intense tissue reaction. At 1:10 dilution it showed



Table 27. Treatment Means for Each Level of Inoculation with the Corresponding  $Q_t$  Values for Group C (20 Days).

Treatments	Means*	
		<u>1:1 Dilution</u>
Culture	0.00000	$Q_t = 4.389 \sqrt{\frac{0.05621}{15}}$ $= 0.269$
Nylon	0.02667	
Steel	0.06667	
Dexon	0.17000	
Plain Catgut	0.32000	
Chromic Catgut	0.36333	
Silk	0.41667	
Mersilene	0.51333	
		<u>1:10 Dilution</u>
Nylon	0.00000	$Q_t = 4.389 \sqrt{\frac{0.04227}{15}}$ $= 0.233$
Culture	0.01333	
Dexon	0.05333	
Steel	0.12000	
Plain Catgut	0.13000	
Mersilene	0.13333	
Chromic Catgut	0.14333	
Silk	0.29667	
		<u>1:100 Dilution</u>
Culture	0.00000	$Q_t = 4.389 \sqrt{\frac{0.01480}{15}}$ $= 0.130$
Nylon	0.00000	
Mersilene	0.03333	
Steel	0.04000	
Dexon	0.04667	
Chromic Catgut	0.16333	
Plain Catgut	0.17000	
Silk	0.19667	

\* Lesion measurements in centimeters.

Any two means whose differences are less than the  $Q_t$  value are connected by a vertical bar. Any pair of means enclosed by the range of any one bar is not significantly different.



Figure 18. Comparison of treatments at each dilution of culture at 20 days.

A. At 1:1 dilution

B. At 1:10 dilution

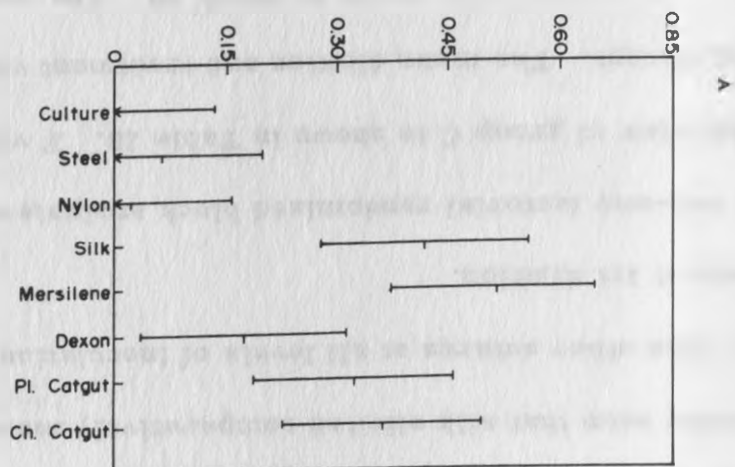
C. At 1:100 dilution

Bars indicate simultaneous confidence intervals. The center of each bar is the mean value. Treatments whose simultaneous confidence intervals overlap are not statistically significant.

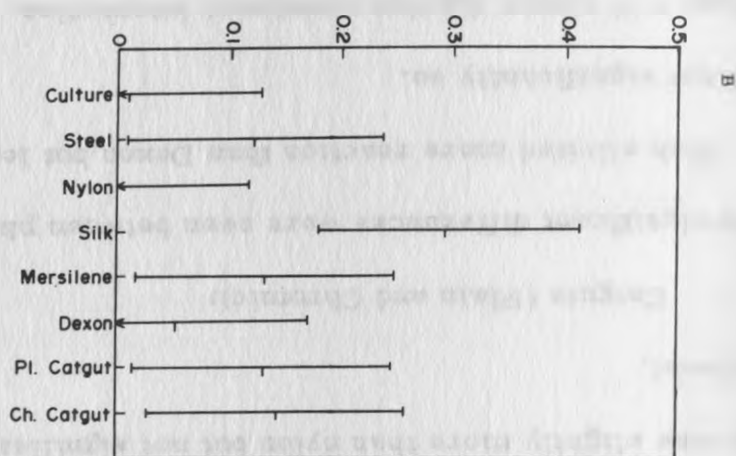
Bars that reach the abscissa (↓) indicate that the mean value is either zero or less than half the simultaneous confidence interval.

Pl. = Plain, Ch. = Chromic.

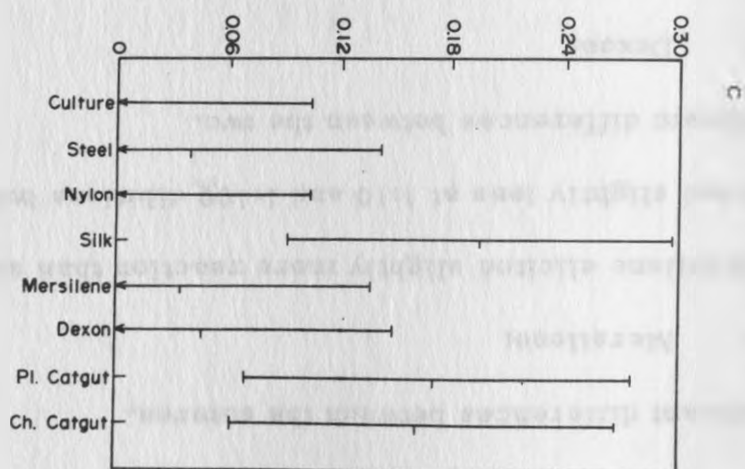
Measurements in centimeters



Measurements in centimeters



Measurements in centimeters



more reaction than nylon and Dexon. At 1:100 dilution there were no significant differences between the sutures.

5. Mersilene:

Mersilene elicited slightly more reaction than silk at 1:1 dilution and slightly less at 1:10 and 1:100 dilutions but there were no significant differences between the two.

6. Dexon:

Dexon stimulated comparatively little tissue reaction. The reaction was slightly more than nylon but not significantly so at any dilution level.

7. Catguts (Plain and Chromic):

No significant differences were seen between plain and chromic catgut. Both elicited more reaction than Dexon but less than silk, although not significantly so.

Figure 19 shows dilution-treatment interaction, comparing treatment means at each of the three dilutions of culture at 20 days. It is readily seen that silk elicited comparatively more tissue reaction than other sutures at all levels of inoculation except for Mersilene at 1:1 dilution.

A two-way factorial randomized block analysis of variance for the global view of group C is shown in Table 28. F values were found significant. The mean dilution and treatment values and their pairwise comparisons are shown in Table 29. The corresponding  $Q_d$  and  $Q_t$  values are also shown.

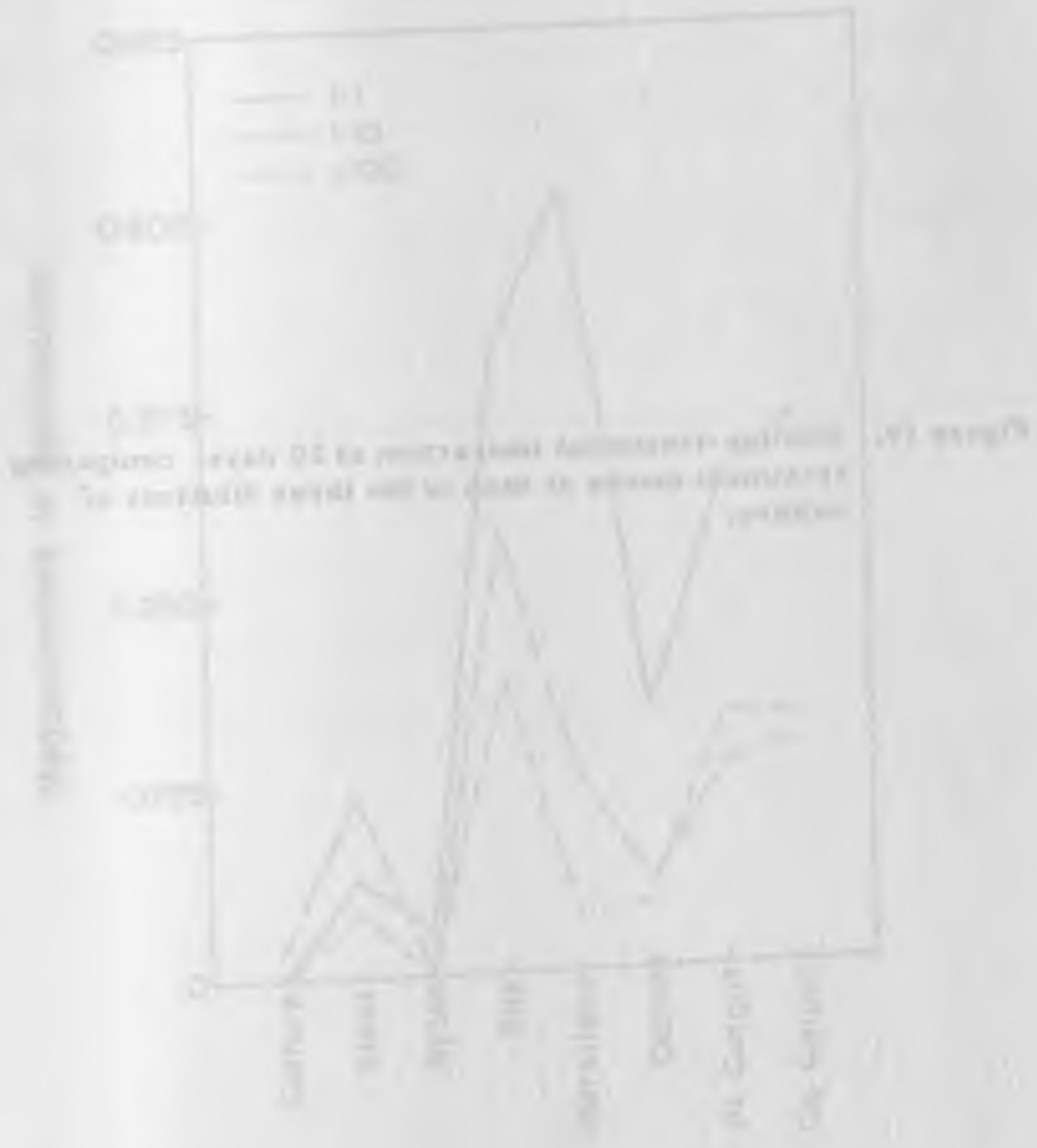
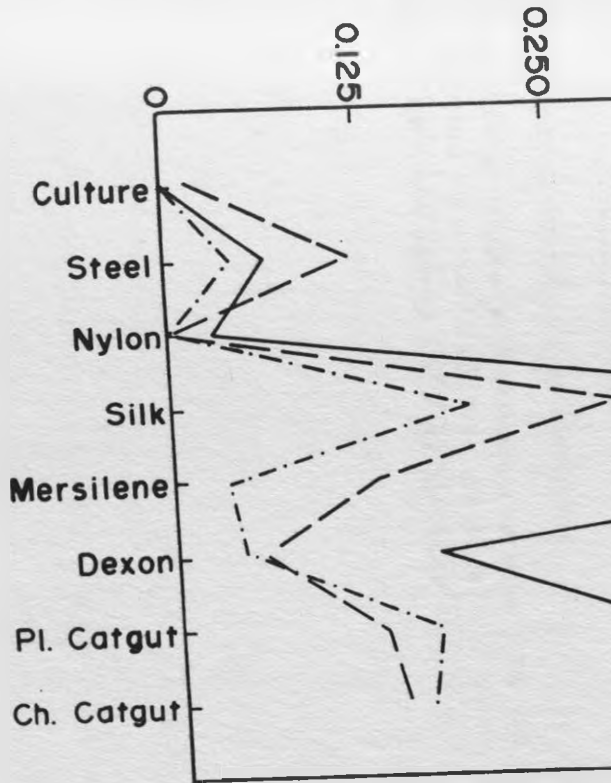


Figure 19. Dilution-treatment interaction at 20 days, comparing treatment means at each of the three dilutions of culture.

# Measurements





in centimeters

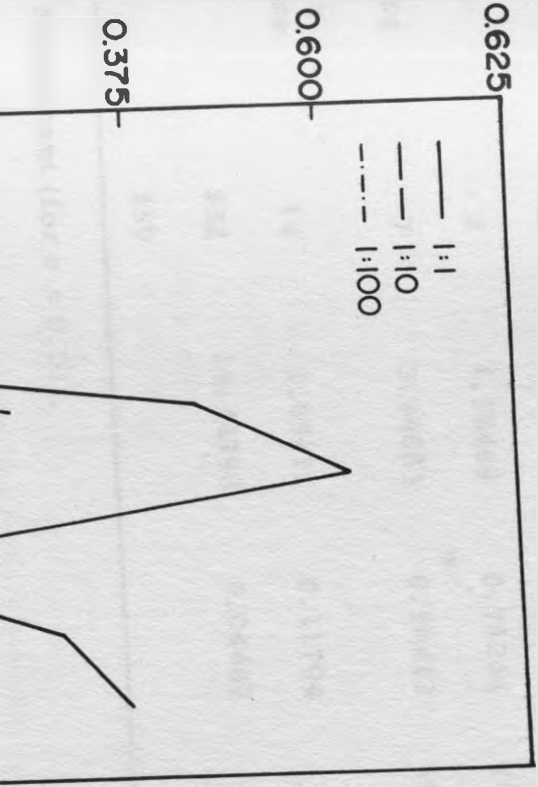


Table 28. Two-Way Factorial Randomized Block Analysis of Variance for the Global View of Group C (20 Days).

Source of Variation	Degrees of Freedom	Sums of Squares	Mean Squares	F
Dogs	14	3.25868	0.23276	
Dilutions	2	1.58489	0.79244	17.661***
Treatments	7	3.94883	0.56412	12.572***
Dilutions x Treatments	14	1.65111	0.11794	2.629***
Error	322	14.44798	0.04487	
Total	359			

\*\*\* Significant (for  $\alpha = 0.01$ ).

Table 29. Dilution and Treatment Means for the Global View with  $Q_d$  and  $Q_t$  Values for Group C (20 Days).

Means*		
<u>Dilutions</u>		
1:1	0.23458	$Q_d = 3.32 \sqrt{\frac{0.04487}{120}}$ $= 0.064$
1:10	0.11125	
1:100	0.08125	
<u>Treatments</u>		
Culture	0.00444	$Q_t = 4.286 \sqrt{\frac{0.04487}{45}}$ $= 0.135$
Nylon	0.00889	
Steel	0.07556	
Dexon	0.09000	
Plain Catgut	0.20667	
Chromic Catgut	0.22333	
Mersilene	0.22667	
Silk	0.30333	

\* Lesion measurements in centimeters.

Any two means whose differences are less than the  $Q_d$  or  $Q_t$  value are connected by a vertical bar. Any pair of means enclosed by the range of any one bar is not significantly different.

Graphic representation of dilution means, with  $Q_d$  range, is shown in Figure 20 A. There were no statistically significant differences between reactions at 1:10 and 1:100 dilutions, but these two were statistically different from the 1:1 dilution level.

Figure 20 B shows overall treatment means with  $Q_t$  range. Pattern of reaction correlates well with reactions at individual dilutions. Silk elicited the most intense reaction. Mersilene and catguts produced slightly less reaction but not significantly different from silk. Dexon produced significantly less reaction than silk. Nylon elicited the least reaction and reaction to steel was between nylon and Dexon.

#### Gross Measurements of Lesions and Statistical Tests for Group D (40 Days)

Measurements of lesion diameters in centimeters in the 1:1, 1:10 and 1:100 dilution rows in each dog in group D are given in Tables 30, 31 and 32.

Results for one-way randomized block analysis of variance for each dilution are shown in Table 33. F values were found significant, at the 1 percent value, for treatments at all three dilutions.

Table 34 shows lesion means and their pairwise comparisons at each dilution. The corresponding  $Q_t$  values are also given. Graphic representation of this is shown in Figure 21 A, B and C.



Figure 20. Global comparison of the three dilutions and the eight treatments at 20 days.

A. Dilutions

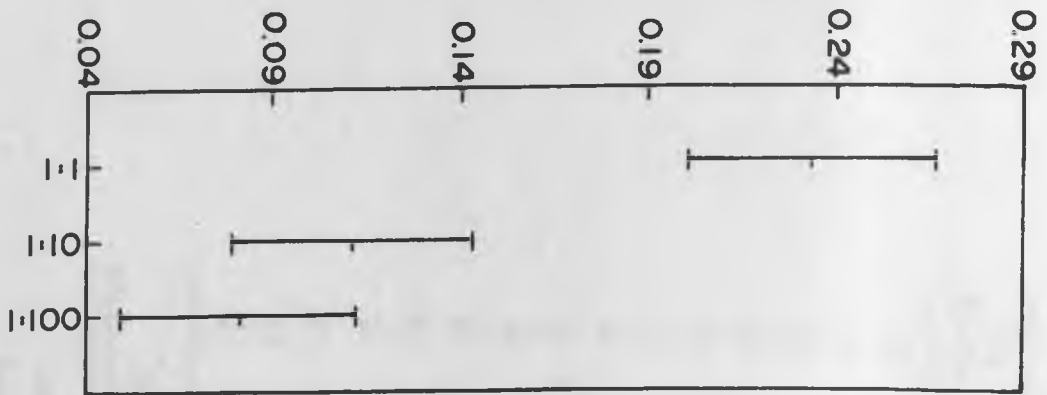
B. Treatments

Bars indicate simultaneous confidence intervals. The center of each bar is the mean value. Treatments whose simultaneous confidence intervals overlap are not statistically significant.

Bars that reach the abscissa (↓) indicate that the mean value is either zero or less than half the simultaneous confidence interval.

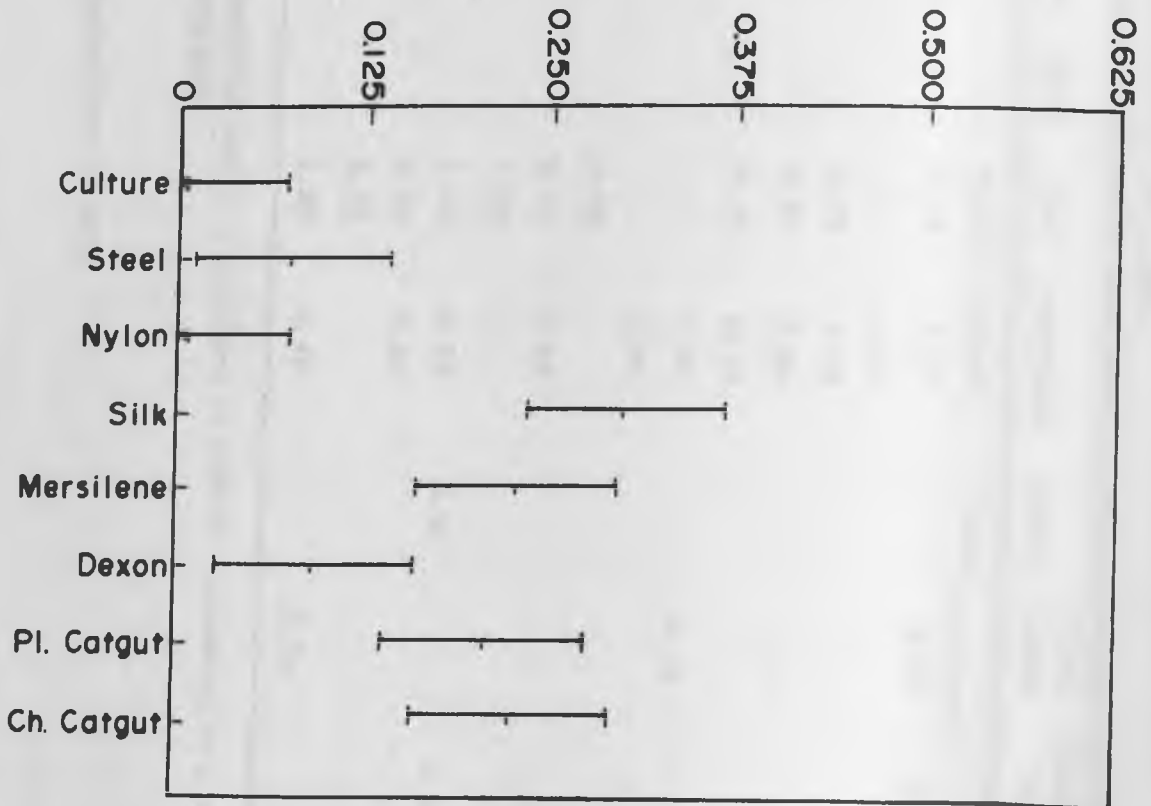
Pl. = Plain, Ch. = Chromic.

Measurements in centimeters



A

Measurements in centimeters



B

Table 30. Gross Measurements of Lesion Diameters in Centimeters in the 1:1 Dilution Rows of Group D (40 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
10				0.55	0.65	1.00		
17		0.25		0.60	0.65		0.40	0.35
15					0.80			
23				0.70	0.60			
14				0.60	0.80			
52				0.35	0.35			
55					0.60		0.30	
37					0.80			
41				0.60				
61		0.45		0.65	0.50			
66				1.00				
67				1.10	0.80	0.45		0.55
77				0.85	0.60			
82				0.50				
83	a			0.80	0.40		0.70	

\* Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^6$  Staphylococcus aureus organisms.

<sup>a</sup> Leaking pocket.

Blanks indicate a measurement of zero.



Table 31. Gross Measurements of Lesion Diameters in Centimeters in the 1:10 Dilution Rows of Group D (40 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
10				0.65	0.45			
17					0.35		0.45	
15				0.40	0.60			
23				0.90				
14					0.60		0.60	
52				0.70				
55				0.75				
37				0.80				0.55
41		a		0.35				
61				0.65	0.75		0.15	0.15
66				0.55	0.70			
67				0.75				
77				0.75				
82				0.60	0.60			
83				0.70				

\* Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^5$  Staphylococcus aureus organisms.

<sup>a</sup> Leaking pocket.

Blanks indicate a measurement of zero.

Table 32. Gross Measurements of Lesion Diameters in Centimeters in the 1:100 Dilution Rows of Group D (40 Days).\*

Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chronic Catgut
10								
17								
15								
23								
14								
52				0.60				
55				0.45				
37								
41				0.55	0.50			0.20
61				0.70				
66								
67				0.50			0.70	
77								
82								
83								

\* Each pocket was inoculated with 0.1 milliliter of culture, containing  $74 \times 10^4$  Staphylococcus aureus organisms.

Blanks indicate a measurement of zero.

Table 33. One-Way Randomized Block Analysis of Variance for Treatment Means at Each Level of Inoculation for Group D (40 Days).

	Source of Variation	Degrees of Freedom	Sums of Squares	Mean Squares	F
1:1 Dilution	Dogs	14	0.86904	0.06207	
	Treatments	7	5.31625	0.75946	17.427***
	Error	98	4.27062	0.04358	
	Total	119			
1:10 Dilution	Dogs	14	0.19854	0.01418	
	Treatments	7	4.34358	0.62051	20.398***
	Error	98	2.98079	0.03042	
	Total	119			
1:100 Dilution	Dogs	14	0.35988	0.02571	
	Treatments	7	0.42767	0.06110	4.128***
	Error	98	1.45046	0.01480	
	Total	119			

\*\*\* Significant (for  $\alpha = 0.01$ ).

Table 34. Treatment Means for Each Level of Inoculation with the Corresponding  $Q_t$  Values for Group D (40 Days).

Treatments	Means*	
		<u>1:1 Dilution</u>
Culture	0.00000	
Nylon	0.00000	
Steel	0.04667	$Q_t = 4.389 \sqrt{\frac{0.04358}{15}}$ $= 0.237$
Chromic Catgut	0.06000	
Plain Catgut	0.09333	
Dexon	0.09667	
Mersilene	0.50333	
Silk	0.55333	
Culture	0.00000	
Steel	0.00000	
Nylon	0.00000	$Q_t = 4.389 \sqrt{\frac{0.03042}{15}}$ $= 0.198$
Dexon	0.00000	
Chromic Catgut	0.04667	
Plain Catgut	0.08000	
Mersilene	0.27000	
Silk	0.57000	
Culture	0.00000	
Steel	0.00000	
Nylon	0.00000	$Q_t = 4.389 \sqrt{\frac{0.01480}{15}}$ $= 0.138$
Dexon	0.00000	
Chromic Catgut	0.01333	
Mersilene	0.03333	
Plain Catgut	0.04667	
Silk	0.18667	

\* Lesion measurements in centimeters.

Any two means whose differences are less than the  $Q_t$  value are connected by a vertical bar. Any pair of means enclosed by the range of any one bar is not significantly different.



Figure 21. Comparison of treatments at each dilution of culture at 40 days.

A. At 1:1 dilution

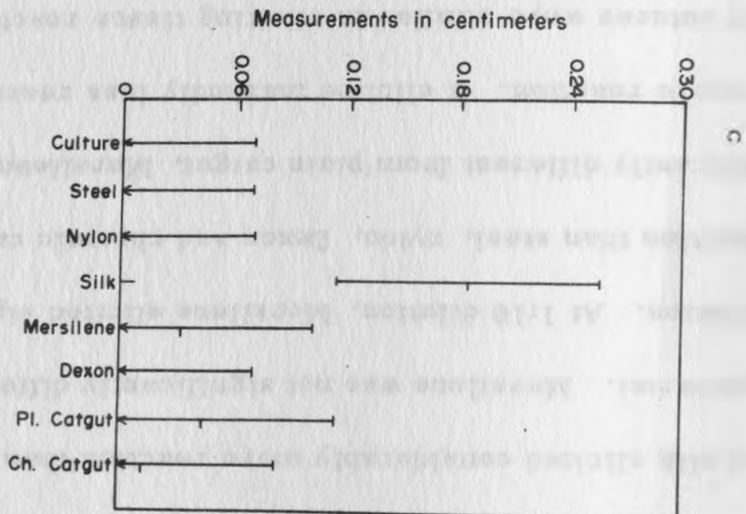
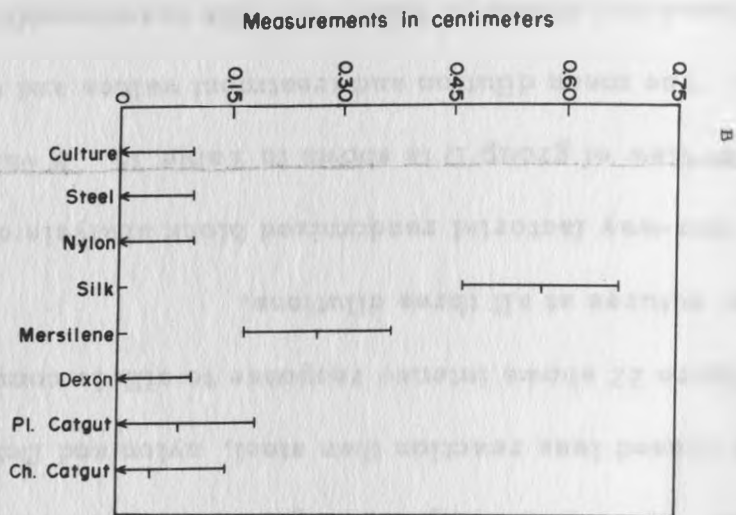
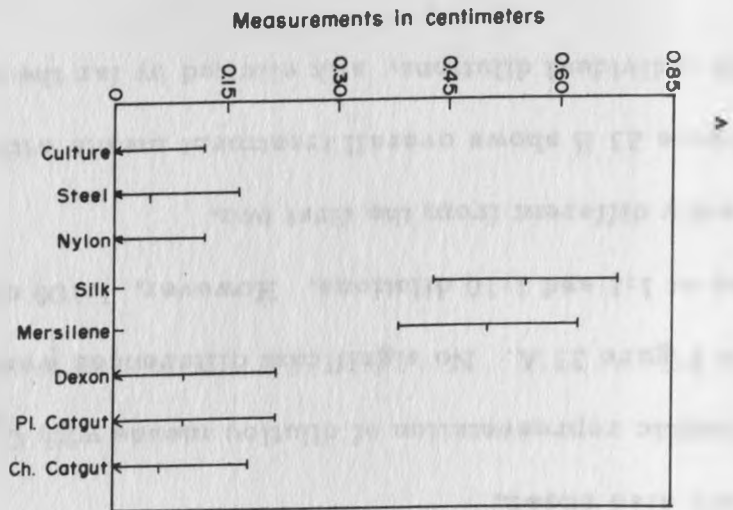
B. At 1:10 dilution

C. At 1:100 dilution

Bars indicate simultaneous confidence intervals. The center of each bar is the mean value. Treatments whose simultaneous confidence intervals overlap are not statistically significant.

Bars that reach the abscissa (↓) indicate that the mean value is either zero or less than half the simultaneous confidence interval.

Pl. = Plain, Ch. = Chromic.



The only significant findings in this group were that at all three dilutions silk elicited considerably more reaction than any other suture material. Mersilene was not significantly different from silk at 1:1 dilution. At 1:10 dilution, Mersilene elicited significantly more reaction than steel, nylon, Dexon and chromic catgut. Although not significantly different from plain catgut, Mersilene caused slightly more reaction. It elicited markedly less reaction than silk. All other sutures were similar in eliciting tissue reactions at all dilutions. Mersilene and plain catgut were intermediate between silk and caused less reaction than steel, nylon and Dexon.

Figure 22 shows intense response to silk in comparison with the other sutures at all three dilutions.

A two-way factorial randomized block analysis of variance for the global view of group D is shown in Table 35. F values were significant. The mean dilution and treatment values and their pairwise comparisons are shown in Table 36. The corresponding  $Q_d$  and  $Q_t$  values are also shown.

Graphic representation of dilution means with  $Q_d$  range is shown in Figure 23 A. No significant differences were seen between reactions at 1:1 and 1:10 dilutions. However, 1:100 dilution was significantly different from the first two.

Figure 23 B shows overall treatment means with  $Q_t$  range. As seen with individual dilutions, silk elicited by far the most intense



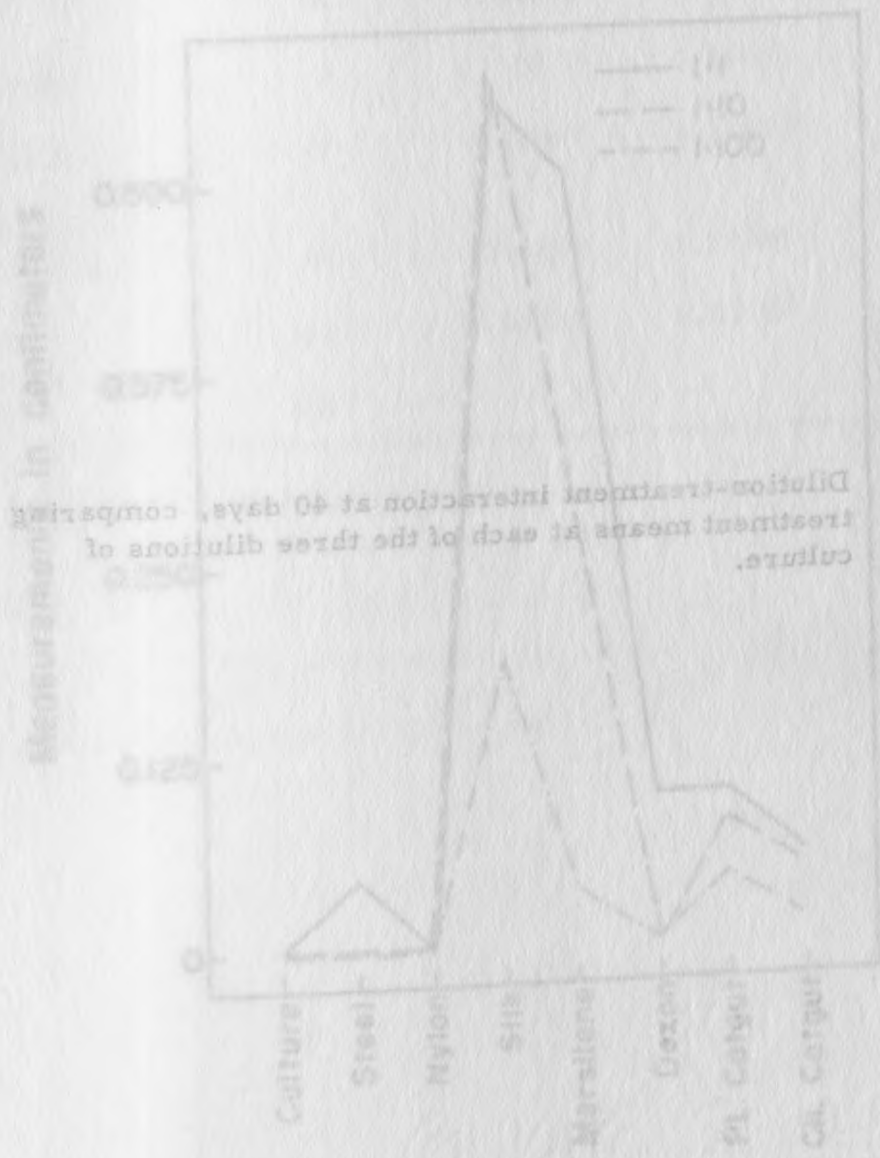


Figure 23. Dilution-treatment interaction at 40 days, comparing treatment means at each of the three dilutions of culture.

Figure 22. Dilution-treatment interaction at 40 days, comparing treatment means at each of the three dilutions of culture.

### Measurements in centimeters

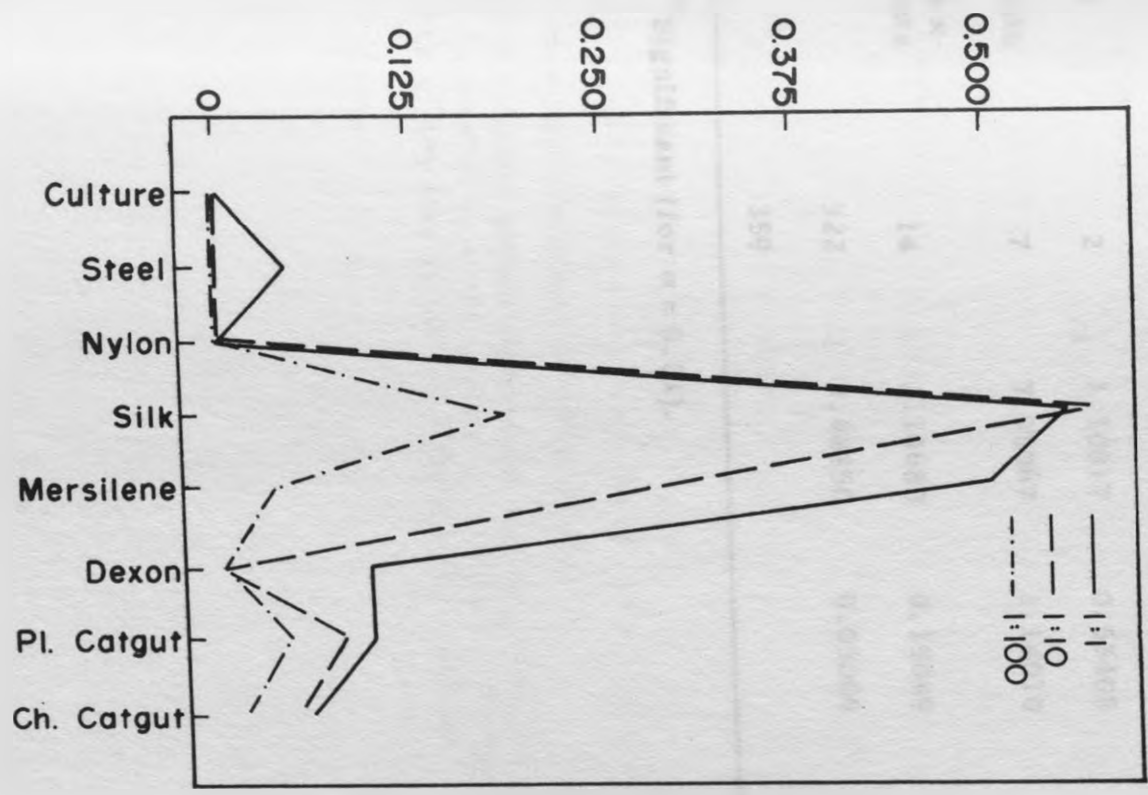


Table 35. Two-Way Factorial Randomized Block Analysis of Variance for the Global View of Group D (40 Days).

Source of Variation	Degrees of Freedom	Sums of Squares	Mean Squares	F
Dogs	14	0.44083	0.03149	
Dilutions	2	1.10817	0.55408	18.415***
Treatments	7	7.98067	1.14010	37.892***
Dilutions x Treatments	14	2.10683	0.15049	5.002***
Error	322	9.68850	0.03009	
Total	359			

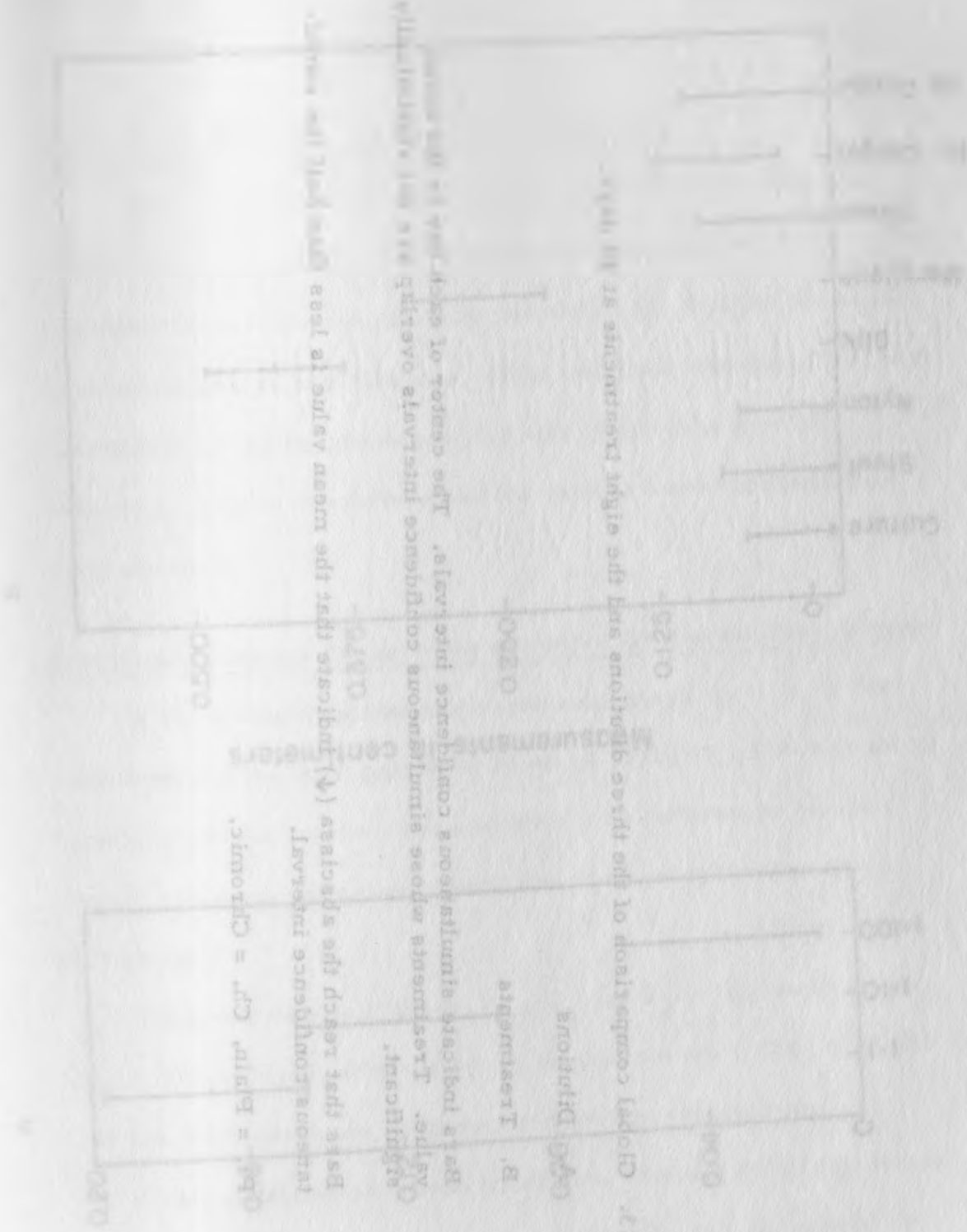
\*\*\* Significant (for  $\alpha = 0.01$ ).

Table 36. Dilution and Treatment Means for the Global View with  $Q_d$  and  $Q_t$  Values for Group D (40 Days).

Means*		
<u>Dilutions</u>		
1:1	0.16917	$Q_d = 3.32 \sqrt{\frac{0.03009}{120}}$ $= 0.053$
1:10	0.12083	
1:100	0.03500	
<u>Treatments</u>		
Culture	0.00000	$Q_t = 4.286 \sqrt{\frac{0.03009}{120}}$ $= 0.111$
Nylon	0.00000	
Steel	0.01556	
Dexon	0.03222	
Chromic Catgut	0.04000	
Plain Catgut	0.07333	
Mersilene	0.26889	
Silk	0.43667	

\* Lesion measurements in centimeters.

Any two means whose differences are less than the  $Q_d$  or  $Q_t$  value are connected by a vertical bar. Any pair of means enclosed by the range of any one bar is not significantly different.



0.20  
0.15  
0.10  
0.05  
0.00

0.500  
0.375  
0.250  
0.125  
0

B' I. resubstrate  
C' D. resubstrate

Control  
100  
200  
300  
400  
500

↑ indicates that the mean value is less than zero (p < 0.05)

↑ indicates that the mean value is significantly different from zero (p < 0.05)

↑ indicates that the mean value is significantly different from the control (p < 0.05)

Figure 17. Chopel comparison of the three questions and the eight treatments at 1000h.

Figure 23. Global comparison of the three dilutions and the eight treatments at 40 days.

A. Dilutions

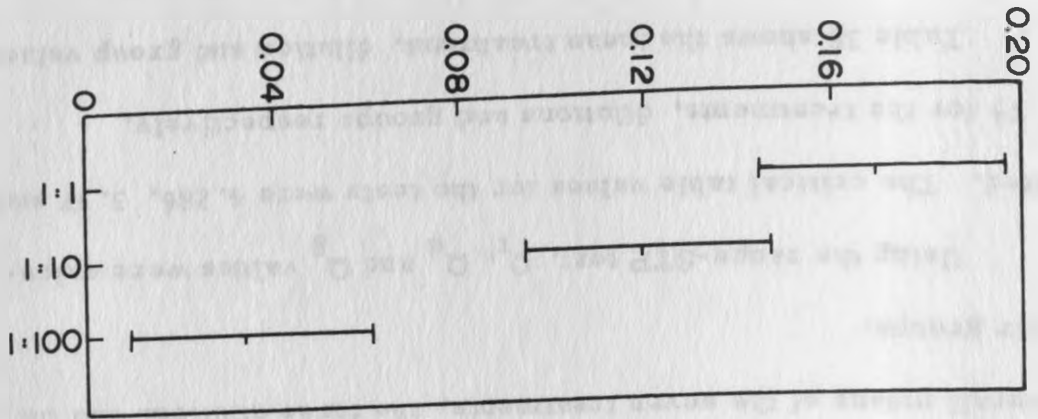
B. Treatments

Bars indicate simultaneous confidence intervals. The center of each bar is the mean value. Treatments whose simultaneous confidence intervals overlap are not statistically significant.

Bars that reach the abscissa (↓) indicate that the mean value is less than half the simultaneous confidence interval.

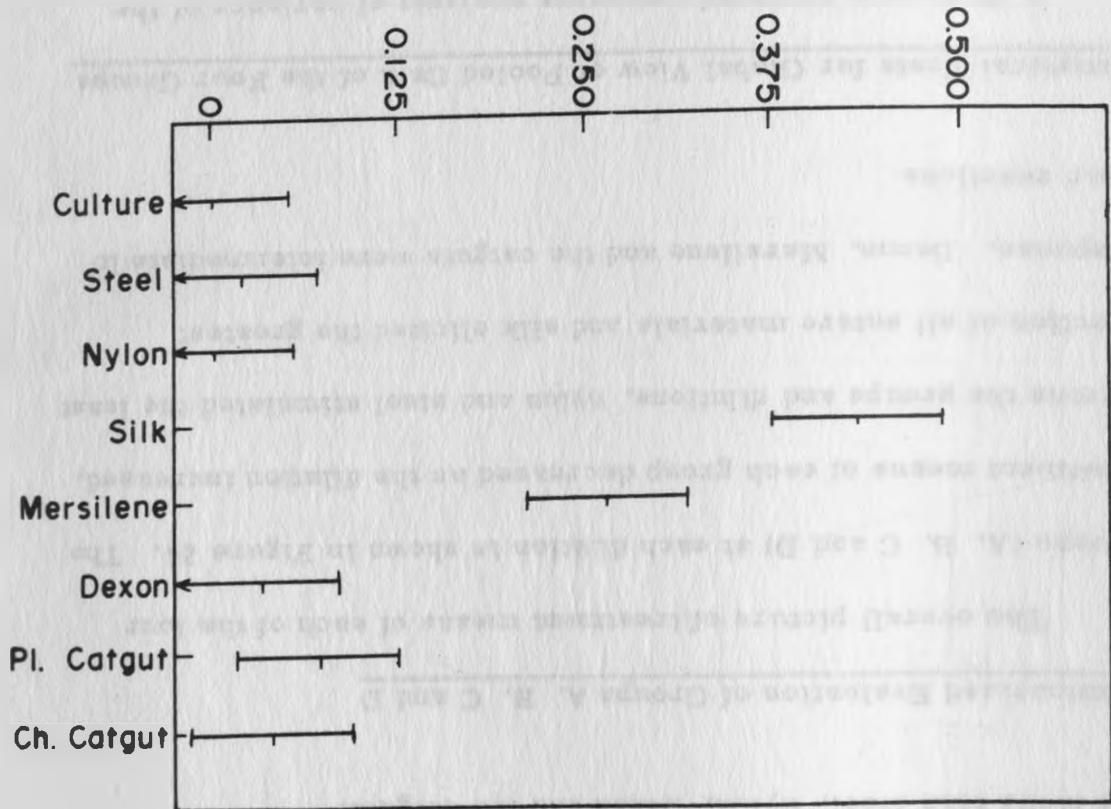
Pl. = Plain, Ch. = Chromic.

Measurements in centimeters



A

Measurements in centimeters



B



reaction. Mersilene stimulated significantly less reaction than silk but more than steel, nylon, Dexon and the catguts.

#### Summarized Evaluation of Groups A, B, C and D

The overall picture of treatment means of each of the four groups (A, B, C and D) at each dilution is shown in Figure 24. The treatment means of each group decreased as the dilution increased. Across the groups and dilutions, nylon and steel stimulated the least reaction of all suture materials and silk elicited the greatest response. Dexon, Mersilene and the catguts were intermediate in their reactions.

#### Statistical Tests for Global View of Pooled Data of the Four Groups

A three-way repeated measures analysis of variance of the pooled data of the four groups is shown in Table 37. F values were found significant, at the 1 percent level, for differences between overall means of the seven treatments, the three dilutions and the four groups.

Using the range-STP test,  $Q_t$ ,  $Q_d$  and  $Q_g$  values were calculated. The critical table values for the tests were 4.286, 3.37 and 3.75 for the treatments, dilutions and groups respectively.

Table 38 shows the mean treatment, dilution and group values with their pairwise comparisons and the  $Q_t$ ,  $Q_d$  and  $Q_g$  values. A graphic representation of this is shown in Figure 25.



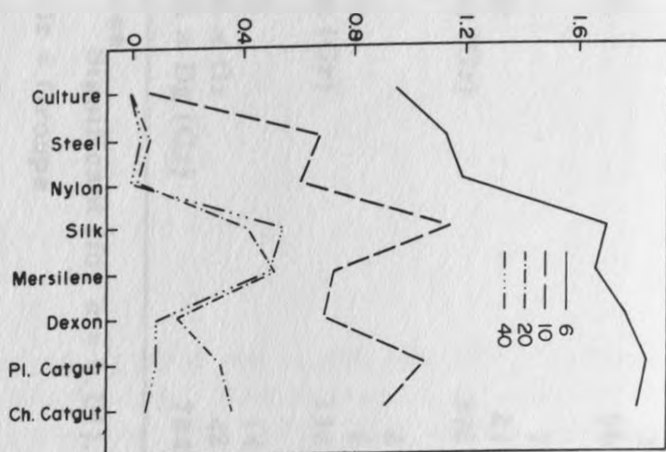
Figure 24. Group-treatment-dilution interaction, comparing treatment means of each of the four groups (6, 10, 20 and 40 days) at each dilution.

A. At 1:1 dilution

B. At 1:10 dilution

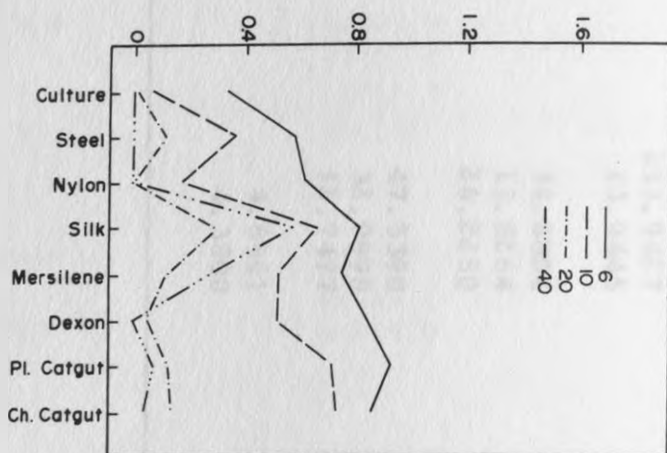
C. At 1:100 dilution

Measurements in centimeters



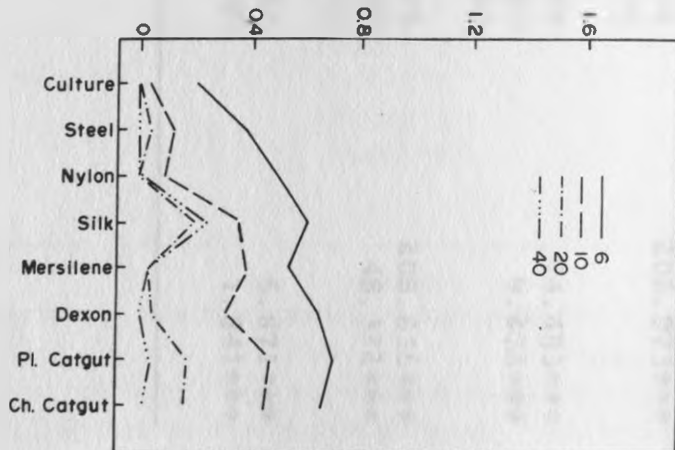
A

Measurements in centimeters



B

Measurements in centimeters



C

Table 37. Three-Way Repeated Measures Analysis of Variance for the Global View of Pooled Data Across the Four Groups.

Source of Variation	Degrees of Freedom	Sums of Squares	Mean Squares	F
Gr	3	153.9427	51.3142	206.073***
Dg (Gr)	56	13.9445	0.2490	
Tr	7	34.8804	4.9829	74.483***
Tr x Gr	21	13.0264	0.6203	9.272***
Tr x Dg (Gr)	392	26.2250	0.0669	
Dl	2	47.5390	23.7695	208.836***
Dl x Gr	6	32.9998	5.5000	48.322***
Dl x Dg (Gr)	112	12.7477	0.1138	
Tr x Dl	14	4.6551	0.3325	5.871***
Tr x Dl x Gr	42	4.3800	0.1043	1.841***
Tr x Dl x Dg (Gr)	784			

\*\*\* Significant (for  $\alpha = 0.01$ ).

Gr = Groups

Dg = Dogs

Tr = Treatments

Dl = Dilutions

Table 38. Treatment, Dilution and Group Means for the Global View of Pooled Data Across the Four Groups, with  $Q_t$ ,  $Q_d$  and  $Q_g$  Values.

Means*		
<u>Treatments</u>		
Culture	0.13944	
Nylon	0.26750	$Q_t = 4.286 \sqrt{\frac{0.06690}{180}}$ $= 0.083$
Steel	0.29361	
Dexon	0.43000	
Mersilene	0.50611	
Chromic Catgut	0.51944	
Plain Catgut	0.54722	
Silk	0.62583	
<u>Dilutions</u>		
1:1	0.66219	$Q_d = 3.37 \sqrt{\frac{0.11382}{480}}$ $= 0.052$
1:10	0.35729	
1:100	0.22896	
<u>Groups</u>		
A (6 Days)	0.91764	$Q_g = 3.75 \sqrt{\frac{0.2440}{360}}$ $= 0.097$
B (10 Days)	0.49225	
C (20 Days)	0.14236	
D (40 Days)	0.10833	

\* Lesion measurements in centimeters.

Any two means whose differences are less than the  $Q_t$ ,  $Q_d$  or  $Q_g$  value are connected by a vertical bar. Any pair of means enclosed by the range of any one bar is not significantly different.

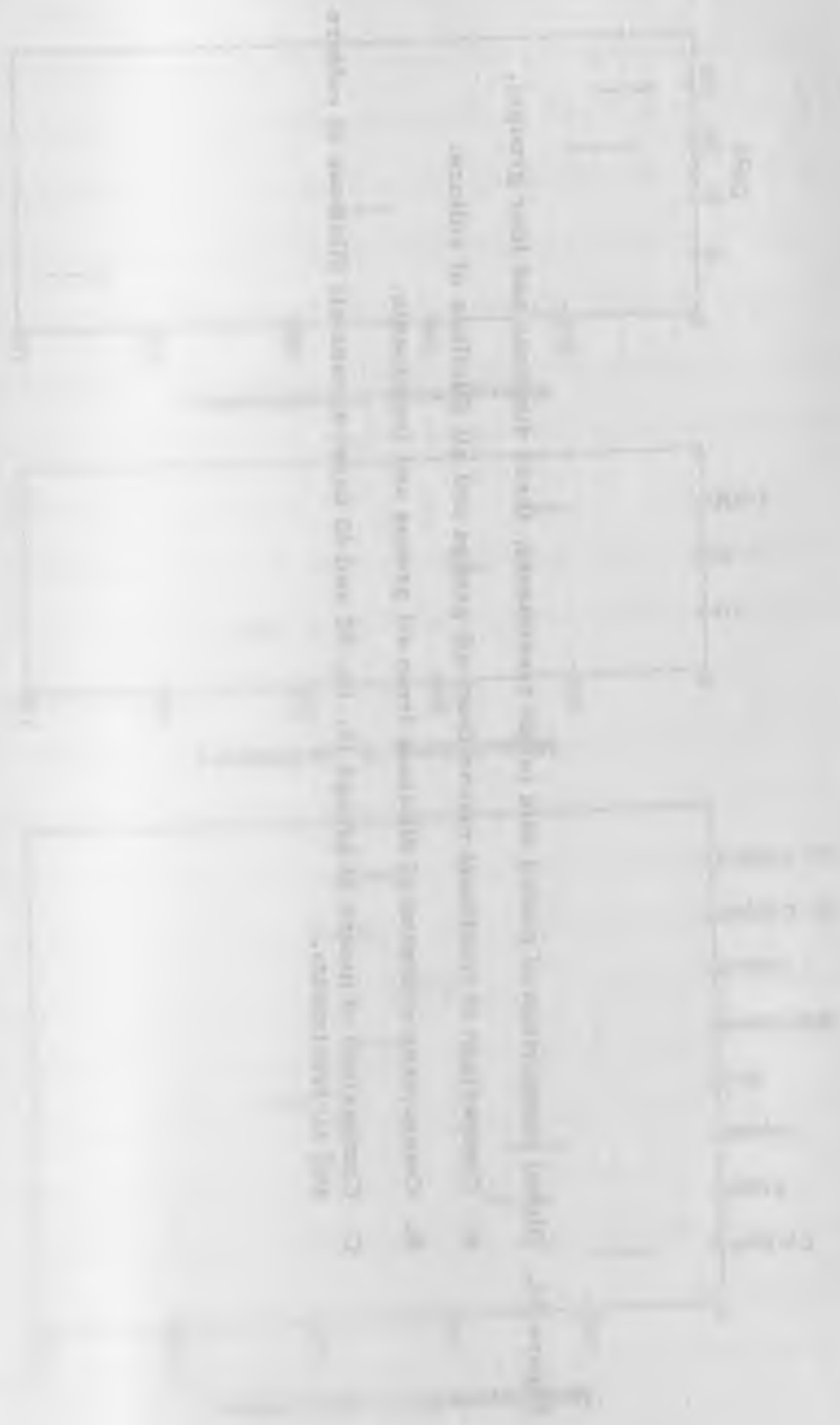


Figure 25. Global comparison of pooled data (eight treatments, three dilutions and four groups).

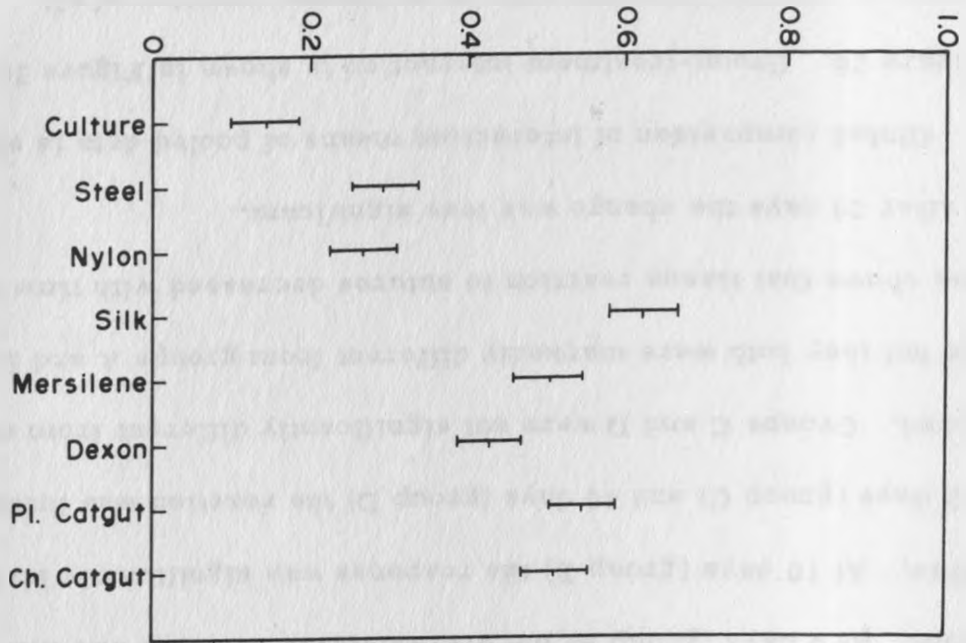
A. Comparison of treatment means from all groups and all dilutions of culture.

B. Comparison of means of dilutions from all groups and treatments.

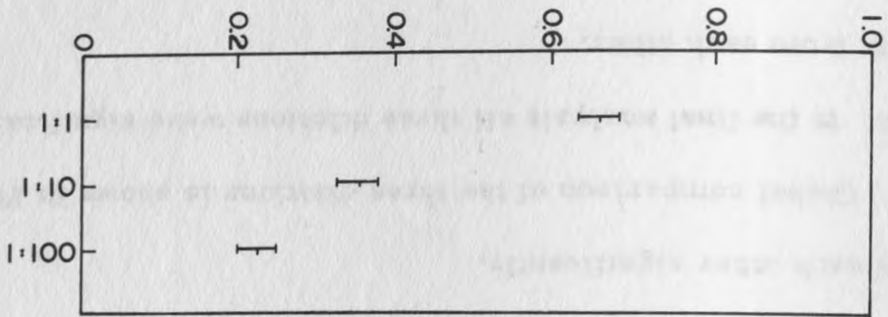
C. Comparison of means of groups (6, 10, 20 and 40 days) across all dilutions of culture and all treatments.



Measurements in centimeters



Measurements in centimeters



Measurements in centimeters

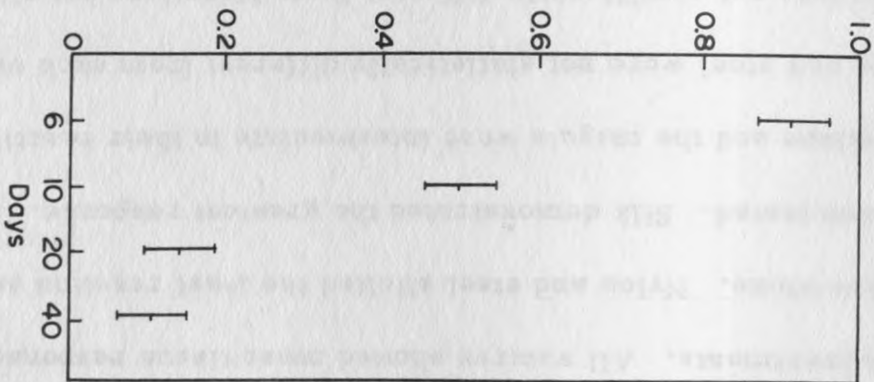


Figure 25 A shows global comparison of pooled data of the eight treatments. All sutures showed more tissue response than culture alone. Nylon and steel elicited the least reaction among the sutures tested. Silk demonstrated the greatest response. Dexon, Mersilene and the catguts were intermediate in their reactions. Nylon and steel were not statistically different from each other. Dexon was not significantly different from Mersilene but elicited less reaction than the catguts. Mersilene and the catguts did not differ from each other significantly.

Global comparison of the three dilutions is shown in Figure 25 B. In the final analysis all three dilutions were significantly different from each other.

Figure 25 C shows comparison of tissue reaction of the four groups. At 6 days (group A) the overall tissue reaction was most intense. At 10 days (group B) the response was significantly less. At 20 days (group C) and 40 days (group D) the reaction was further reduced. Groups C and D were not significantly different from each other but they both were markedly different from groups A and B. It was shown that tissue reaction to sutures decreased with time but that after 20 days the change was less significant.

Global comparison of interaction means of pooled data is shown in Figure 26. Group-treatment interaction is shown in Figure 26 A. In group A, catguts stimulated the most intense reaction of all

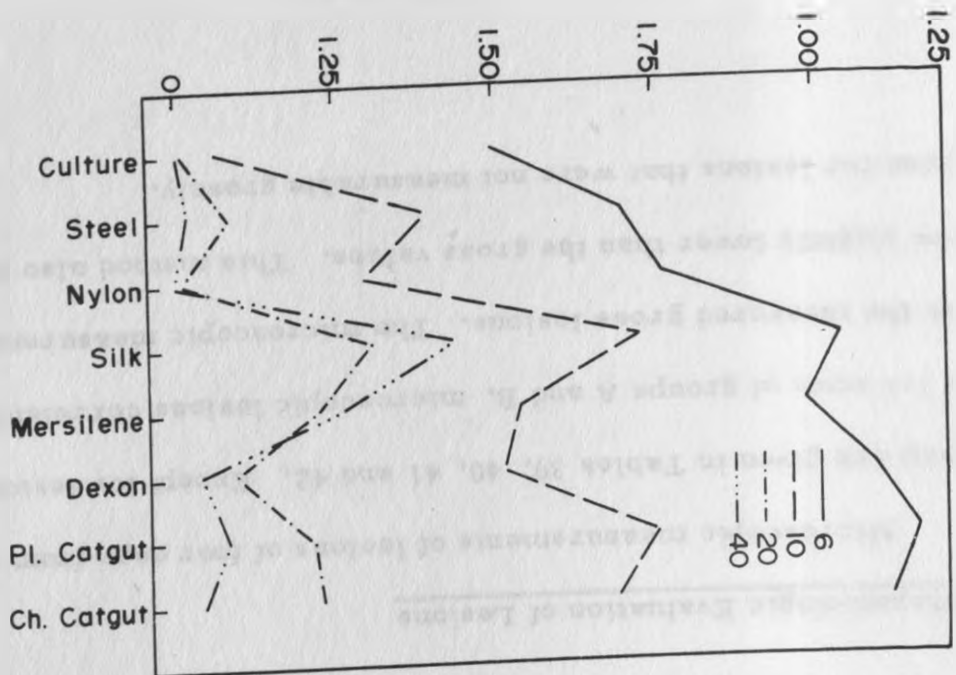


Figure 26. Global comparison of interaction means of pooled data.

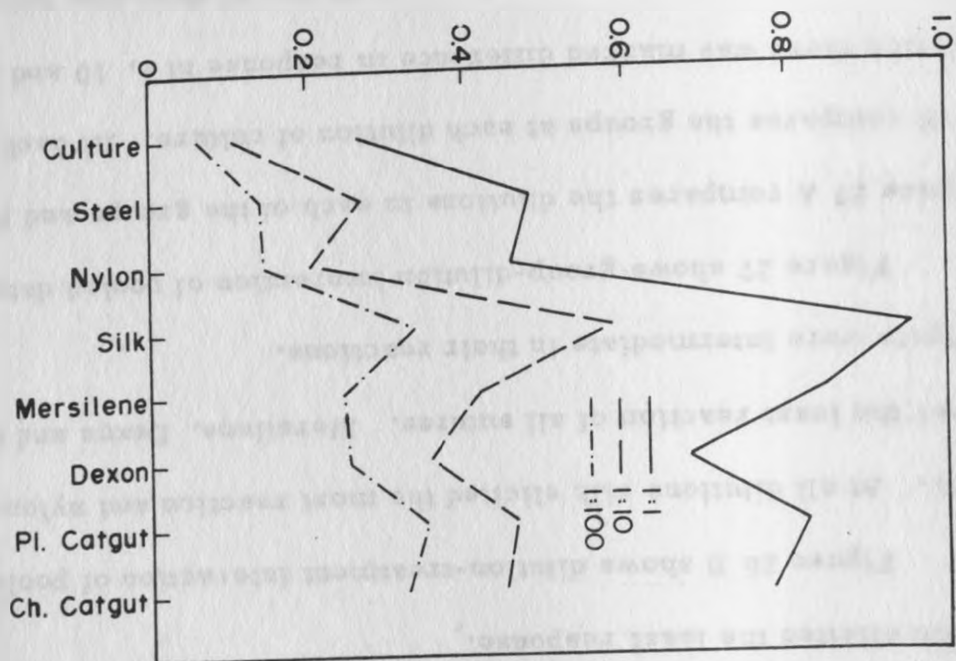
A. Group-treatment interaction

B. Dilution-treatment interaction

Measurements in centimeters



Measurements in centimeters



sutures. Dexon and silk caused slightly less reaction. In groups B, C and D silk elicited the greatest response. In all groups steel and nylon elicited the least response.

Figure 26 B shows dilution-treatment interaction of pooled data. At all dilutions silk elicited the most reaction and nylon and steel the least reaction of all sutures. Mersilene, Dexon and the catguts were intermediate in their reactions.

Figure 27 shows group-dilution interaction of pooled data. Figure 27 A compares the dilutions in each of the groups and Figure 27 B compares the groups at each dilution of culture. At each dilution there was marked difference in response at 6, 10 and 20 days. Difference in response between 20 and 40 days was not significant. Differences between dilutions were marked at 6 and 10 days but were not significant at 20 and 40 days.

#### Histopathologic Evaluation of Lesions

Microscopic measurements of lesions of four dogs from each group are given in Tables 39, 40, 41 and 42. Except for lesions in the 1:1 rows of groups A and B, microscopic lesions correlated well with the measured gross lesions. The microscopic measurements were slightly lower than the gross values. This method also provided values for lesions that were not measurable grossly.

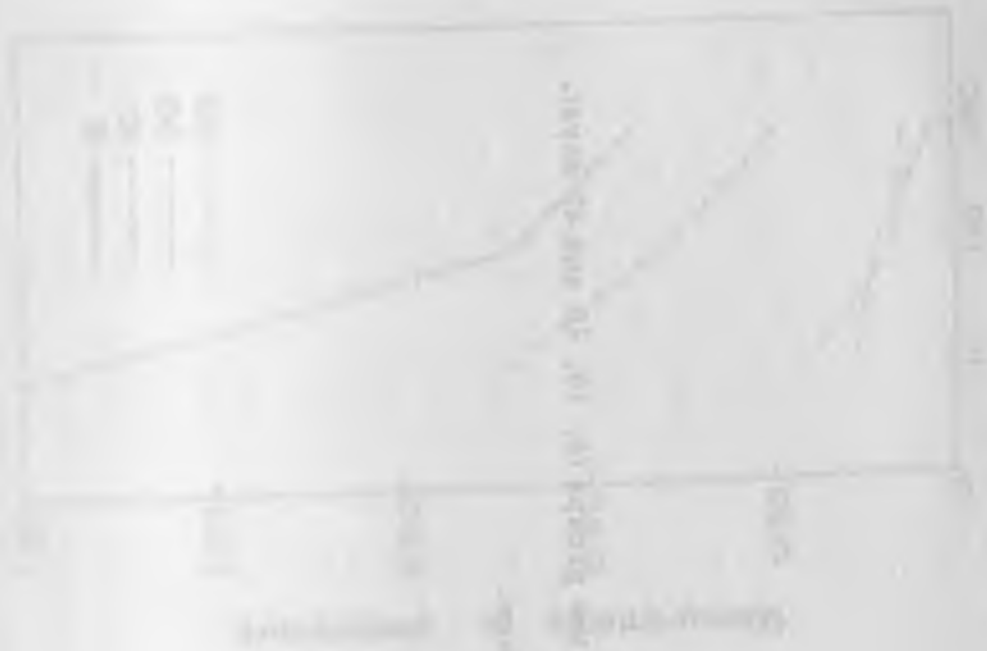


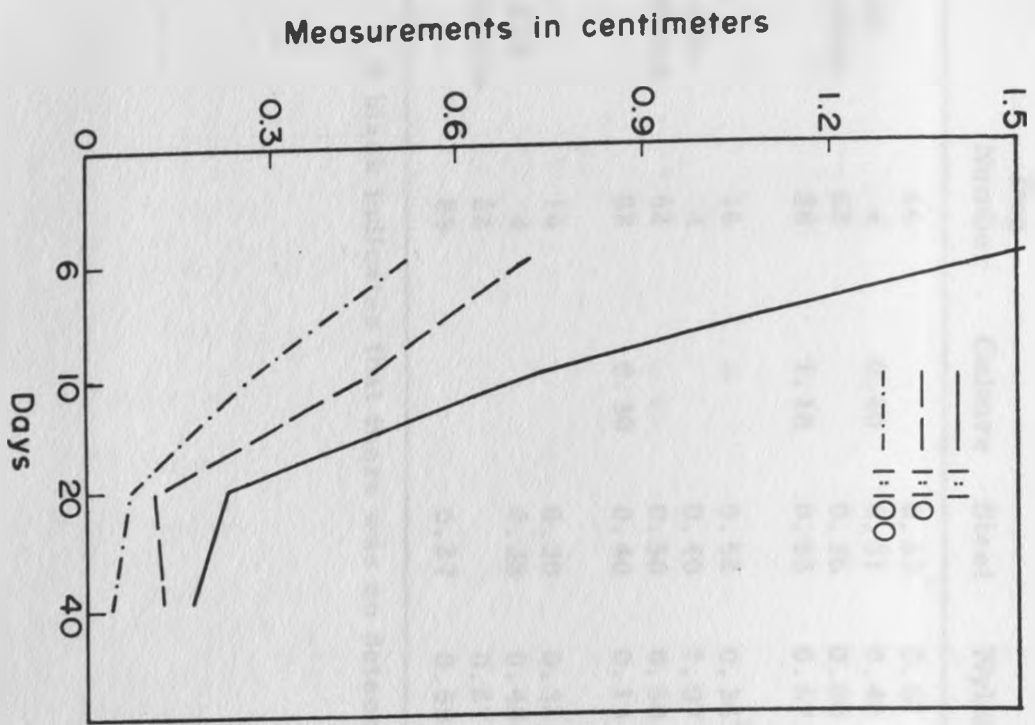
Figure 1: Comparison of two data series over time.

Figure 2: Comparison of two data series over time.

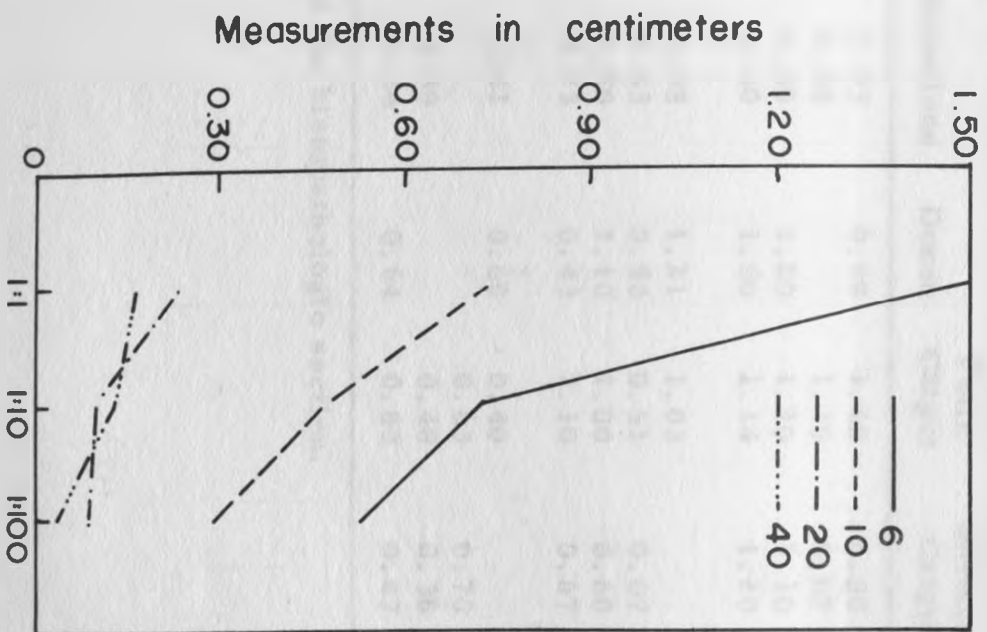
Figure 27. Group-dilution interaction of pooled data.

- A. Comparing all dilutions of culture in each of the groups (6, 10, 20 and 40 days).
- B. Comparing all groups at each dilution of culture.





A



B

Table 39. Microscopic Measurements of Lesions in Centimeters of Group A (6 Days).

	Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
1:1 Dilution	16		0.53	0.62	1.33	0.93	0.68	1.60	0.80
	4	0.40	0.31	0.40	0.62	0.58		1.19	0.67
	62		0.36	0.80	1.10	1.30	1.20	1.20	1.10
	88	1.10	0.53	0.67	1.80	0.40	1.60	1.14	1.20
1:10 Dilution	16		0.52	0.34	0.47	0.58	1.21	1.03	
	4		0.40	1.07	0.67	0.53	0.53	0.51	0.67
	62		0.50	0.50	0.70	0.60	1.10	1.00	0.60
	88	0.30	0.40	0.13	0.47	0.73	0.43	1.10	0.67
1:100 Dilution	16		0.30	0.32	0.53	0.51	0.60	0.40	
	4		0.38	0.48	0.54			0.63	0.70
	62			0.27	0.50	0.39		0.40	0.36
	88		0.27	0.50	0.63	0.34	0.64	0.83	0.67

A blank indicates that there was no detectable lesion in the histopathologic section.

Table 40. Microscopic Measurements of Lesions in Centimeters of Group B (10 Days).

	Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
1:1 Dilution	31		0.40	0.50	0.65	0.31	0.40	0.89	0.80
	40			0.13	1.07	0.43	0.14		0.25
	11		0.22	0.26	0.40	0.40	0.36	0.71	0.76
	76		0.06		0.20	0.21	0.49	0.40	0.53
1:10 Dilution	31		0.51	0.22	0.40	0.46	0.28	0.54	
	40		0.21	0.20	0.60		0.47	0.21	0.40
	11	0.08	0.34	0.44	0.53	0.53	0.46	0.54	0.41
	76		0.23	0.34	0.47	0.01	0.24	0.50	
1:100 Dilution	31		0.08	0.23	0.40	0.40	0.27	0.39	
	40				0.44	0.51	0.20	0.47	0.37
	11				0.60	0.34			0.37
	76			0.22	0.24	0.50	0.38	0.54	0.45

A blank indicates that there was no detectable lesion in the histopathologic section.

Table 41. Microscopic Measurements of Lesions in Centimeters of Group C (20 Days).

	Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
1:1 Dilution	12		0.08	0.31		0.65	0.42	0.13	0.28
	47			0.18	0.64	0.30	0.13	0.38	0.40
	65				0.54	0.52	0.15	0.20	0.52
	71	0.08		0.10	0.60	0.40	0.40		0.55
1:10 Dilution	12	0.13	0.09	0.08	0.19	0.07			
	47		0.22	0.14	0.28	0.52	0.25	0.34	0.53
	65			0.20	0.16		0.26	0.30	0.18
	71			0.20	0.46	0.65		0.44	0.32
1:100 Dilution	12			0.20	0.21	0.18	0.40	0.35	
	47			0.18	0.30	0.36	0.31		
	65		0.13		0.33	0.18		0.20	0.27
	71		0.35	0.27	0.69	0.18			

A blank indicates that there was no detectable lesion in the histopathologic section.

Table 42. Microscopic Measurements of Lesions in Centimeters of Group D (40 Days).

	Dog Number	Culture	Steel	Nylon	Silk	Mersilene	Dexon	Plain Catgut	Chromic Catgut
1:1 Dilution	17		0.13	0.37	0.64	0.60	0.19		
	55		0.08	0.10		0.59		0.32	
	61		0.22		0.70	0.42		0.27	0.33
	83		0.27	0.22	0.45		0.24	0.77	0.31
1:10 Dilution	17			0.26	0.33	0.46		0.87	0.20
	55		0.10	0.21	0.85	0.31	0.39	0.26	0.20
	61			0.01	0.44	0.47	0.18		0.24
	83			0.01	0.56	0.17	0.23	0.24	0.31
1:100 Dilution	17			0.17	0.30	0.39		0.31	0.52
	55				0.40	0.50		0.39	
	61		0.13	0.07	0.60	0.27	0.20		
	83		0.12	0.16	0.53	0.40		0.22	0.22

A blank indicates that there was no detectable lesion in the histopathologic section.

Mean differential cell counts for each treatment at each dilution are given in Tables 43, 44, 45 and 46 for groups A (6 days), B (10 days, C (20 days) and D (40 days) respectively.

Significant observations in histopathologic sections for each treatment are described below.

1. Culture:

Very few histopathologic sections showed detectable lesions when culture was injected alone. No lesions were seen at 40 days. When seen, the inflammatory reaction zone was the smallest of all treatments.

The predominant cells were neutrophils at 6 days. Macrophages and fibroblasts were few at 6 days. They increased at 10 and 20 days, while the relative number of neutrophils decreased. Lymphocytes, eosinophils and plasma cells were occasionally seen.

2. Steel:

Several histopathologic sections were difficult to evaluate as in the process of removing the steel suture from the lesion, the inflammatory reaction around it was disturbed. Generally, the reaction zones were very small.

At 6 days there were less than 50 percent neutrophils at all dilutions. The predominant cells were macrophages, with some fibroblasts. At 10 and 20 days the number of neutrophils decreased and macrophages increased. At 20 days there were less than 10

Table 43. Mean Differential Cell Counts (Percentages) in Histopathologic Sections of Group A (6 Days).\*

		N	M	F	G	L	P	E
1:1 Dilution	Culture	78	11	9	0	1	0	1
	Steel	44	45	10	0	0	0	1
	Nylon	48	34	15	1	1	1	0
	Silk	59	20	15	0	6	0	0
	Mersilene	73	20	4	2	0	1	0
	Dexon	83	3	3	2	4	5	0
	Plain Catgut	63	11	17	0	9	0	0
	Chromic Catgut	63	20	15	1	1	0	0
1:10 Dilution	Culture	78	17	2	1	1	1	0
	Steel	33	48	16	3	0	0	0
	Nylon	60	30	8	2	0	0	0
	Silk	65	14	15	5	1	0	0
	Mersilene	53	21	20	4	2	0	0
	Dexon	69	17	6	0	6	0	2
	Plain Catgut	56	28	16	0	0	0	0
	Chromic Catgut	67	27	6	0	0	0	0
1:100 Dilution	Culture <sup>a</sup>							
	Steel	20	47	31	0	2	0	0
	Nylon	60	15	25	0	0	0	0
	Silk	58	30	5	4	3	0	0
	Mersilene	55	30	12	1	0	0	2
	Dexon	58	12	12	0	18	0	0
	Plain Catgut	56	24	10	2	4	4	0
	Chromic Catgut	62	20	16	0	2	0	0

\* Mean values of five microscopic fields (1125X) in each of the four dogs.

<sup>a</sup> No detectable lesions in the histopathologic sections.

N = Neutrophils, M = Macrophages, F = Fibroblasts, G = Giant Cells, L = Lymphocytes, P = Plasma Cells, E = Eosinophils.

Table 44. Mean Differential Cell Counts (Percentages) in Histopathologic Sections of Group B (10 Days).\*

		N	M	F	G	L	P	E
1:1 Dilution	Culture	55	34	6	0	2	3	0
	Steel	42	40	17	1	0	0	0
	Nylon	15	53	28	2	0	2	0
	Silk	55	30	13	0	2	0	0
	Mersilene	66	20	10	1	1	2	0
	Dexon	52	32	11	1	3	1	0
	Plain Catgut	51	20	25	0	4	0	0
	Chromic Catgut	47	25	22	0	6	0	0
1:10 Dilution	Culture	50	35	15	0	0	0	0
	Steel	13	60	23	2	1	1	0
	Nylon	8	38	48	1	2	2	1
	Silk	60	13	18	2	5	2	0
	Mersilene	30	50	15	5	0	0	0
	Dexon	35	48	12	0	3	1	1
	Chromic Catgut	58	28	10	0	4	0	0
1:100 Dilution	Culture <sup>a</sup>							
	Steel	10	54	32	1	0	2	1
	Nylon	20	30	40	0	10	0	0
	Silk	25	56	15	1	1	2	0
	Mersilene	30	40	25	5	0	0	0
	Dexon	41	29	18	0	12	0	0
	Chromic Catgut	52	24	21	0	3	0	0

\* Mean values of five microscopic fields (1125X) in each of the four dogs.

<sup>a</sup> No detectable lesions in the histopathologic sections.

N = Neutrophils, M = Macrophages, F = Fibroblasts, G = Giant Cells, L = Lymphocytes, P = Plasma Cells, E = Eosinophils.



Table 45. Mean Differential Cell Counts (Percentages) in Histopathologic Sections of Group C (20 Days).\*

		N	M	F	G	L	P	E
1:1 Dilution	Culture	40	40	16	0	0	4	0
	Steel	5	52	38	0	3	1	1
	Nylon	5	58	32	2	0	3	0
	Silk	59	26	12	0	0	2	1
	Mersilene	68	22	10	0	0	0	0
	Dexon	36	46	10	0	6	2	0
	Plain Catgut	42	24	30	1	3	0	0
	Chromic Catgut	40	27	30	0	1	2	0
1:10 Dilution	Culture	28	43	24	0	0	1	4
	Steel	8	58	26	0	3	5	0
	Nylon	3	35	60	2	0	0	0
	Silk	56	23	16	1	3	0	1
	Mersilene	46	42	10	0	0	2	0
	Dexon	28	57	11	1	0	3	0
	Plain Catgut	40	30	21	0	4	5	0
	Chromic Catgut	35	34	23	0	4	4	0
1:100 Dilution	Culture <sup>a</sup>							
	Steel	4	50	43	1	1	0	1
	Nylon	3	39	57	0	0	1	0
	Silk	20	60	16	1	2	0	1
	Mersilene	5	52	34	6	0	3	0
	Dexon	14	60	21	0	2	3	0
	Plain Catgut	38	37	20	0	0	0	5
	Chromic Catgut	33	35	25	0	0	5	2

\* Mean values of five microscopic fields (1125X) in each of the four dogs.

<sup>a</sup> No detectable lesions in the histopathologic sections.

N = Neutrophils, M = Macrophages, F = Fibroblasts, G = Giant Cells, L = Lymphocytes, P = Plasma Cells, E = Eosinophils.

Table 46. Mean Differential Cell Counts (Percentages) in Histopathologic Sections of Group D (40 Days).\*

		N	M	F	G	L	P	E
		Culture <sup>a</sup>						
1:1 Dilution	Steel	1	45	45	0	0	9	0
	Nylon	0	50	49	1	0	0	0
	Silk	31	38	23	1	0	7	0
	Mersilene	40	39	20	0	0	1	0
	Dexon	1	33	57	1	0	8	0
	Plain Catgut	33	37	26	1	2	1	0
	Chromic Catgut	34	34	26	1	0	5	0
		Culture <sup>a</sup>						
1:10 Dilution	Steel	1	40	58	0	0	1	0
	Nylon	3	32	57	0	0	8	0
	Silk	40	36	20	1	0	3	0
	Mersilene	35	38	21	1	1	4	0
	Dexon	1	57	30	1	1	10	0
	Plain Catgut	20	42	34	0	0	4	0
	Chromic Catgut	13	33	44	1	0	9	0
		Culture <sup>a</sup>						
1:100 Dilution	Steel	1	31	67	0	0	1	0
	Nylon	1	28	67	0	0	4	0
	Silk	37	37	20	0	0	6	0
	Mersilene	3	54	33	0	0	10	0
	Dexon	1	44	49	0	0	6	0
	Plain Catgut	3	42	47	1	0	7	0
	Chromic Catgut	4	45	45	1	0	5	0

\* Mean values of five microscopic fields (1125X) in each of the four dogs.

<sup>a</sup> No detectable lesions in the histopathologic sections.

N = Neutrophils, M = Macrophages, F = Fibroblasts, G = Giant Cells, L = Lymphocytes, P = Plasma Cells, E = Eosinophils.

percent neutrophils and at 40 days there were only 1 percent neutrophils at all three dilutions. The predominant cells were fibroblasts. Plasma cells were also seen. Representative photomicrographs at 10 and 40 days are shown in Figure 28.

### 3. Nylon:

Except at 6 days, microscopic measurements for nylon were very similar to those of steel. At 6 days, nylon elicited slightly more reaction.

The predominant cells at 6 days were neutrophils, with less macrophages and fibroblasts and occasional giant cells, lymphocytes, plasma cells and eosinophils. At 10 days there were less than 20 percent neutrophils, the predominant cells being macrophages and fibroblasts. Lymphocytes were more prominent than at 6 days. At 20 and 40 days there were less than 5 percent neutrophils. The reaction consisted mainly of fibroblasts and macrophages and a few plasma cells. Representative photomicrographs at 10 and 40 days are shown in Figure 29.

### 4. Silk:

Silk demonstrated the greatest inflammatory reaction of all sutures at 20 and 40 days. The reaction was similar to, or slightly less than, Dexon and the catguts at 6 days and only the catguts at 10 days.



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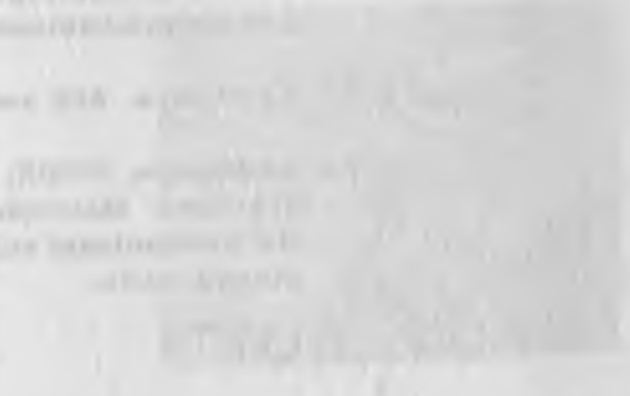


Figure 28. Photomicrographs of tissue reaction to steel at 10 and 40 days, at 1:1 dilution of culture.

A & B. At 10 days, 88X and 312X

C. At 10 days, 2250X, showing the cellular reaction. Neutrophils and macrophages are the predominant cells.

D & E. At 40 days, 88X and 312X

F. At 40 days, 2250X, showing the cellular reaction. Macrophages and fibroblasts are the predominant cells. Also seen are some plasma cells.



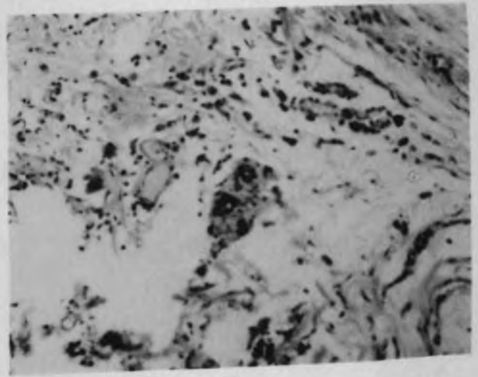
A



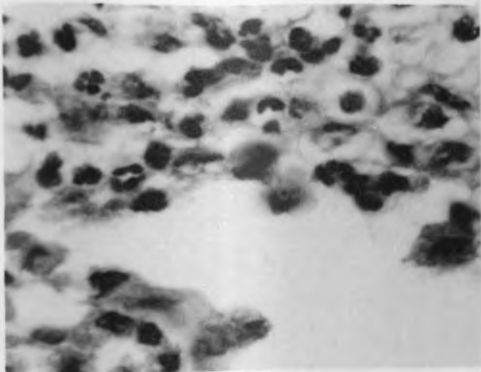
D



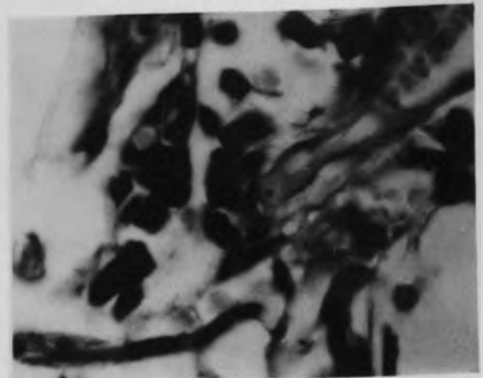
B



E



C



F



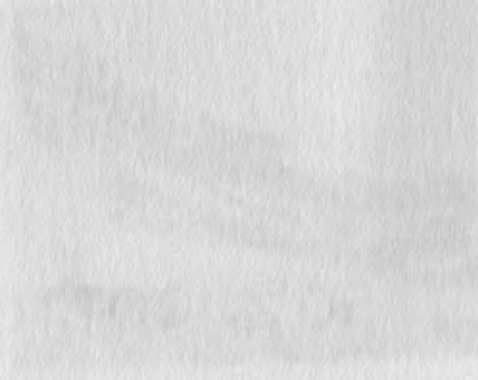
Figure 27. Photomicrographs of tissue reaction to nylon at 10 and 20 days, at 1:1 dilution of culture.

A & B. At 10 days, 28X and 112X

C. At 10 days, 2250X, showing the cellular reaction. Macrophages, fibroblasts and some epithelial cells are seen.

D & E. At 20 days, 88X and 352X, showing a mild fibrous zone.

F & G. At 20 days, 2250X, showing the cellular reaction. The predominant cells are macrophages and fibroblasts.



C

D

Figure 29. Photomicrographs of tissue reaction to nylon at 10 and 40 days, at 1:1 dilution of culture.

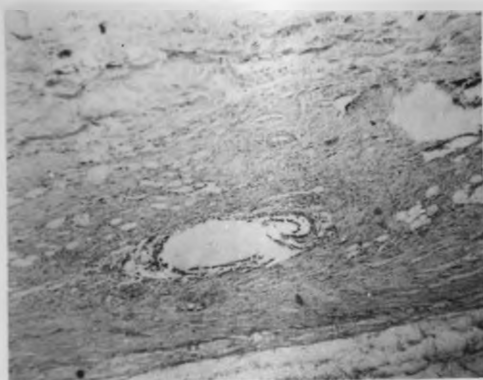
A & B. At 10 days, 88X and 312X

C. At 10 days, 2250X, showing the cellular reaction. Macrophages, fibroblasts and some neutrophils are seen.

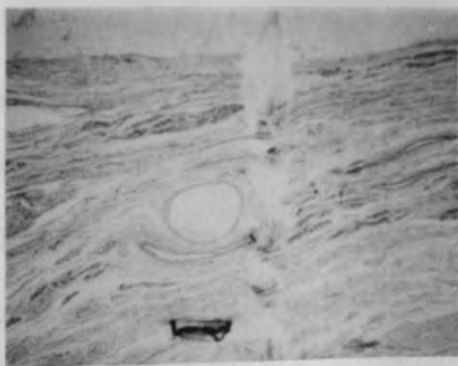
D & E. At 40 days, 88X and 312X, showing a mild reaction zone.

F. At 40 days, 2250X, showing the cellular reaction. The predominant cells are macrophages and fibroblasts.





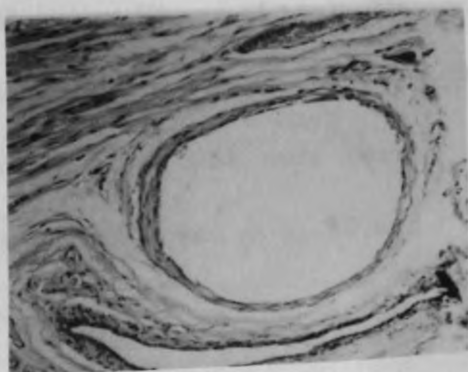
A



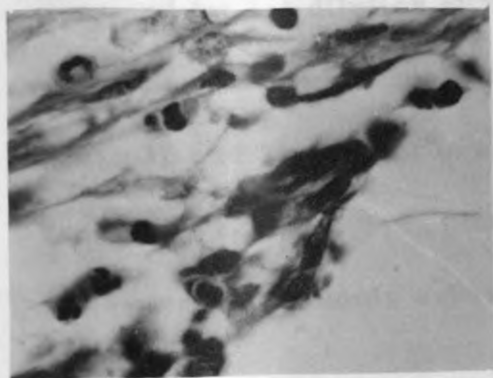
D



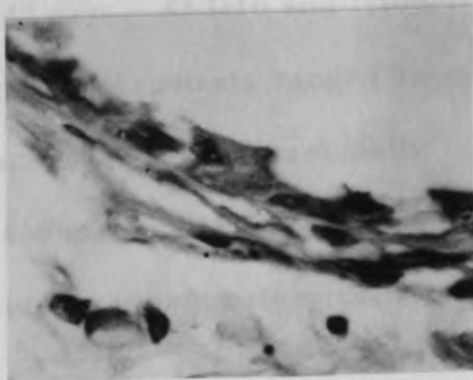
B



E



C



F

Neutrophils were the predominant cells at 6 days. There were fewer macrophages and fibroblasts. Lymphocytes were occasionally seen. The reaction was similar at 10 and 20 days except at 1:100 dilution of culture, where the predominant cells were macrophages. At 40 days there were less than 40 percent neutrophils at all three dilutions. Other predominant cells were macrophages and fibroblasts. Plasma cells ranged between 3 and 7 percent. Inflammatory cells were commonly seen between individual suture fibers. Figure 30 shows representative photomicrographs at 10 and 40 days.

An eosinophilic deposit around individual fibers of multifilament silk suture, similar to the Splendore-Hoeppli phenomenon (SHP), was observed in some sections at 40 days. This was only seen with silk suture. The phenomenon was not observed at 6, 10 and 20 days and was not consistent at 40 days.

#### 5. Mersilene:

Microscopic measurements of lesions with Mersilene were similar to silk.

The predominant cells were neutrophils for all dilutions at 6 days and for 1:1 dilution at 10, 20 and 40 days. At 1:10 and 1:100 dilutions, there were more macrophages. Fibroblasts ranged between 10 and 35 percent in all sections. Other cells occasionally seen were lymphocytes, plasma cells and giant cells. Mersilene fibers were commonly separated by invading inflammatory cells.



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Figure 30. Photomicrographs of tissue reaction to silk at 10 and 40 days, at 1:1 dilution of culture.

A & B. At 10 days, 88X and 312X. Inflammatory reaction around the suture strands is seen.

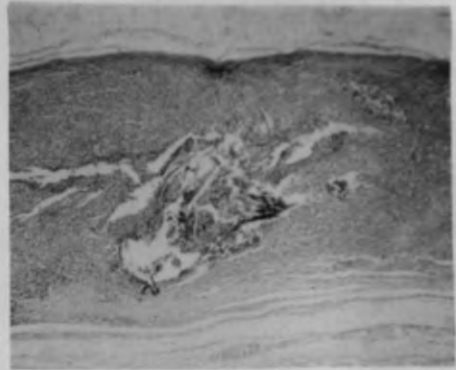
C. At 10 days, 2250X, showing cellular reaction in greater detail. Predominantly neutrophils are seen around the suture.

D & E. At 40 days, 88X and 312X.

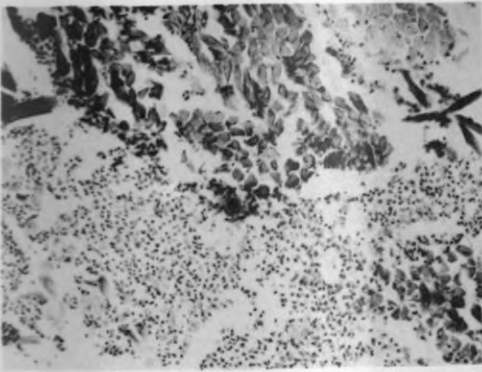
F. At 40 days, 2250X, showing cellular reaction around the suture. Neutrophils, macrophages and fibroblasts are the predominant cells. Occasional plasma cells are also seen.



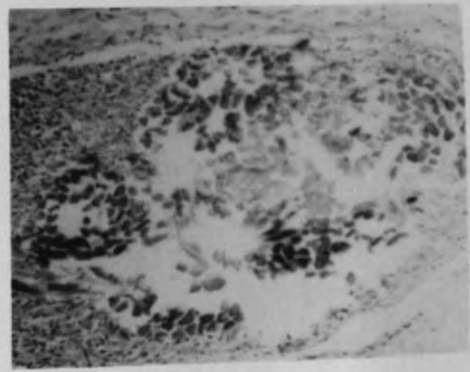
A



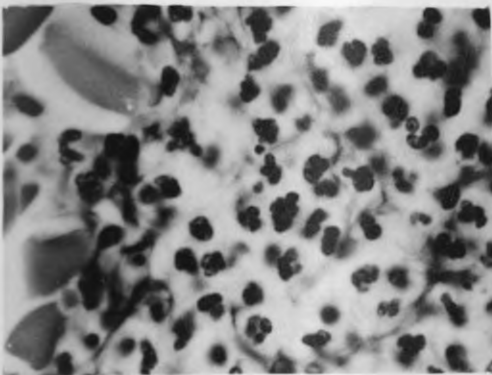
D



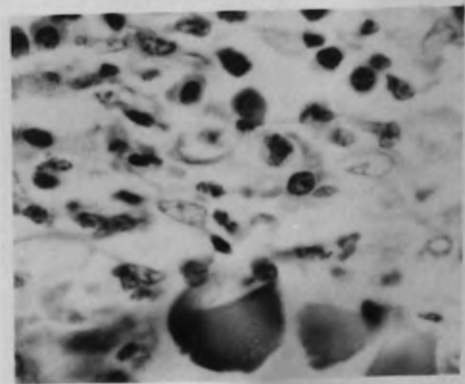
B



E



C



F

Representative photomicrographs at 10 and 40 days are shown in Figure 31.

6. Dexon:

Dexon showed similar reaction to silk and Mersilene at 6 days but generally less reaction at 10, 20 and 40 days.

Neutrophils were the predominant cells at 6 days. Macrophages and fibroblasts together were less numerous than the neutrophils. Lymphocytes were frequently seen. Plasma cells, giant cells and eosinophils were less common. At 10 days the relative number of neutrophils had decreased and macrophages had increased. At 20 days there were more macrophages than neutrophils, and at 40 days there were only 1 percent neutrophils at all three dilutions. Predominant cells were macrophages and fibroblasts. There were between 6 and 10 percent plasma cells at 40 days.

Inflammatory cells were commonly seen between individual suture fibers. The fibers were splitting at 20 days and many fibers were broken, suggesting that they were being absorbed. This was more evident at 40 days when many sections contained no suture material. Representative photomicrographs at 10 and 40 days are shown in Figure 32.

7. Catguts (Plain and Chromic):

Plain and chromic catgut were similar in stimulating tissue reaction at all stages. Plain catgut showed slightly larger reaction



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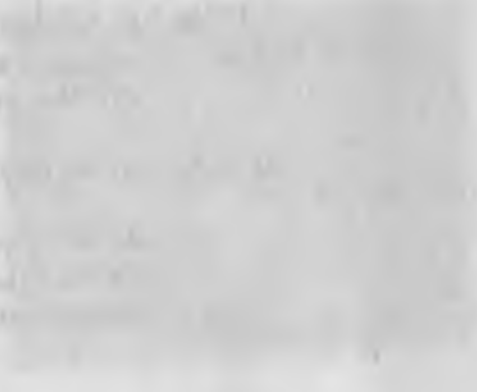


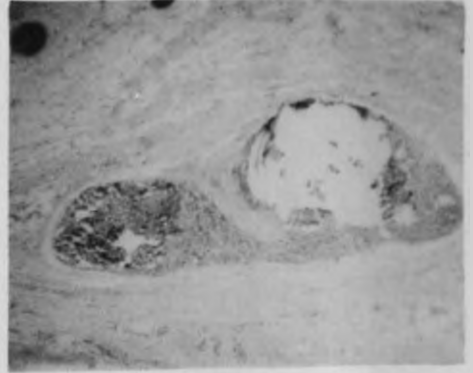
Figure 31. Photomicrographs of tissue reaction to Mersilene at 10 and 40 days, at 1:1 dilution of culture.

- A & B. At 10 days, 88X and 312X. Suture strands are seen in cross-section, with the inflammatory reaction around them.
- C. At 10 days, 2250X, showing cellular reaction between and around suture strands. Neutrophils are the predominant cells.
- D & E. At 40 days, 88X and 312 X.
- F. At 40 days, 2250X, showing cellular reaction around the suture strands. Macrophages and fibroblasts predominate, with some neutrophils.

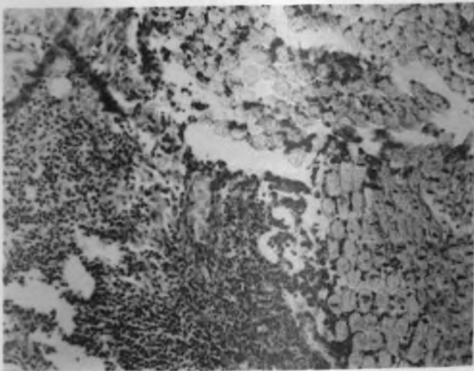




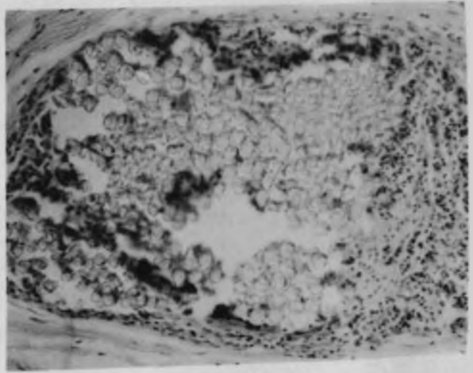
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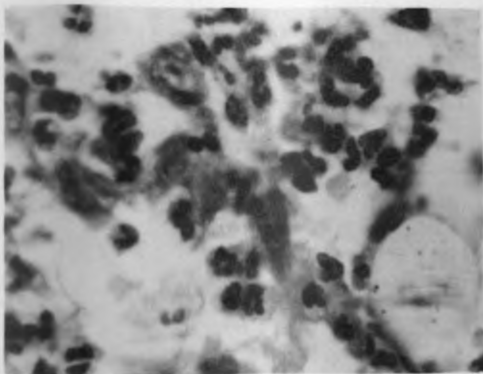
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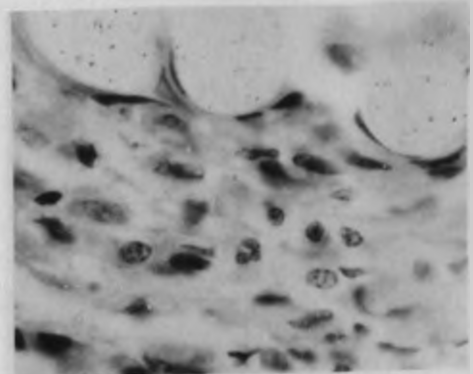
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C



F



Figure 11. Distribution of the species in the study area. The map shows the distribution of the species in the study area.

Table 1. Summary of the data collected during the study. The table shows the number of individuals of each species collected during the study.

Table 2. Summary of the data collected during the study. The table shows the number of individuals of each species collected during the study.

Species	Number of individuals
Species 1	10
Species 2	20
Species 3	30
Species 4	40
Species 5	50
Species 6	60
Species 7	70
Species 8	80
Species 9	90
Species 10	100



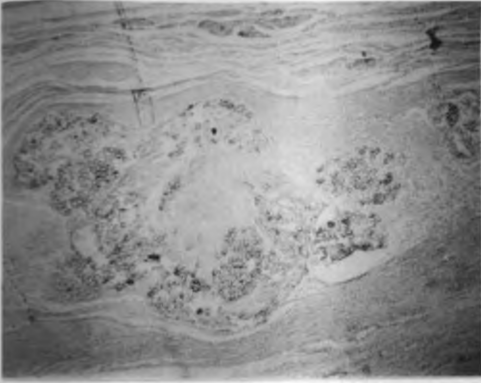
Figure 32. Photomicrographs of tissue reaction to Dexon at 10 and 40 days, at 1:1 dilution of culture.

A & B. At 10 days, 88X and 312X. Individual suture strands are seen in cross-section.

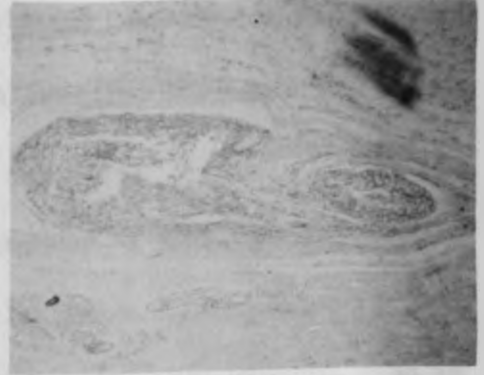
C. At 10 days, 2250X, showing cellular reaction around the suture. Predominantly neutrophils and macrophages are seen.

D & E. At 40 days, 88X and 312X.

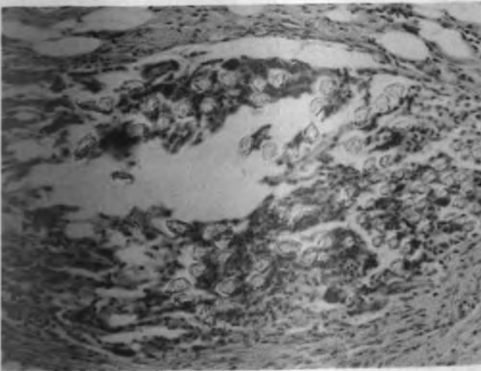
F. At 40 days, 2250X. Suture strands are being split. Predominant cells are macrophages and fibroblasts with some plasma cells. Reaction is seen between suture fibers.



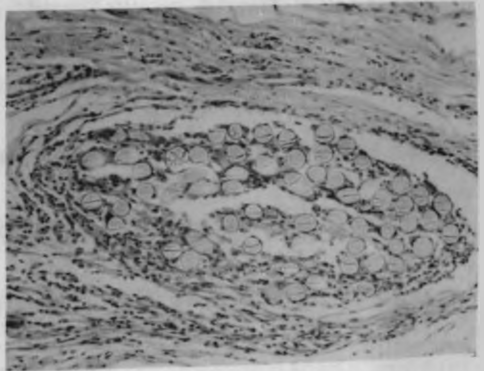
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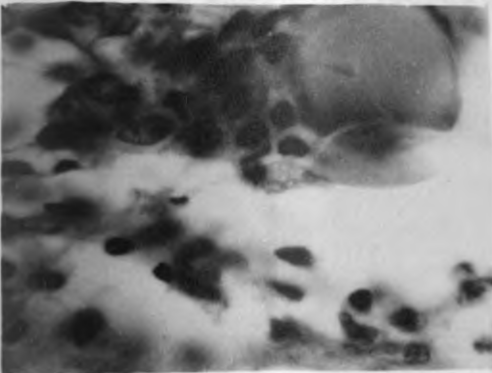
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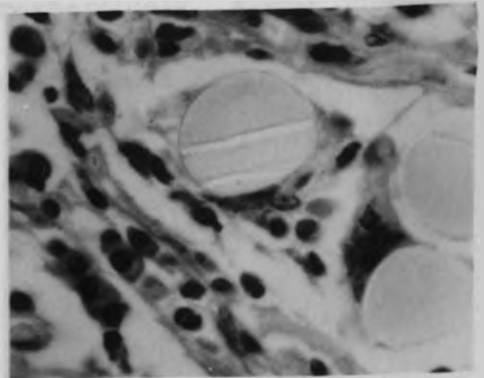
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zones. Both types of catgut elicited the most intense reaction of all sutures at 6 and 10 days but caused less reaction than silk at 20 and 40 days.

The predominant cells were neutrophils at 6 and 10 days. Macrophages and fibroblasts were less common. Lymphocytes were seen commonly and plasma cells and giant cells occasionally. At 20 days there were relatively fewer neutrophils and the relative number of macrophages and fibroblasts increased. Lymphocytes and plasma cells were seen occasionally. At 40 days the pattern was different for individual dilutions. At 1:1 dilution there were about equal numbers of neutrophils and macrophages. At 1:10 dilution there were fewer neutrophils and more macrophages and fibroblasts. There were very few neutrophils (3 to 4 percent) at 1:100 dilution. Macrophages and fibroblasts were equal in number. Plasma cells were seen at all three dilutions (1 to 9 percent).

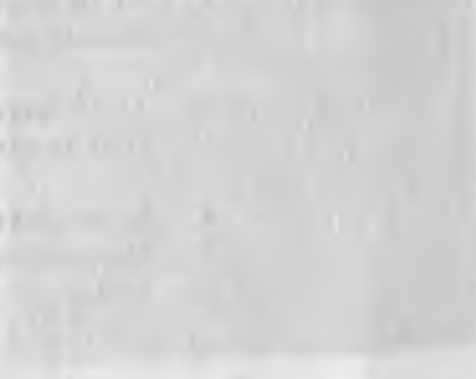
Catgut was disintegrating at 20 days and absorption was more complete at 40 days. Catgut was absorbed faster than Dexon. Representative photomicrographs at 10 and 40 days for plain and chromic catgut are shown in Figures 33 and 34 respectively. Color photographs of several sutures at 40 days are shown in Figures 35, 36 and 37.



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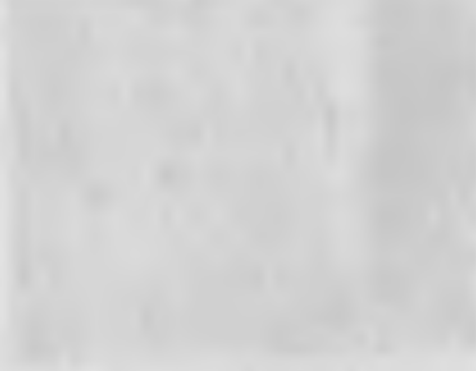


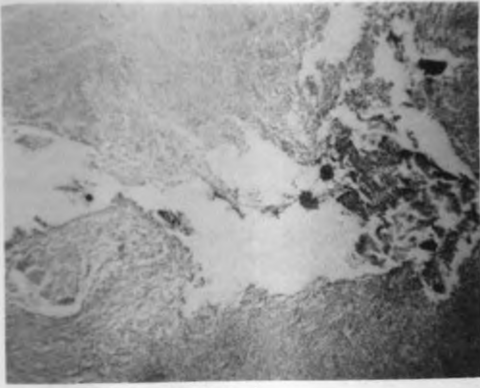
Figure 33. Photomicrographs of tissue reaction to plain catgut at 10 and 40 days, at 1:1 dilution of culture.

A & B. At 10 days, 88X and 312X. Broken down suture fibers are seen.

C. At 10 days, 2250X. Cellular reaction shows mainly neutrophils.

D & E. At 40 days, 88X and 312X. Suture fibers are still present.

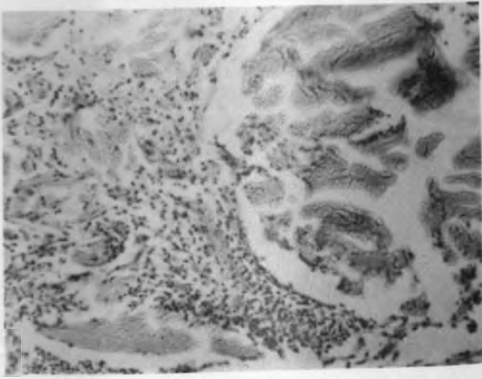
F. At 40 days, 2250X, showing cellular reaction. Macrophages and fibroblasts are the predominant cells.



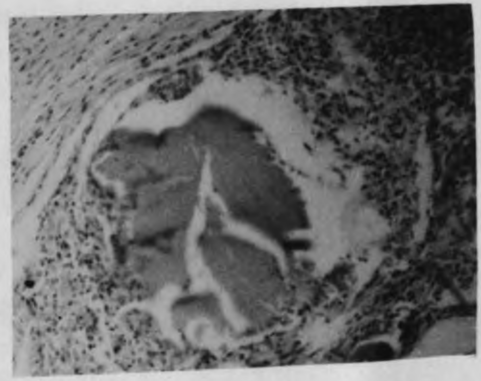
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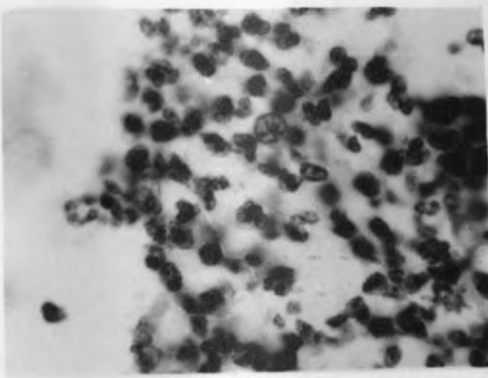
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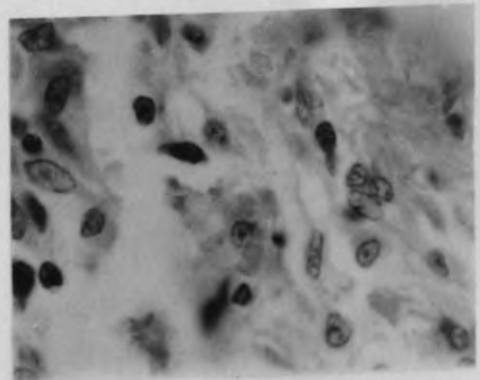
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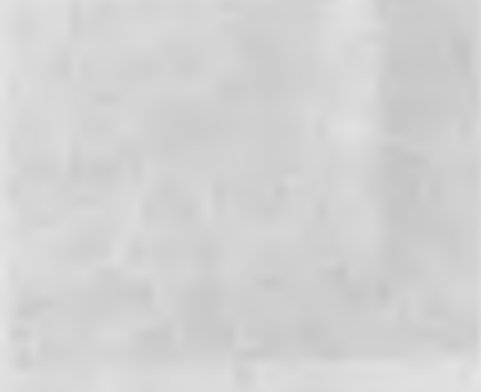


Figure 34. Photomicrographs of tissue reaction to chromic catgut at 10 and 40 days, at 1:1 dilution of culture.

A & B. At 10 days, 88X and 312X. Suture fibers are seen.

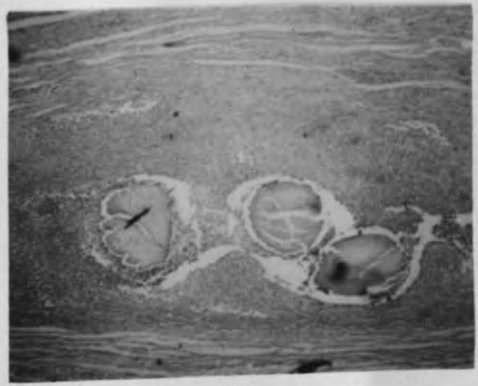
C. At 10 days, 2250X, showing cellular reaction. Mainly neutrophils and macrophages are seen around the suture strand.

D & E. At 40 days, 88X and 312X. Suture is still present.

F. At 40 days, 2250X. Cellular reaction consists of neutrophils, macrophages and plasma cells.



A



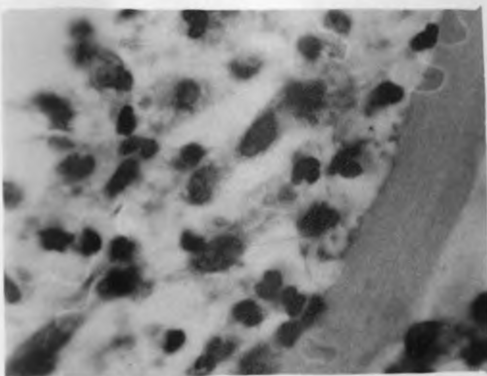
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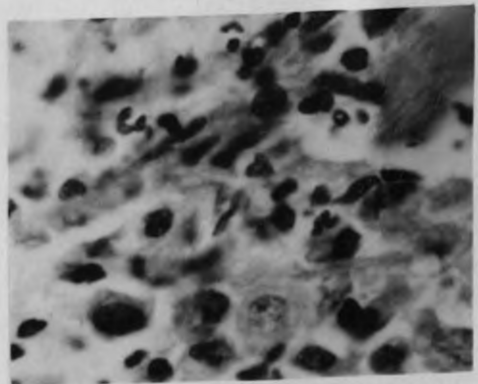
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E



C



F

Figure 35. Color photomicrographs of tissue reaction to sutures at 40 days, showing suture materials in cross-section (hematoxylin-eosin stain).

Mersilene, 1000X. Note cellular reaction between suture strands.

Dexon, 1000X. Note cellular reaction between suture strands.

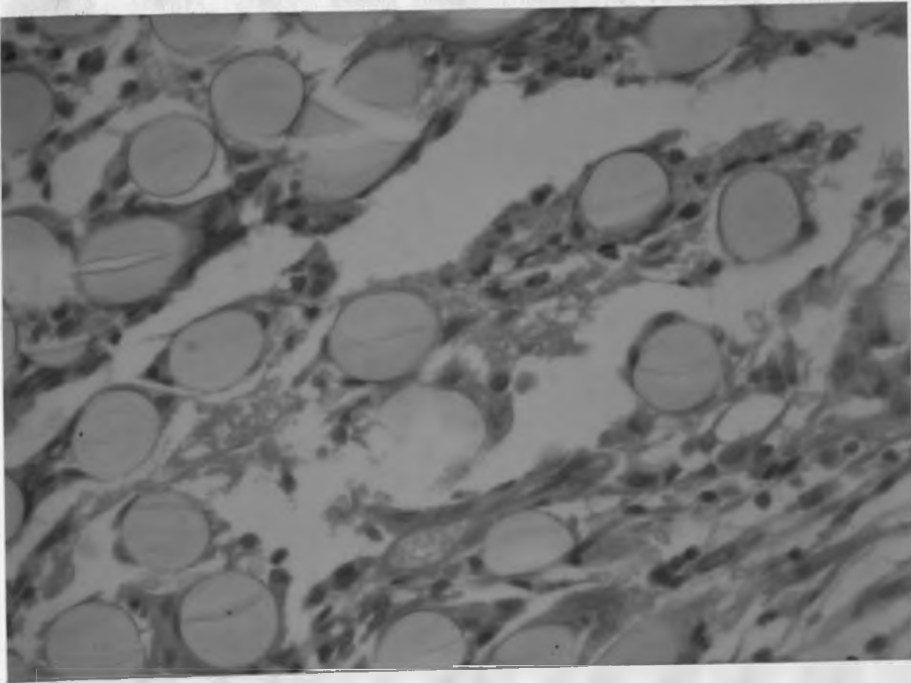
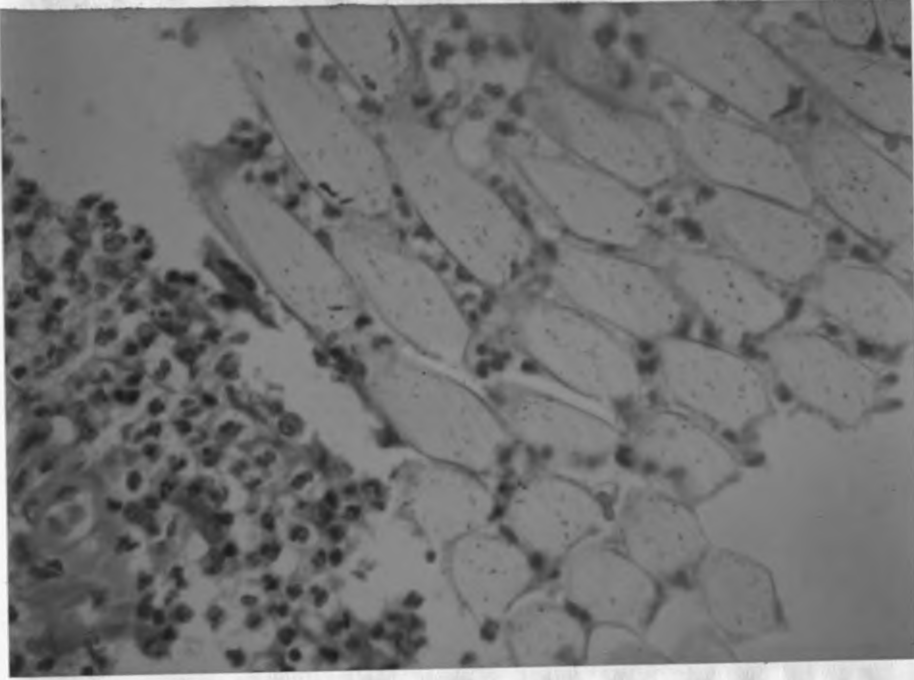


Figure 30. Color and topography of the surface of the  
 200 mesh sieve in the region of the  
 100 mesh sieve in the region

Fig. 30. Color and topography of the surface of the  
 200 mesh sieve in the region of the  
 100 mesh sieve in the region

Figure 36. Color photomicrographs of tissue reaction to sutures at 40 days, showing suture materials in cross-section (hematoxylin-eosin stain).

Plain catgut, 1000X.

Silk, 2250X, showing no eosinophilic deposit around the suture strands.

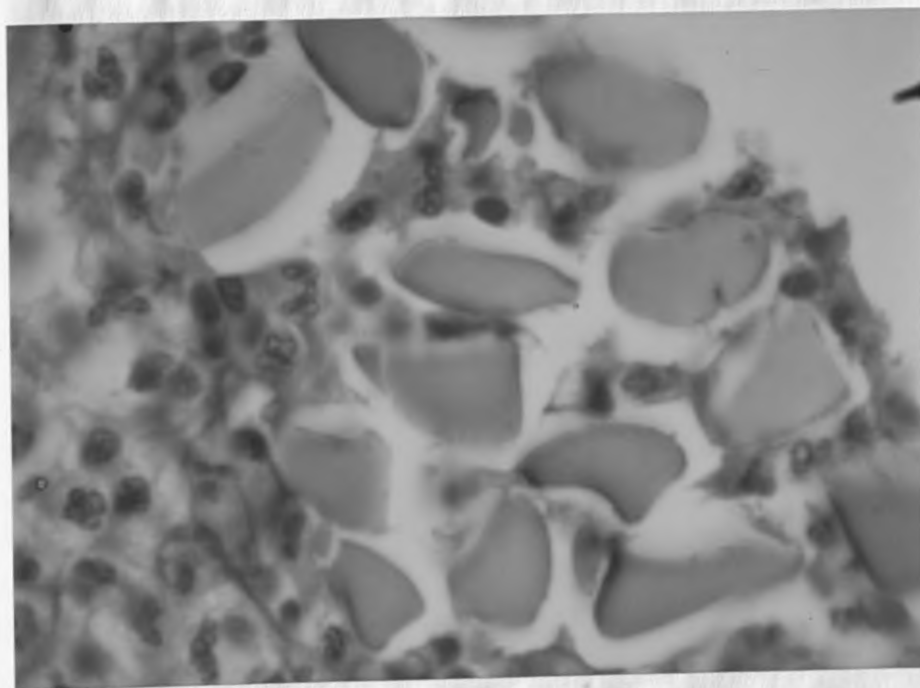
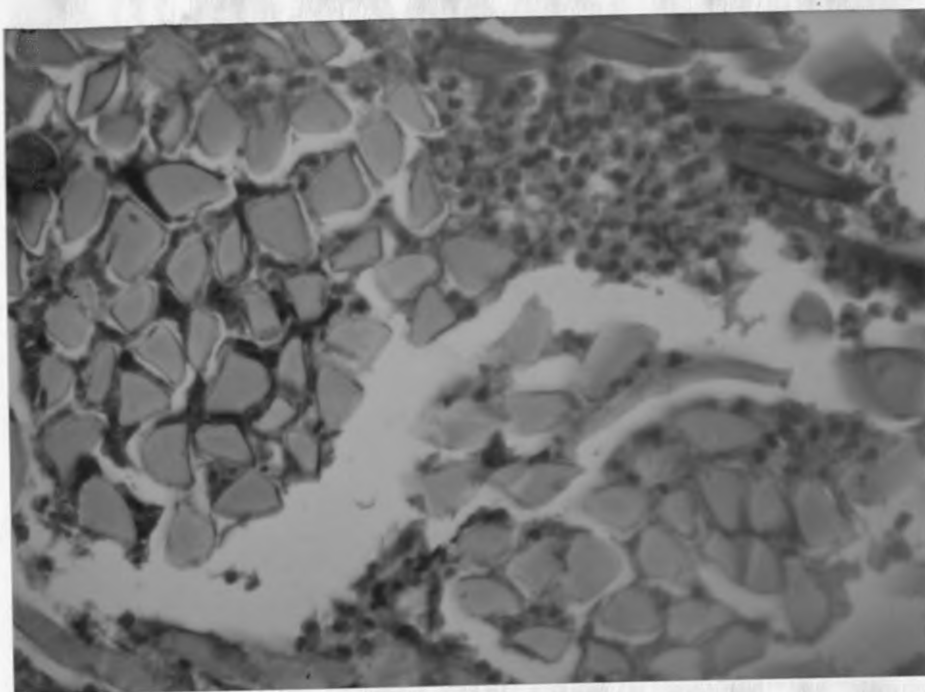




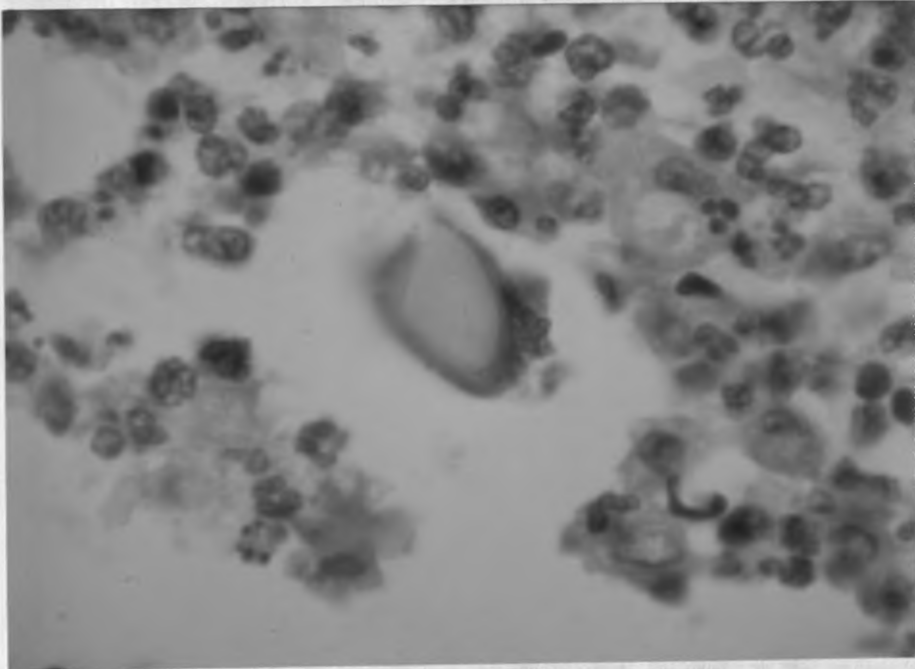
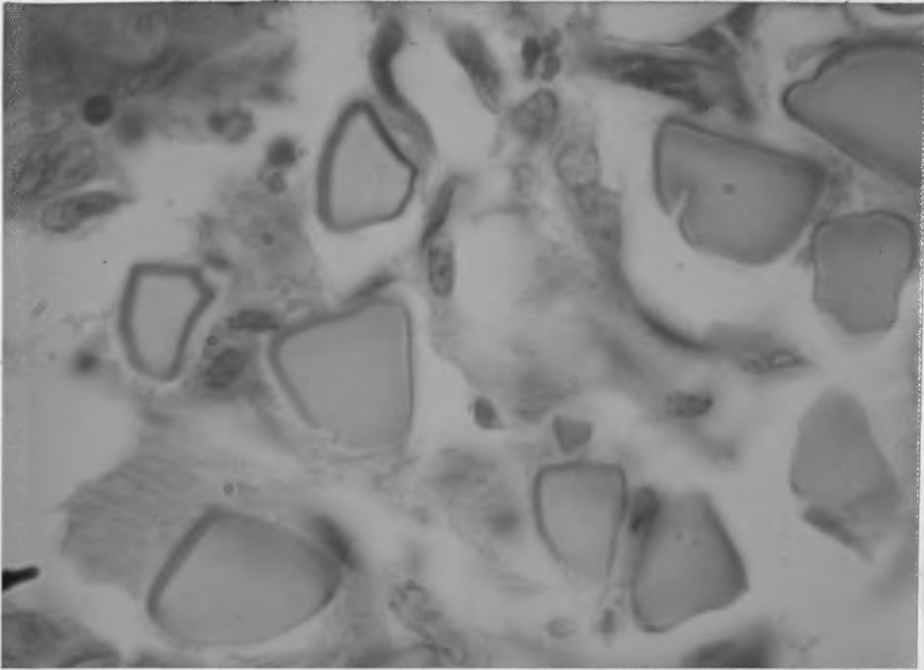
Figure 2. Coronal section of the lower jaw showing the position of the teeth and the surrounding bone structure.

Figure 3. Coronal section of the lower jaw showing the position of the teeth and the surrounding bone structure.

Figure 37. Color photomicrographs of tissue reaction to sutures at 40 days, showing suture materials in cross-section (hematoxylin-eosin stain).

Silk, 2250X, showing mild eosinophilic deposit (Splendore-Hoeppli Phenomenon) around several suture strands.

Silk, 2250X, showing marked Splendore-Hoeppli phenomenon around the individual suture strand.



## CHAPTER V

### DISCUSSION AND CONCLUSIONS

Several investigations have been conducted to compare tissue reaction elicited by different surgical sutures in "clean" wounds but such studies in infected wounds have been limited. It has long been recognized that suture materials can lead to development or persistence of local infection (Elek and Conen, 1957; James and MacLeod, 1961). It has also been suggested that the degree of enhancement of infection by a suture material is proportional to the inflammatory reaction elicited by the material itself (James and MacLeod, 1961).

With the advent of polyglycolic acid, a new synthetic absorbable material, Dexon, was developed and made available for clinical use. It has been claimed to cause minimal tissue reaction. A comparative investigation of this material and other conventional sutures in the presence of infection was conducted by the author in an earlier study (Varma, 1973). The degree of infection observed with Dexon in contaminated wounds was significantly more than steel and nylon, and not different from plain or chromic catgut. This finding was at variance with claims that Dexon was a relatively inert suture material.

The present study was undertaken to compare the varying degrees of infection enhanced by different suture materials in

contaminated wounds, and to evaluate tissue reactions elicited by them at various stages of implantation. This was done to test whether the response to Dexon over a prolonged period was indeed as great as it was at six days and whether relative reactions to other sutures varied at later stages.

To compare with the author's previous work, evaluation of sutures in comparatively acute stages of infection was repeated in one group of dogs which were sacrificed six days after surgery. Dogs in other groups were sacrificed at 10, 20 and 40 days to provide a more chronic and complete spectrum of observations.

Tryptose broth, the culture medium used, did not produce any reaction at any inoculation site indicating that it played no role in the formation of abscesses observed in pockets inoculated with culture.

There was no correlation between suture materials and draining abscesses. Although more abscesses drained with Dexon (9 of 180), this was not statistically significant. All the abscesses drained in the acute stage of infection (less than 10 days). This could not be related to the phenomenon of "spitting" sutures which is observed as a late reaction to some sutures.

The pattern of reactions elicited by different sutures in the six day survival group of the present study was generally similar to the previous work. However, the tissue response was always greater with all treatments in the previous study (Figure 38). This can be

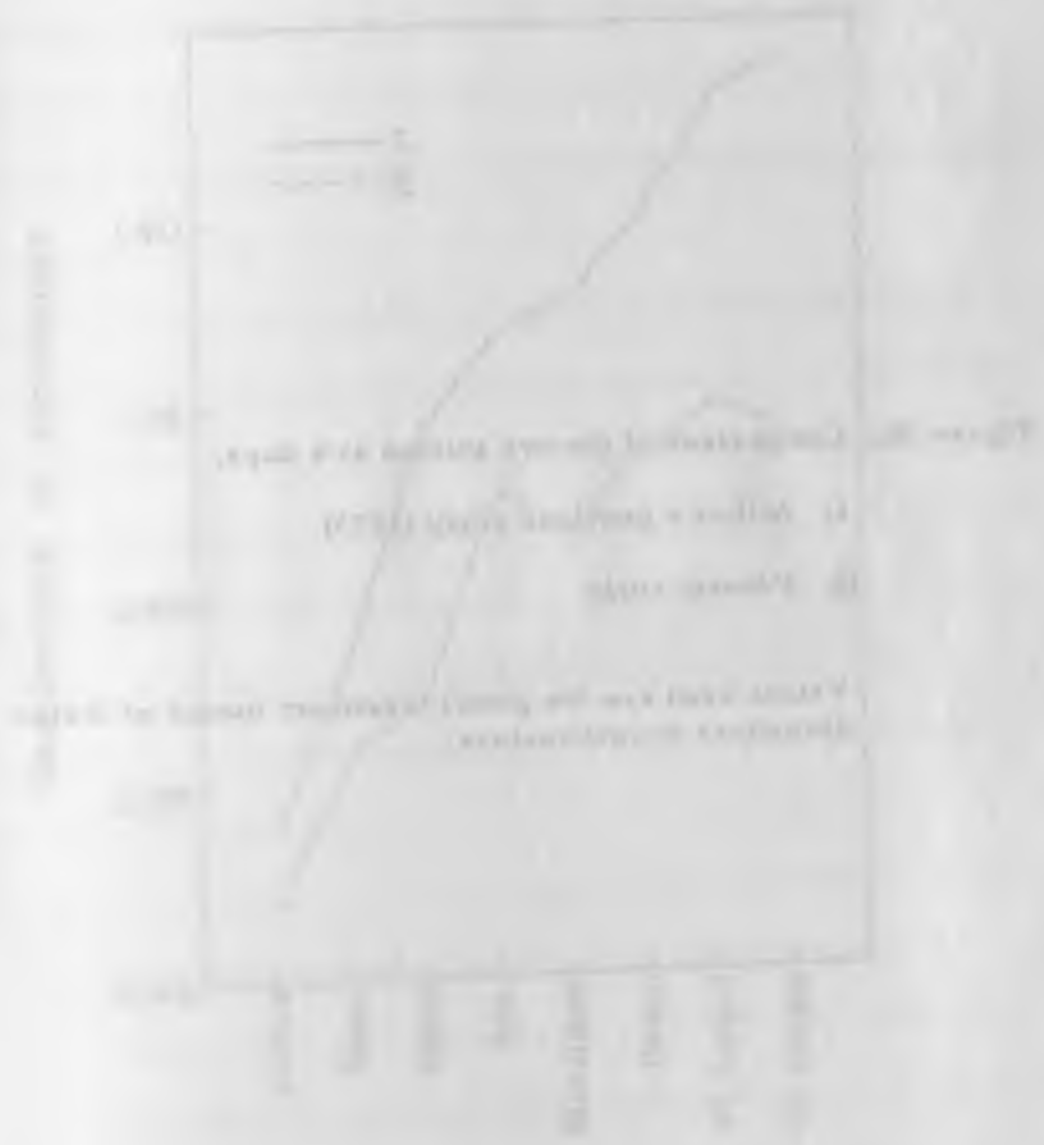


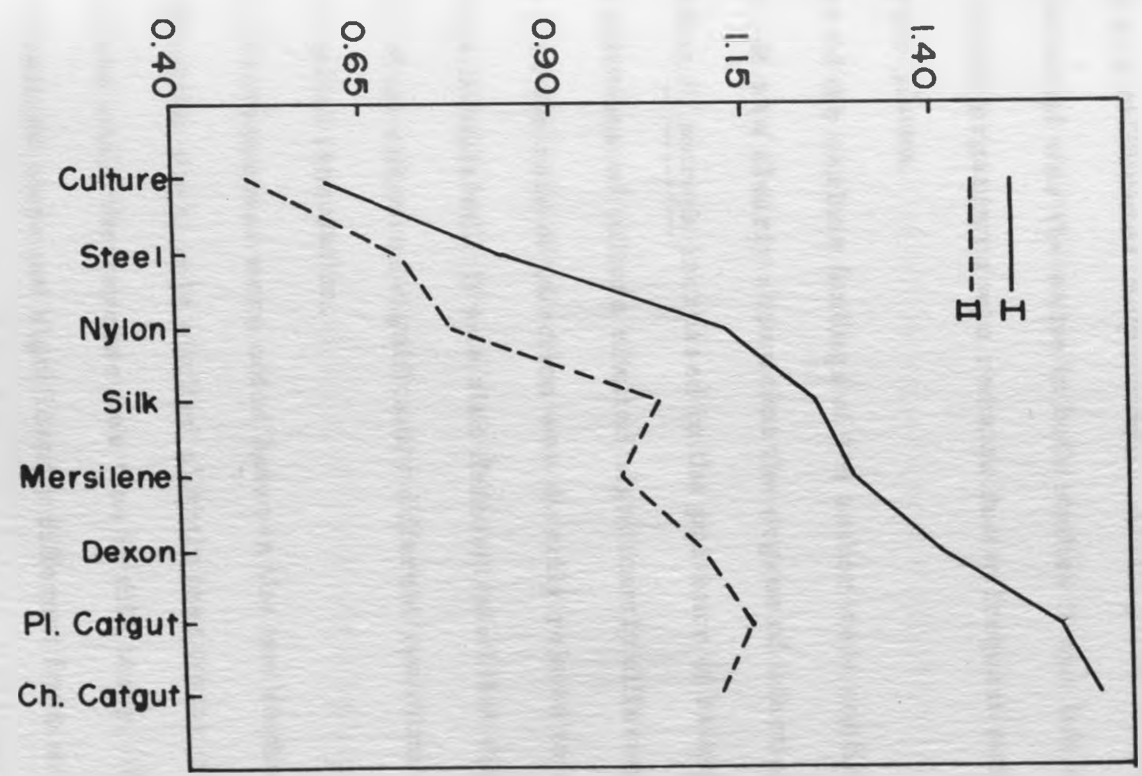
Figure 38. Comparison of the two studies at 6 days.

I. Author's previous study (1973)

II. Present study

Values used are the global treatment means of lesion diameters in centimeters.

Measurements in centimeters





explained by the dose of infective material used. In the first study the 1:1, 1:10 and 1:100 dilutions of S. aureus culture contained  $93 \times 10^7$ ,  $93 \times 10^6$  and  $93 \times 10^5$  organisms per milliliter of broth while in the present study there were  $74 \times 10^7$ ,  $74 \times 10^6$  and  $74 \times 10^5$  organisms per millimeter respectively. Although the source of the infective material was the same in both studies it was felt that the virulence of the organisms was reduced due to frequent subculturing on blood agar plates.

Some of the earlier findings of the author were confirmed in this study. It was clearly shown that the degree of infection induced by inoculating S. aureus increased in the presence of suture material. The three dilutions of culture elicited significantly different reactions, indicating that the rate of infection was directly related to the number of organisms inoculated. It was also demonstrated that different suture materials enhanced significantly different reactions at the same level of bacterial inoculation.

Few discrepancies were noted between the two studies. In the author's previous work, silk elicited slightly less tissue reaction than Mersilene while the reverse was true in this study. However, silk and Mersilene were not significantly different from each other in either study. The same observation was made between plain and chromic catgut. In this study plain catgut elicited slightly more tissue reaction than chromic catgut, similar to the findings of

Alexander et al. (1967), Herrmann et al. (1970) and Van Winkle and Hastings (1972). Tissue reaction elicited by Dexon was similar in both studies. It was between silk and Mersilene and the catguts. Steel and nylon caused the least tissue reaction in both studies. One of the reasons for variation between the current and the previous study could be explained on the basis that most of the suture materials in the current study were made by a different company.

It was interesting to note that steel elicited the least reaction of all sutures in acute stages of infection irrespective of its physical configuration. Monofilament steel was used in the previous work, whereas multifilament steel was substituted for it in this study. Variation in reaction to individual suture materials in the two studies was less with steel than with any other suture material. This finding does not agree with the author's earlier claim that the physical nature of a suture material is a contributing factor in the degree of infection induced around the suture. Recently, Edlich et al. (1973, 1974) have also suggested that the physical configuration of the suture plays a relatively unimportant role in the development of early surgical infection. The chemical structure of the suture may be a more important factor in the enhancement of infection, as has already been hypothesized (Edlich et al., 1973, 1974).

At 10 days the three dilutions of culture continued to elicit significantly different responses. The relative reactions to different

sutures was basically the same as at six days, except that silk showed slightly more reaction than Mersilene and Dexon was very similar to the catguts in stimulating tissue reaction. The catguts still caused the most reaction and nylon and steel the least. At 20 days, differences between dilutions of culture were less marked although the 1:1 dilution enhanced significantly more reaction than the 1:10 and 1:100 dilutions. Significant differences between sutures were markedly reduced; however, Dexon showed relatively little tissue reaction and silk elicited the most reaction. Mersilene and the catguts were intermediate between Dexon and silk. Nylon and steel remained the most inert of all sutures. At 40 days differences between the three dilutions were again noted although they were not very marked. Of all the sutures, silk elicited by far the greatest response and Mersilene produced slightly less. All other sutures showed minimal tissue response. It was evident that the relative tissue reaction to silk increased with time, and it elicited the largest suture granulomas at 40 days. Mersilene also produced comparatively more tissue reaction at later stages of implantation, but reaction to it was slightly less than that of silk. Although causing marked reaction in acute stages of infection, Dexon demonstrated little tissue reaction in later stages. This compared favorably with the findings of most workers. Reaction elicited by the catguts changed relative to alterations in tissue reactions to silk, Mersilene

and Dexon. Overall, the catguts elicited marked reactions at 6, 10 and 20 days, but not at 40 days. Multifilament steel and monofilament nylon stimulated the least reaction of all sutures at all periods of study.

Looking at the pooled data of all groups it was clearly found that the presence of suture material in an infected pocket enhanced tissue response, as culture alone elicited minimal reaction. Silk elicited the most reaction of all sutures and steel and nylon the least. Dexon, Mersilene and the catguts were intermediate. This demonstrated that monofilament or multifilament nature of the suture material did not play an important role in tissue reaction elicited. Overall reactions at 6, 10 and 20 days differed markedly from each other but there were no significant differences between reactions at 20 and 40 days. This suggested that after 20 days chronic suture granulomas showed little change.

Microscopic measurement of lesions was found to be a useful adjunct to gross measurements. More measurements could be made by this method. Gross and microscopic measurements showed good correlation except at 1:1 dilutions in Group A (6 days) and Group B (10 days). This was thought to be due to loss of pus and suture knot on sectioning the abscess, with marked reduction in size of the lesion. Several histopathologic reactions to steel were difficult to evaluate, as in the process of removing the suture from the lesion the inflammatory reaction around it was disturbed.

Several comparative studies have been made to evaluate histologic tissue reaction to various sutures. Detailed cellular reactions in acute and long-term implantations have been described by Postlethwait (1968; 1970a, b) and Postlethwait et al. (1975). No reports have appeared in the literature describing histopathologic reactions to infected sutures. The technique of histopathologic grading of tissue reactions described by Sewell et al. (1955) was not very applicable to comparative studies at various stages of implantation and for infected wounds. However, it was found to be a useful guideline.

The predominant inflammatory cells at six days were neutrophils due to the bacterial infection. Less predominant cells seen were macrophages and fibroblasts. Lesions with Dexon contained the greatest number of neutrophils and steel the least. Silk, Dexon and the catguts attracted more lymphocytes than other sutures at six days. At 10 days Dexon, steel and nylon showed less neutrophils and more macrophages and fibroblasts. The change was dramatic with nylon; reduction in neutrophils with other sutures was not so marked. At 20 days lesions with steel and nylon had very few neutrophils. More macrophages were seen. Mersilene, silk and the catguts still showed neutrophils as predominant cells. Dexon was intermediate in its neutrophilic response. At 40 days neutrophils were minimal with steel, nylon and Dexon and there were more

macrophages and fibroblasts. Dexon had a high number of plasma cells. Mersilene, silk and the catguts retained high numbers of neutrophils even at 40 days, suggesting persistence of local infection. These sutures also had the greatest tissue response as evidenced grossly by suture granulomas. Plasma cells were commonly seen with these sutures. The cellular reaction observed at later stages of implantation was similar to that described by Postlethwait (1968, 1970a, b) and Postlethwait et al. (1975).

Lesions with Dexon contained large numbers of neutrophils in early stages of infection, probably explaining the large tissue reaction to Dexon in acute infection. However, there was rapid reduction in neutrophils in later stages with relatively mild tissue response. Mersilene, silk and the catguts had persistent local infection and hence exaggerated tissue reaction.

Macrophages appeared as early as six days and were more prominent at later stages. These cells accumulate at sites of acute or chronic inflammation, exercising their function in defense against microbial agents and in removal of effete and damaged cells (Schalm et al., 1975). Plasma cells were seen more at later stages of implantation. Schalm et al. (1975) have suggested that plasma cells appear in areas of antigenic deposition.

Occasionally an eosinophilic deposit around individual silk fibers was observed at 40 days. Its characteristics were similar to

the Splendore-Hoeppli phenomenon (SHP) described around silk by Liber and Choi (1973). No SHP was found about steel, nylon, Mersilene, Dexon or the catguts. It has been suggested that SHP represents an antigen-antibody reaction and the formation of deposit is related to the biochemical and immunological nature of silk. This finding gave support to strong allergenic properties of silk. Reaction to silk sutures due to allergy has been reported previously (Getzen and Jansen, 1966; Roe, 1975; \_\_\_\_\_, 1976).

Assessment of absorption of Dexon and the catguts was difficult as some lesions lost the suture material during sectioning, especially at 6 and 10 days. Evaluation of lesions at 20 and 40 days suggested that catgut was absorbed earlier than Dexon. Unlike catgut, reaction to Dexon was minimal during the absorption stages. This finding is similar to that observed by Postlethwait (1970b).

## CHAPTER VI

### SUMMARY

A comparison was made between Dexon (braided polyglycolic acid), plain catgut, chromic catgut, multifilament steel, monofilament nylon, braided silk and Mersilene (braided dacron) in infected wounds. The findings of this study in acute (6 days) and chronic (10, 20 and 40 days) implantations are summarized below:

1. Suture materials play an important role in the enhancement of infection produced in surgical wounds.
2. The amount of reaction produced in an infected wound is proportional to the number of organisms in the wound. Suture materials elicited significantly different reactions with each of the three dilutions (1:1, 1:10, 1:100) of S. aureus culture in acute stages of infection. This was less marked in chronic implantations.
3. At 6 days postimplantation, the seven suture materials produced different reactions when challenged with similar numbers of S. aureus organisms. Multifilament steel and monofilament nylon elicited the least reaction. Plain and chromic catgut stimulated the most reaction. Dexon, Mersilene and silk were intermediate.
4. At later stages of implantation (10, 20 and 40 days) reaction to steel and nylon remained minimal. Silk showed a



comparative increase in its reaction while Dexon decreased comparatively. Mersilene elicited slightly less reaction than silk. The catguts did not alter much in the comparative reactions elicited by them.

5. Tissue reaction to sutures decreased with time but the change was not significant after 20 days.

6. Across all groups and dilutions, silk elicited by far the greatest tissue reaction, and steel and nylon the least.

7. Although Dexon showed intense reaction in acute infection, the reaction was mild in chronic implantations. Dexon appeared to be absorbed more slowly than plain or chromic catgut and tissue reaction was minimal in the absorption stages.

8. Cellular reaction varied with different suture materials. In general, neutrophils were the predominant cells in acute infection, but in later stages macrophages and fibroblasts predominated. Occasionally plasma cells, lymphocytes, eosinophils and giant cells were seen. Lesions with plain and chromic catgut, silk and Mersilene showed large numbers of neutrophils even at 40 days post-implantation, suggesting persistence of local infection. The numbers of neutrophils rapidly decreased with nylon, steel and Dexon.

9. Correlation between physical configuration of sutures, monofilament or multifilament, and their ability to enhance infection in contaminated wounds was not definitely established. Although

nylon, the only monofilament suture used, generally elicited the least reaction of all sutures, multifilament steel compared very favorably with it at all stages of implantation. Multifilament silk, Mersilene and the catguts elicited considerably more tissue reaction. Enhancement of infection by sutures may be related to their chemical structure rather than their physical configuration. Reduced reaction to Dexon in chronic implantations may be related to absorption properties of the material.

10. A reaction similar to the Splendore-Hoeppli phenomenon was seen around multifilament silk strands at 40 days postimplantation.

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APPENDIX



Table A1. Parasitologic Results (Eggs per Gram of Feces) of All Dogs in the Four Groups at the Time of Acquisition.

Group A			Group B			Group C			Group D		
Dog Number	<u>Ancylostoma caninum</u>	<u>Toxocara canis</u>	Dog Number	<u>Ancylostoma caninum</u>	<u>Toxocara canis</u>	Dog Number	<u>Ancylostoma caninum</u>	<u>Toxocara canis</u>	Dog Number	<u>Ancylostoma caninum</u>	<u>Toxocara canis</u>
2	1100	775	8	950	--	12	350	625	10	400	--
16	7000	--	18	1850	--	20	125	--	17	2325	--
29	725	--	25	--	--	6	200	25	15	525	--
32	1900	--	31	1975	--	38	225	125	23	100	--
27	75	25	35	4150	25	34	7775	25	14	2375	--
4	375	--	42	8470	--	47	75	--	52	100	--
26	225	325	40	1625	--	53	3225	--	55	2025	--
56	--	--	44	1225	--	36	1625	50	37	75	--
63	4700	--	33	6275	550	54	--	--	41	1625	50
13	1750	175	64	350	1225	65	25	100	61	700	--
62	900	1825	11	450	--	59	75	25	66	3825	--
70	200	--	5	2200	1700	69	300	--	67	925	--
78	300	--	68	1850	--	74	1725	--	77	--	--
72	150	--	51	5475	--	71	800	--	82	6750	200
88	875	--	76	--	--	81	--	--	83	850	225

Table A2. Group A - Hematologic Values at Time of Surgery.

Dog Number	PCV	TP	WBC	Differential Count (%)			
				TN	L	M	E
2	47.0	8.8	8.1	73	24	0	3
16	40.0	8.6	8.6	57	34	3	6
29	40.0	8.2	15.6	39	45	5	11
32	43.0	7.8	13.6	63	28	2	7
27	47.0	8.2	12.8	49	40	0	11
4	45.0	6.2	17.2	56	35	0	9
26	39.0	6.4	16.3	44	51	0	5
56	48.0	8.0	15.2	50	43	1	6
63	42.0	8.8	11.6	56	36	0	8
13	38.0	7.0	17.9	46	43	0	11
62	36.0	9.4	13.9	58	36	0	6
70	36.0	7.6	11.6	57	38	1	4
78	41.0	7.0	12.3	48	44	0	8
72	52.0	7.2	17.5	60	27	2	11
88	36.0	8.0	13.7	48	34	0	18

PCV Packed Cell Volume (Hematocrit) in %.

TP Total Protein in grams per 100 milliliters.

WBC White Blood Cell Count in thousands per cubic millimeter.

TN Total Neutrophils - expressed in % of total WBC.

L Lymphocytes - expressed in % of total WBC.

M Monocytes - expressed in % of total WBC.

E Eosinophils - expressed in % of total WBC.

Table A3. Group B - Hematologic values at Time of Surgery.

Dog Number	PCV	TP	WBC	Differential Count (%)			
				TN	L	M	E
8	41.0	7.6	13.3	71	26	0	3
18	42.0	7.8	9.1	72	28	0	0
25	48.0	10.2	15.8	70	23	0	7
31	49.0	7.8	13.2	54	40	0	6
35	39.0	7.2	15.6	57	33	0	10
42	47.0	7.2	17.8	54	40	6	0
40	50.0	8.0	9.6	42	51	1	6
44	39.0	7.4	13.6	74	22	0	4
33	51.0	7.0	17.2	60	33	0	7
64	48.0	8.0	13.6	63	26	0	11
11	46.0	7.4	14.7	51	43	0	6
5	48.0	7.2	14.3	68	25	0	7
68	42.0	7.6	11.9	63	21	5	11
51	35.0	6.8	12.9	64	28	2	6
76	49.0	7.8	12.6	54	38	0	7

PCV Packed Cell Volume (Hematocrit) in %.

TP Total Protein in grams per 100 milliliters.

WBC White Blood Cell Count in thousands per cubic millimeter.

TN Total Neutrophils - expressed in % of total WBC.

L Lymphocytes - expressed in % of total WBC.

M Monocytes - expressed in % of total WBC.

E Eosinophils - expressed in % of total WBC.

Table A4. Group C - Hematologic Values at Time of Surgery.

Dog Number	PCV	TP	WBC	Differential Count (%)			
				TN	L	M	E
12	40.0	7.8	8.1	59	31	3	7
20	45.0	7.8	13.7	73	25	1	1
6	47.0	8.2	15.5	60	26	5	9
38	41.0	7.2	12.4	68	29	0	3
34	39.0	6.4	18.0	69	20	0	11
47	42.0	7.2	7.4	51	41	0	8
53	38.0	7.0	15.6	54	43	1	2
36	37.0	7.6	14.1	70	26	0	4
54	43.0	7.8	8.5	50	48	0	2
65	48.0	7.8	12.2	63	32	0	5
59	37.0	8.6	16.3	55	35	2	8
69	38.0	7.8	13.7	65	23	0	12
74	43.0	8.6	12.7	56	35	1	8
71	40.0	7.2	15.2	64	32	0	4
81	38.0	9.0	9.9	56	26	0	18

PCV Packed Cell Volume (Hematocrit) in %.

TP Total Protein in grams per 100 milliliters.

WBC White Blood Cell Count in thousands per cubic millimeter.

TN Total Neutrophils - expressed in % of total WBC.

L Lymphocytes - expressed in % of total WBC.

M Monocytes - expressed in % of total WBC.

E Eosinophils - expressed in % of total WBC.

Table A5. Group D - Hematologic Values at Time of Surgery.

Dog Number	PCV	TP	WBC	Differential Count (%)			
				TN	L	M	E
10	52.0	7.8	12.1	56	42	0	2
17	49.0	7.6	16.6	67	26	1	6
15	45.0	7.2	12.5	45	44	0	11
23	41.0	7.2	17.6	51	27	0	22
14	36.0	6.6	17.0	46	44	0	10
52	44.0	7.2	10.9	46	52	0	2
55	36.0	7.4	16.4	70	30	0	0
37	36.0	6.0	15.3	51	36	0	13
41	46.0	6.0	9.3	56	34	0	10
61	45.0	6.0	11.8	50	44	0	6
66	47.0	7.6	16.7	63	29	2	6
67	39.0	8.2	13.6	60	35	0	5
77	36.0	7.2	15.0	45	42	1	12
82	35.0	6.8	6.4	50	38	0	12
83	35.0	7.4	17.8	62	37	0	1

PCV	Packed Cell Volume (Hematocrit) in %.
TP	Total Protein in grams per 100 milliliters.
WBC	White Blood Cell Count in thousands per cubic millimeter.
TN	Total Neutrophils - expressed in % of total WBC.
L	Lymphocytes - expressed in % of total WBC.
M	Monocytes - expressed in % of total WBC.
E	Eosinophils - expressed in % of total WBC.

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