THE MECHANICAL AND BIOLOGICAL PERFORMANCE OF THE ALTERNATING SLIDING KNOTS WITH DIFFERENT PATTERNS IN ABDOMINAL WOUND CLOSURE

by

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M.S. in Biomedical Engineering, Boğaziçi University, 1993

Submitted to the Institute of Biomedical Engineering
in partial fulfillment of the requirements
for the degree of
Doctor
of
Philosophy

Boğaziçi University
March, 1998

ACKNOWLEDGMENTS

I would like to thank my advisor Prof. Dr. Sabri Altıntaş for his orientation, Assoc. Prof. Dr. Özcan Gülçür for his advises, Dr. Aykut Sümer for his assistance, Assoc. Prof. Dr. Sabri Ergüney, Assoc.Prof. Dr. Süha Göksel, Prof. Dr. Müzeyyen Mamal Torun, Research Assistant Hrisi Bahar and Prof. Dr. Imer Okar for their valuable contributions in this study.

I would like to also thank Prof. Dr. Burak Erman for the approval to use facilities of the Polymer Research Center laboratory, Doğsan Ltd. company for providing the surgical sutures, and Boğaziçi University Research fund under project no 95HX0009 for providing financial support.

Finally, I owe it all to my parents.

ABSTRACT

New knot configurations, consisting of alternating strands with different patterns, have been studied from mechanical and biological perspectives in order to determine whether they would be suitable for abdominal surgery, as compared with conventional sliding knots. Mechanical properties of these new knots were compared with those of the classical sliding knots and single threads for silk and nylon sutures under dry conditions. From the mechanical perspective, the new knots showed better knot holding capacity and efficiency.

In the in vivo implantation tests performed on the rat abdominal wall, the alternating sliding knots with different patterns were found to be more efficient and secure than the classical sliding knots. The knot configuration, postoperative period, suture material and size were important factors in determining the knot holding capacity.

From the biological perspective, these new knots provoked tissue reaction similar to the classical sliding knots. Because nylon is less pliable than silk, its use resulted in higher effective knot volumes, causing more pronounced tissue reaction.

To test the bacterial adherence to the knots, in vitro and in vivo tests were performed in rats. The degree of the elicited infection correlated well with the capability of bacteria to bind to the suture. It was observed that the knot configurations and the suture sizes did not have much effect on bacterial adherence. Due to the presence of interstices between throws, the knots had greater capacity to retain bacteria than the single threads for both silk and nylon, thus promoting infection.

The elasticity and stress-relaxation properties of these knots were compared to those of single threads of silk and nylon. The elasticity of the knots, in general, was higher than that of the threads for both materials. The silk showed decreased elasticity at high extension levels, while nylon showed increased elasticity. In stress relaxation tests, the residual load fraction of the knots was found to be higher than that of the threads at all extension levels.

A model was created to study the effect of several factors on the suture pullout force in the abdominal wall. Incisional direction, knot configuration, strain rate and tissue healing strength were important factors in determining the suture pullout force. In conclusion, we do recommend the use of the alternating sliding knots with different patterns in abdominal surgery instead of the currently used sliding knots.

Keywords: Alternating sliding knots with different patterns, abdominal surgery, knot security, knot holding capacity, suture pullout force.

KARIN YARALARININ KAPANMASINDA FARKLI BİÇİMLİ KAYAN DÜĞÜMLERİN MEKANİK VE BİYOLOJİK DAVRANIŞI

ÖZET

Karın cerrahisinde, klasik kayan düğümlere kıyasla daha uygun olup olmadıklarını belirleyebilmek için farklı biçimli, yer-değiştirmeli kayan düğümlerden oluşan yeni düğüm biçimleri mekanik ve biyolojik yönden incelendi. Kuru koşullarda, ipek ve naylondan yapılan tek iplik ve düğümler için, bu yeni düğümlerin mekanik özellikleri klasik kayan düğümlerle karşılaştırıldı. Mekanik açıdan, yeni düğümlerin düğüm tutma kapasitelerinin ve verimlerinin daha iyi olduğu gösterildi.

Sıçan karın duvarında yapılan "in vivo" implantasyon testlerinde, farklı biçimli kayan düğümlerin klasik kayan düğümlerden daha verimli ve daha güvenli olduğu saptandı. Düğüm biçimi, cerrahiden sonra geçen zaman, cerrahi ip materyali ve çap düğüm tutma kapasitesini belirleyen önemli faktörlerdi.

Biyolojik yönden bu yeni düğümlerin klasik kayan düğümlerle aynı doku reaksiyonuna yol açtıkları görüldü. Naylon ipek kadar yumuşak olmadığından, naylon iple yapılan düğümlerin etkin hacimlerinin daha büyük olduğu, ve bu nedenle de daha belirgin doku reaksiyonuna sebep oldukları saptandı.

Düğümlere bakteri yapışmasını test etmek için sıçanlarda "in vivo" ve "in vitro" testler yapıldı. Oluşan enfeksiyon bakterinin ipliğe yapışabilme kapasitesiyle orantılı bulundu. Düğüm biçimi ve iplik çapının bakteri yapışmasında etkili bir rol almadığı gözlemlendi. Hem ipek hem de naylon için, bağlar arasındaki boşluklar nedeniyle düğümlerin bakteri tutma kapasitelerinin tek ipten daha fazla olduğu bulundu.

İpek ve naylon için, yeni düğümlerle tek ipliklerin esneme ve gerilim-gevşeme özellikleri karşılaştırıldı. Genel olarak, düğümlerin esneme yeteneklerinin tek ipliklerden daha yüksek olduğu saptandı. Yüksek uzama düzeylerinde, ipek ip ve düğümlerin esneme yeteneklerinde azalma görülürken, naylonda artma görüldü. Gerilim gevşemesi testlerinde, tüm uzama düzeyleri için, kalan yük fraksiyonunun, düğümlerde iplerden daha fazla olduğu bulundu. Çeşitli faktörlerin, karın duvarında ip çekilme kuvveti üzerindeki etkilerini incelemek amacıyla bir model geliştirildi. Kesme yönü, düğüm biçimi, gerilme oranı ve doku iyileşme gücü ipin çekilme kuvvetini belirleyen önemli faktörlerdi.

Sonuç olarak, karın ameliyatlarında, halen kullanılmakta olan kayan düğümler yerine farklı biçimli kayan düğümlerin kullanılması önerilmektedir.

Anahtar kelimeler: Yer-değiştiren farklı biçimli kayan düğümler, karın cerrahisi, düğüm güvenliliği, düğüm tutma kapasitesi, cerrahi ip çekilme kuvveti.

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LIST OF ABBREVIATIONS

KHC	Knot Holding Capacity
KE	Knot Efficiency
ASTM	American Standards for Tensile Materials

1. INTRODUCTION

1.1 Motivations and Objectives

This work has been undergone to provide a good insight into the application of sliding knots in surgery. The aim is to introduce new sliding knot configurations and test their performance from many different aspects. As will be stated lately, the sliding knots are well used in the clinical practice; but they have not been closely examined. The reason surgeons use sliding knots more frequently than flat knots seems to be due to their convenience and speed of tying. Unfortunately, they were condemned by surgeons and they have been judged of having less performance when compared with the flat knots. Because of their high use, they should gain more attention. Consequently, they should not be avoided. On the contrast, their frequent use in clinical practice should be encouraging researchers to conduct more studies concerning their application. Therefore, in this thesis, we were interested in their application in abdominal surgery. In the category of the sliding knots, a new type exist that has not been studied so far. It is the type where the strand of suture alternates using different patterns than the preceding one (discussed lately). In this thesis, the performance of this type of sliding knots is examined from mechanical and biological aspects as well.

1.2 Thesis Outline

The thesis is composed of nine chapters. The first chapter presents a general overview on sutures, their role in wound healing and the complications they cause. The second chapter gives a study of the mechanical properties of silk and nylon sutures in thread configuration. The third chapter studies the mechanical properties of the same sutures in the loop configuration. The fourth chapter compares the different sliding knots mechanical performance under dry conditions. The fifth chapter compares the different sliding knots mechanical performance under in vivo conditions in the first part. In the second part, it studies the tissue reaction to these knots. The sixth chapter studies the bacterial adherence to these knots under in vitro and in vivo conditions. The seventh chapter studies two important mechanical properties: the elasticity and stress relaxation of sutures in thread and loop configurations. The eight chapter introduces a model in the rat abdominal wall to study the suture pullout force and the effect of certain factors like the incisional direction, knot configuration, postoperative days, strain rate suture material and size on the suture pullout force . Finally, chapter nine presents the conclusions and recommendations the study has to provide.

1.3 History of Sutures

The history of sutures may be divided into two periods. The beginning of the first dates back to at least four thousands years ago [1]. Going back to the year 1862, and relating to the Egyptian medicine, a surgical papyrus was found by Edwin Smith in Thebes. It was called the Edwin Smith papyrus and it is dated from about 1600 B.C. [2], [3]. On this papyrus, stitching, splints and the use of fire drill were mentioned [1], [2], [3]. It also mentioned the use of sutures, most probably linen, for wound edge approximation [1]. Dried gut, dried tendon, strips of hide, horsehair, women's hair, bark fibers were also used [3].

In Sushruta [2], the Indian famous medical manuscript written in Sanskrit, (622-542 B.C.), the following fundamental surgical techniques were mentioned: excision, incision, scarification, punction, probing, extraction, squeezing and sewing. Charaka, the summary of Susrhuta belongs to the first century A.D. [2]. Artandi [1] stated that around two thousands and six hundered years ago, Sushruta listed plated horse hair, cotton, strips of leather and fibers from tree bark along with sinews [1].

In Roman medicine, Aulus Cornelius Celsus (3-64 A.D.) who was from Latin race, descibed the surgical practice including the use of the ligature [2]. He referred to twisted sutures [1]. Galen [2], (130-201 A.D.) was a first rate anatomist and a great dissector. He mentioned catgut about one thousand eight hundred years ago for the first time [1, 4]. Catgut sutures were a compelling interest of such surgical giants as Lister [4]. Lister has introduced the concept of antisepsis in 1867 [3]. He promoted, also, the chrome treatment of catgut to delay its absorption in tissues and thus contributing to longer lasting strength of sutures [3]. The use of silk for ligatures was mentioned by Galen [1]. Halsted was instrumental in establishing silk as an important suture [1], [5]. Halsted was the first in the United States of America to promulgate the philosophy of safe surgery. He advocated the gentle handling of tissues, careful hemostasis, the avoidance of dead space and a meticulous surgical technique at a time when speed, inattention to hemostatsis and rough handling of tissues were the rule [5]. He also improved methods of sterilization. Gold wire sutures were first utilized in the sixteenth century and silver wire in the nineteenth century. In the thirties, interest was generated in cotton sutures [1], [3], [4]. At that time, the use of stainless steel wire was popularized ; it came into use around 1930 [1], [3].

The end of the first period saw the appearance of new fibers as the result of the fast growing polymer science and technology. The characteristic of the first period was that, whatever fiber became available, it was tried in operations, although none of the products were designed or developed for surgical application. Perhaps, this may be why everyone is still looking for the ideal suture, the elusive dream [1]. Until 1930, the suture materials used were predominantly catgut and silk and to a lesser degree, linen and cotton [3]. Synthetic fibers were introduced on a large scale during and after the world war II, starting with nylon in 1941 [3]. A large number of suture materials with various physical and biological properties flooded the market. They appeared first in the textile market and their use as suture materials came as a secondary offshoot.

The second period starting in the fifties is characterized by efforts toward synthesizing new polymers specially suited for surgical suture applications as the demand for a synthetic absorbable surgical suture material has stimulated research in different laboratories [1], [3]. Therefore, the first product possessing reasonable physical and biological properties was created in the sixties by Du Pont research laboratories. It was a braided polyester suture made of poly-L-lactide. The suture was strong and inert but absorbed extremely slowly in comparison with the absorbable sutures available at that time, catgut and collagen. The new polyester sutures commercially produced later on, are degraded by hydrolysis, introduction of water and do not require cellular activity as catgut and cotton [1].

Although attempts were made in the 1960's to perfect the absorbable synthetic suture , it was until 1970 that the synthetic suture made of the polyglycolic acid polymer was introduced by Davis and Geck laboratories. They presented a synthetic absorbable suture: Dexon [3], [4] A second synthetic absorbable suture, polygactin was introduced in 1974 by, Ethicon that presented polygactin 910, or Vicryl [3],[4]. In 1995, Lam [6], stated that many commercially available sutures exit to meet different needs of the surgeon because the ideal suture that meets all the requirements is yet to be developped. He suggested (PLLA) Poly (L-Lactic acid) as a biocompatible, biodegradable polymer with increasing possibilities for clinical applications.

The Polyester sutures, while they possess greater strength than catgut or collagen, are also much stiffer and harsher fibers. The suture has to be braided to provide good handling characteristics and carefully tied to avoid loosening of the first throw. So, in order to overcome the abrasive quality of these fibers and improve tying, an absorbable coating was developed and successfully tested clinically that allows sliding of the knot for better control [1], [2], [3].

Until recently, among the nonabsorbable materials, only commercially available polymers or fibers had been used for sutures. However, all nonabsorbable monofilaments are still wiry and more difficult to tie than braided materials [1,2]. Recently, fabrics of Dacron, nylon, Teflon, Polypropylene as well as metals such as stainless steel have been widely used as suture materials and as reinforcing meshes in the repair of diaphragmatic defects and in abdominal repair of hernia [7].

Although the second period of suture development is only twenty five years old, it is amazing to see how greatly polymer chemistry has revolutionized the manufacture of sutures, and it is still only the beginning. The selection of proper molecules as building blocks and their well designed architectural arrangement into unique structures provides a powerful capability for meeting any reasonable combination of physical and biologic properties for a great variety or novel medical and surgical products [1], [2], [7].

1.4 Literature Background

1.4.1 Mechanical Studies

In 1938, Taylor compared the square, surgeon's and the "useless square" sliding knots so-called at that time, with two throws between wax-coated, untreated silk and catgut sutures to conclude the superiority of the square knot over the other two and suggest a triple throw squared knot [8].

In 1948, in an attempt to offer practical suggestions that give surgeons better confidence in ligatures, Price [9], desribed the tension suture dealing with loops of square, granny and square-square , square-granny and granny-granny tension sutures. He measured the amount of tension needed to break these loops with a tensiometer.

In 1949, Douglas measured the tensile strength of surgeon's knot as compared to the straight pull based on the B.P.C. method for various suture materials [10]. In 1973, Herrman run experiments on the three squared throws knot, to evaluate the changes in breaking strength and knot security in the early postoperative period for silk, cotton, PGA, Dacron and plain gut sutures [11]. In 1974, Holmund provided a study on the physical properties of various commercially known suture materials [12].

Thacker, in 1975, offered a comprehensive analysis of the mechanical performance of the square, granny and surgeon's knot by determining the number of throws to reach knot break [13]. In 1977, Thacker compared the recommended configuration of a knot with specified suture materials as ascertained by mechanical performance tests to that employed by a group of board-certified surgeon's [14]. In the same year, Holmund came up with a model study of silk knot properties, using the three square throws knot [15].

In 1980, an important characterization of suture materials based on their mechanical properties was given by Chu [16]. Then, Trimbos studied the security of various knots, square, surgeon's and sliding for the first time in 1984; to go on with the study of the performance of the sliding knots in monofilament and multifilament suture materials in 1986 [17, 18]. In 1987, Gunderson proposed the rational alternative to the square knot: the half-hitch or sliding knot [19] . In 1989, Chu published a study of the quantitative evaluation of stiffness of commercial sutures [20].

In 1990, Trimbos et al performed a comparative study of the mechanical performance of both the sliding and square knots [21]. In 1991, Bouwers tested the square and sliding knots under dynamic loading [22]. In 1992, Brown studied the performance of square, surgeon's and double knots [23]. In 1993, Tomita analyzed the handling characteristics of braided suture materials [24]. In 1994, Tomita et al, studied the effects of cross-sectional stress-relaxation on handling characteristics of suture materials [25]. In the same year, Greenwald made a comparison of ten suture materials before and after in vivo incubation [26] and Israelsson provided a study of the physical properties of self locking and conventional surgical knots [27]. In 1996, Dinsmore, in an attempt towards understanding surgical knot security, offered a proposal to standardize literature [28] .

1.4.2 Biological Studies

In 1943, Localio studied the bacteriology and pathology of wounds contaminated with bacteria in relation to suture materials [29]. An experimental and clinical evaluation of surgical suture materials was made in 1953 and 1958 Madsen [30-32].

In 1955, Sewell and Wiland came up with a new method of comparing sutures of ovine catgut with those of bovine catgut in three species [33]. In 1959, Postlethwait evaluated surgical suture materials on the basis of tensile strentgth as well as tissue reaction [34]. In 1961, James studied infected wounds with inoculated sutures in mice [35].

Haxton, in 1965, discussed the influence of suture materials and methods on the healing of abdominal wounds [36]. The role of percutaneous suture in surgical wound infection was investigated by Carpendale, at the same year [37]. The next year, Alexander studied the role of suture materials in the development of wound infection with silk, silicone treated silk, wax treated and untreated twist silk sutures [38]. In 1967, a quantitative study of impaired healing resulting from infection was made by Smith [39]. Postlethwait, 1969, performed a long term comparative study of nonabsorbable sutures on the basis of strength and tissue reaction [40].

In 1973, Edlich investigated the role of the physical and chemical configuration of sutures in the development of surgical infection [41]. Following, in 1974, Postlethwait studied the human tissue reaction to various suture materials [42] . Whether the choice of suture material affects the wound infection was searched in 1975, Mouzas [43]. In 1980, Mcgeehan studied the relationship between synergistic wound sepsis and suture materials [44]. To determine another factor in suture induced infection, Katz studied the bacterial adherence to sutures at the same year [45].

In 1982, Greany performed a clinical and experimental study of suture sinuses in abdominal wound [46], followed by Sharp at the same year too [47] who studied suture resistance to infection. In 1988, Gittes incorporated nonabsorbable suture transcutaneously in rats and studied them histologically [48]. In 1989, Trimbos and Rijssel studied the tissue reaction to surgical knots: the effect of suture size, knot configuration and knot volume [49]. In 1993, Ahlberg, compared the mono- versus the multifilament absorbable sutures for abdominal closure [50].

1.5 Towards Sliding Knots: The "Alternating Strands with Different Patterns" Combinations

1.5.1 Basic Concepts And Definitions

Sutures remain the most common method of approximating the divided edges of tissue. Ideally, the physician should construct a knot that can be advanced to the wound surface, providing a preview of the apposition of the wound edges. When the advanced knot provides meticulous coaptation of the wound edges, the physician can construct additional throws to the knot that prevent slippage and ensure knot security [51].

A suture is defined as a thread that either coapts adjacent cut surfaces of the wound or compresses blood vessels to stop bleeding. The mode of operation of a suture is the creation of a loop secured by a knot. A tied suture has three components, as shown in Figure 1.1 [51, 13]. First, the loop, created by the knot, maintains the apposition of tissue. Second, the knot is composed of a number of throws snugged against each other. A throw is a wrapping of weaving of two strands. Finally, the ears are the cut ends of the suture. The physician's side of the knot is defined as the side of the knot with the ears, or the side to which the tension is applied during tying. The patient side is the side of the knot adjacent to the loop.

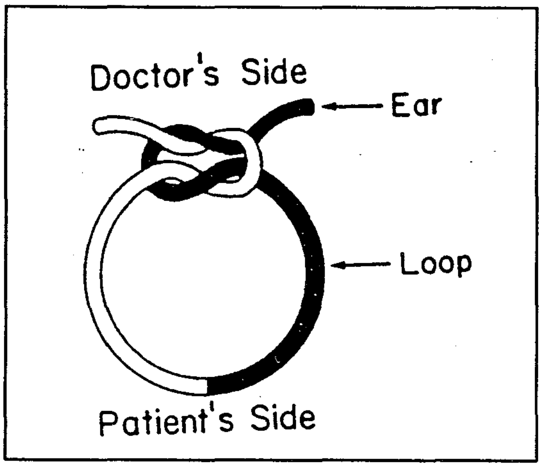
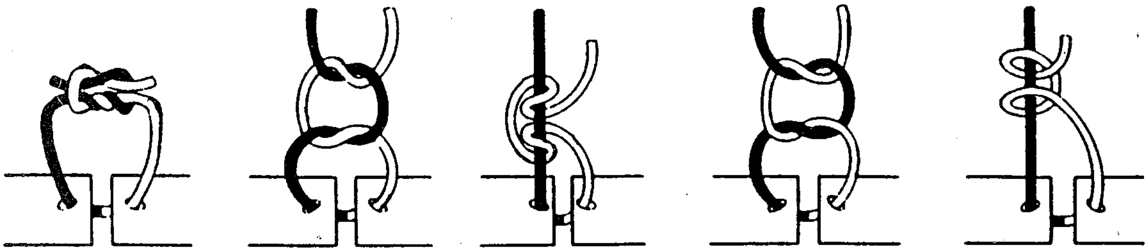


Figure 1.1 Components of a tied suture [51].

1.5.2 Configurations of Knots and Applied Nomenclature

1.5.2.1 Knots Configurations

The configuration of a knot can be classified into two general types by the spatial relationship between the knot ears and the loop. When the right ear and loop of a two-throw knot exit on the same side of the knot and are parallel to each other, the type of the knot is square [51], [28], [22], [13], as shown in Figure 1.2 [51]. The left ear and loop come out from the square knot in a position that is directly opposite to that of the right ear and loop. Tension is applied to the ears in directions that are perpendicular to the wound in horizontal planes that are parallel to the underlying tissue [51], [19] as illustrated in Figure 1.3 [19]. The knot is considered a granny type if the right ear and loop cross or exit different sides of the knot. A surgeon's knot is formed by making an initial double wrap throw, which is followed by a single throw, as shown in Figure 1.2 [51], [28], [22] . The relationship between the knot's ear and loop permit the surgeon's knot to be classified either as surgeon's knot square or granny [51], [28].



SURGEON'S KNOT SQUARE 2=1

SQUARE KNOT 1=1

SQUARE SLIP KNOT 1=1

GRANNY KNOT 1x1

GRANNY SLIP KNOT 1x1

Figure 1.2 Different knots configurations [51]

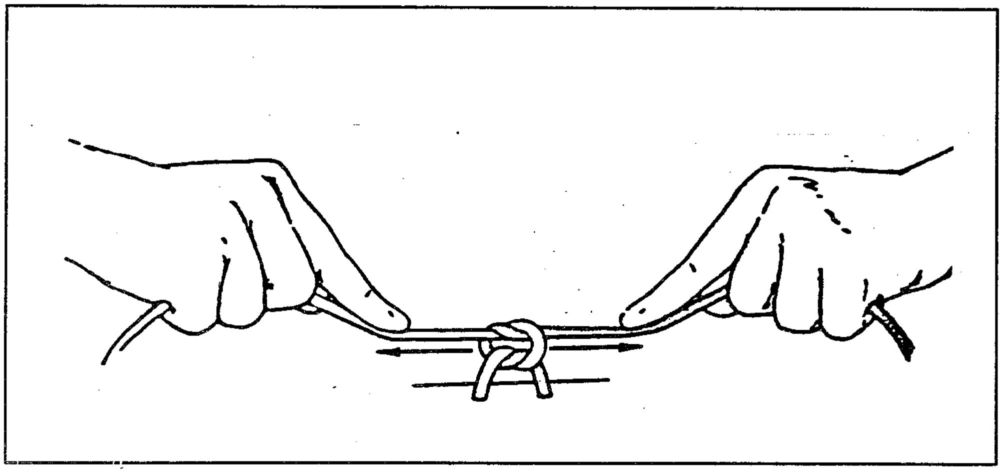


Figure 1.3 Tying a square knot, the ears are pulled down in a line perpendicular to knot axis and in horizontal plane parallel to the tissue [19].

Traditionally, square knots are used to tie sutures. The knot consists of two or more level throws. Each alternating right- and left hand throw adds another bend to each strand, thus adding to the security of the knot. However, tying a square knot is no simple matter. In order for the throw to be snugged down square and level, tension on each strand must be applied in a straight line centered on the knot. If tension on both strands is not applied in a line or if the line is not perpendicular to the axis of the knot, the snugged down throw can only become a sliding knot [51, 28, 22, 19, 18] Figure 1.4 [19]. In a sliding knot, there is no need to keep the strands in line when snugging down a throw, so, it is simpler to tie. It does not require as many throws to make a sliding knot as secure as a square knot. The addition of one more throw to a sliding knot yields equivalent security to square knot.

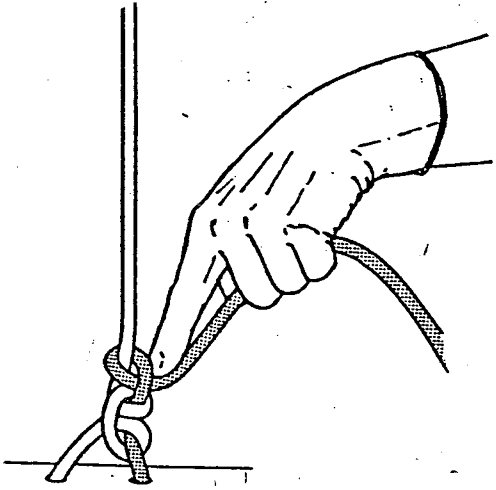


Figure 1.4 Schema of a sliding knot [19].

When compared to each other, in the flat or square knot, the threads reach from one side of the throw to the opposite side. In a sliding knot, the threads enter and leave the throw at the same side, as shown in Figure 1.5 [18].

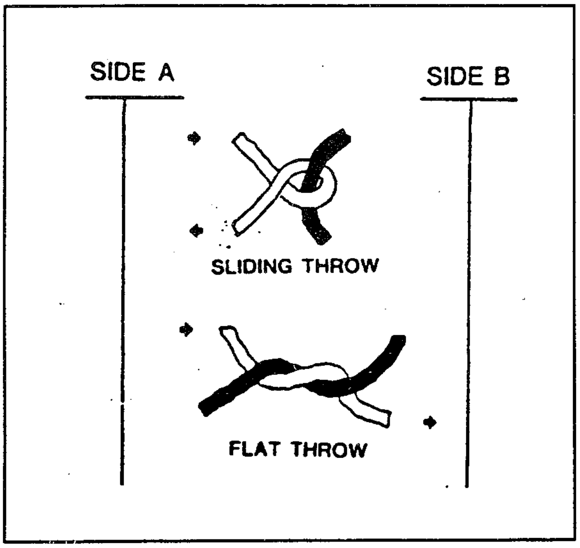


Figure 1.5 Configuration of sliding and flat knots [18]

Many other configurations that are derived from either the square or the sliding knot are used by surgeons. In his study, Trimbos [17], stated that during abdominal hysterectomy, as many as 60 knots might be used, accounting for one-sixth of the total duration of the operation at an operating time of roughly one hour and a knot tying time of ten seconds each. He tested twelve kinds of knots commonly used in surgical practice. They are shown in Figure 1.6 [17].

In an attempt to compare the knot configurations used in surgery to other currently used configurations, different marine knot configurations are shown in Figure 1.7.

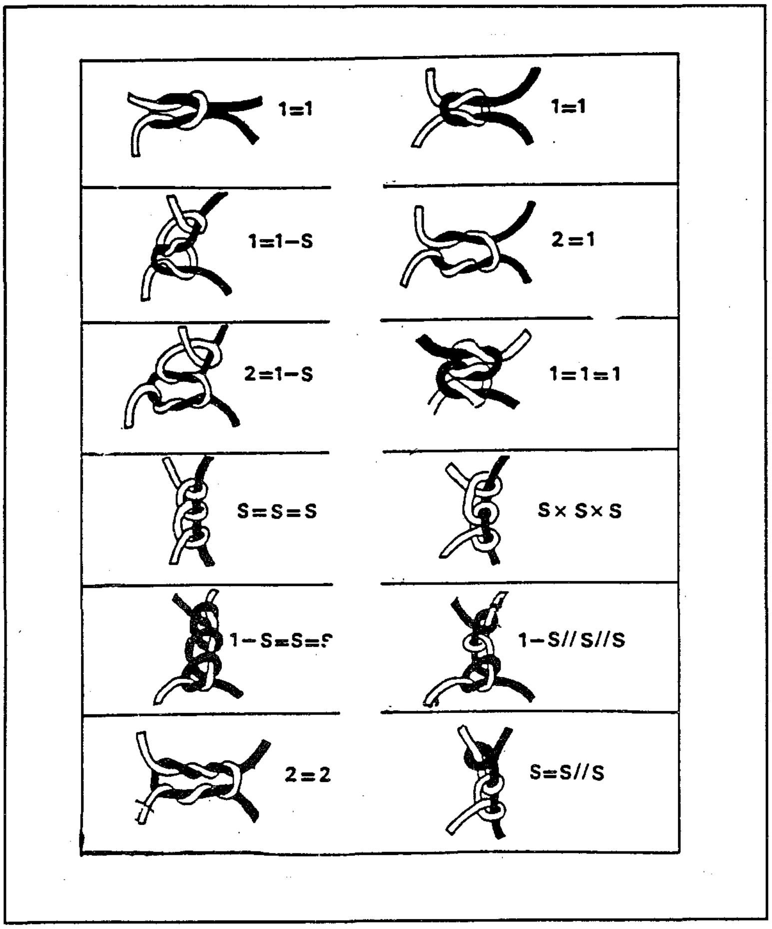


Figure 1.6 Knot configurations tested by Trimbos, 1984 [17]

Figure 1.7 Marine knot configurations

1.5.2.2 Knots and Nomenclature

To establish a nomenclature that is both easily understood and that can be used to describe all of the commonly used surgical knots, it is necessary to consider not only the various types of knots, but their relationships to one another. The commonly used surgical knots arranged according to their structure are illustrated in Figure 1.8 [28].

As Dinsmore has stated [28], there was a nomenclature used for describing flat knots (square). It was introduced by Tera and Aberg in 1976. A standard nomenclature for sliding knots was developped by Trimbos in 1984 [28], [17]. Tera's nomenclature was based on the knots appearance, whereas Trimbos codified the knots on the basis of the way they are formed, rather than the appearance alone. The conflict arises from the fact that sliding knots are derived from square and granny knots.

Granny knots are composed of two identical throws, consisting of one turn wrapped in the same direction. Using Tera's nomenclature, this knot is symbolized as 1X1 as shown in Figure 1.9 [28]. This is because according to this nomanclature, the ear and loop exit at

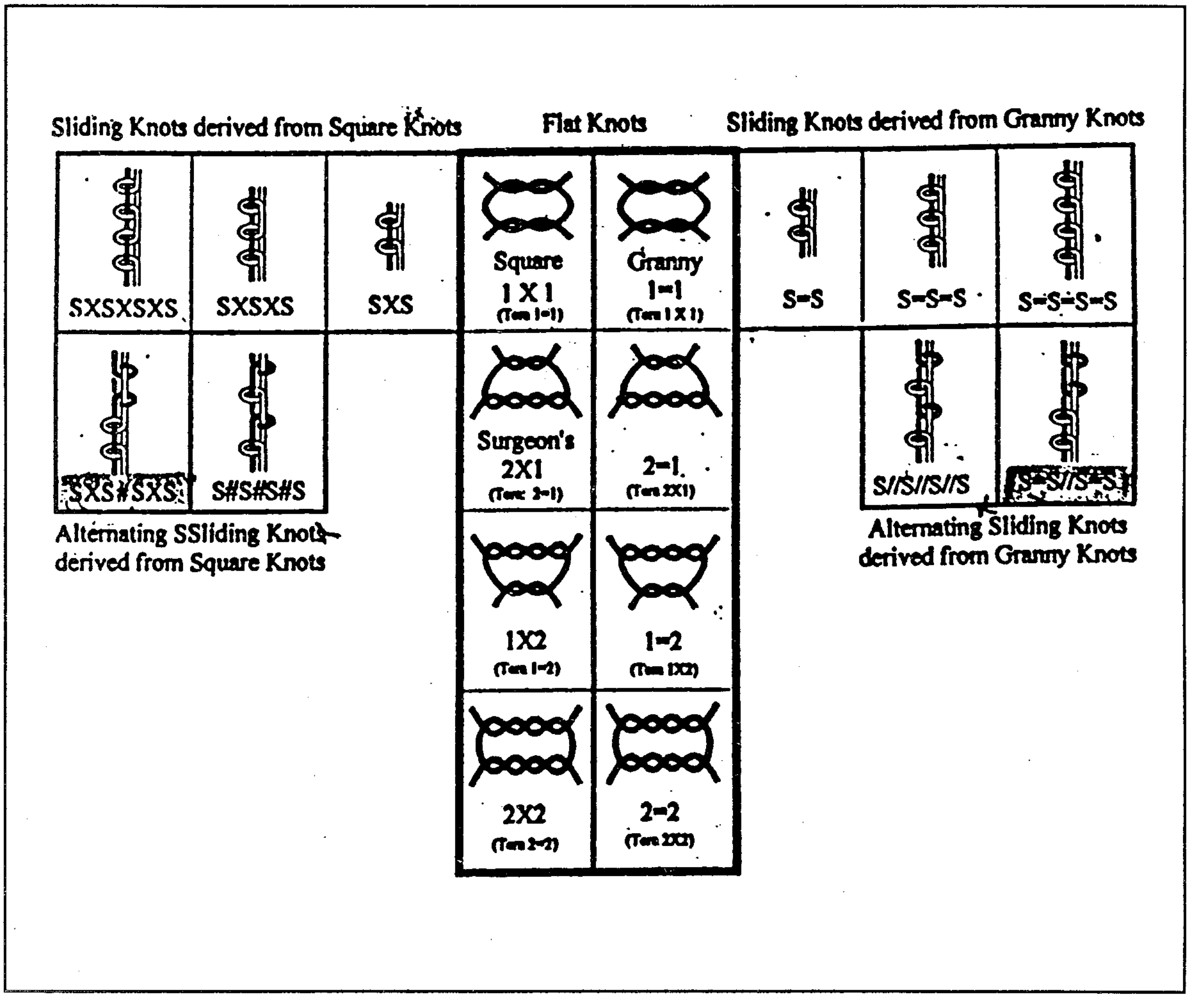


Figure 1.8 Structures of common surgical knots [28].

different sides of knot. But according to Trimbos, it is codified as 1=1 because each throw turns in the same direction as the preceding one (counterclockwise). When a granny knot is converted to a sliding knot, the knot formed is codified as S=S because each throw is identical to the one preceding it Figure 1.9. The number of wraps involved in each throw is indicated by appropriate arabic number and S is used to denote that the knot is sliding. The = symbol denotes that different entrance and exit of ear in relation to the loop in Tera's nomenclature while it indicates identical throws in same direction in Trimbos's. Therefore, the square knot, wher each throw turns in opposite direction of the preceding (right over left, left over right) is codified 1X1 according to Trimbos.

Square knot	Sliding knot	Granny knot	Sliding knot
Tera: 1=1	Trimbos: SXS	Tera: 1x1	Trimbos: S=S
1st throw: CCW	1st throw: CCW	1st throw: CCW	1st throw: CCW
2nd throw: CW	2nd throw: CW	2nd throw: CCW	2nd throw: CCW

CW: Clockwise, CCW: Counterclockwise

Figure 1.9 Conversion of flat knots into sliding knots [28]

A unified nomenclature based on the actual structure of the knot is used by Dinsmore [28]. Using this nomanclature, the = sign means that the subsequent throw turns in the same direction as the one preceding it. The X sign means in the ooposite direction. For sliding knots, in which the strand alternates, the // sign is used to show the change when the following throw turns in the same direction . When the axial strand changes and the throw turns in opposite direction the # sign is used.

1.5.2.3 Towards Sliding Knots

It is observed that publications on knot reliability and security dealt only with square knot, granny knot or surgical knots, frequently of a complicated type, such as the double square or double-double granny and triple surgical square knots... Tying of these knots is relatively time consuming and involves as mentioned the prerequisite that the knot be snugged down with the thread ends in opposite directions within the same plane [19].

In the daily routine of the operating thearter, the sliding knot use is observed too and though most surgeons warn against it, the prevalence of this knot in actual use is high. That is when most surgeons keep constant tension on one strand of the knot instead of both to prevent the first throw from loosening, this, inevitably results in the formation of a sliding rather than a square knot [17]. In his study, Trimbos [17], questioned gynecologists about the type of knot they mostly use. He found out that 80% of them used sliding knots. Still more striking was the finding that most of them interviewed were convinced that they made square knots. So, they confirmed Haxton's statement that "the use of methods and materials for suturing is usually a matter of habit, guesswork or tradition."

The reason sliding knots are preferred seems unclear. Crossing the sutures after the first throw unavoidably releases the tension applied to the knot, and this can easily lead to slippage of the first throw. Crossing of the surgeon's hands during knot tying induces clumsiness to a performance that should "do the job expeditiously with no loss of time and with great precision". Furthermore, the tying of deep seated ligatures of sutures is most easily performed by keeping one of the suture ends constantly under tension, and this also gives rise to a sliding knot [17].

Therefore, considering the frequency with which these knots are used, it is surprising that they have not been more closely examined. Studies on the physical qualities of the various sliding knots are still lacking [17].

According to Dinsmore [28], the performance of sliding knots must be further investigated in the area of testing existing suture materials. He went further to declare that there are no studies comparing knots in which the strand alternated using a pattern other than with every throw (example: SXS#SXS or S=S//S=S).

2. WOUND HEALING AND SUTURES

2.1 Introduction

A wound is a body injury caused by physical means. Wound healing is the process by which this injured part of the body is repaired. The Halsted technique [5], the most accepted technique, promotes rapid wound healing through certain practices: hemostasis, asepsis and gentleness to tissue. Hemostasis involves stopping the bleeding in the wound. Asepsis keeps the wound free from infection, and gentleness to tissue minimizes tissue trauma.

Sutures function to hold tissue together until wound healing takes place. Twenty-one days is often regarded as the critical wound healing period, during which time sutures provide strength to the wound after the incision is closed and allow tissue approximation with minimal scarring.

2.2 Primary Tissue Layers and their Characteristics

The characteristics of the tissue layer being sutured determine the type of suture to be used, as well as the method of suturing. The primary tissue layers are shown in Figure 2.1.

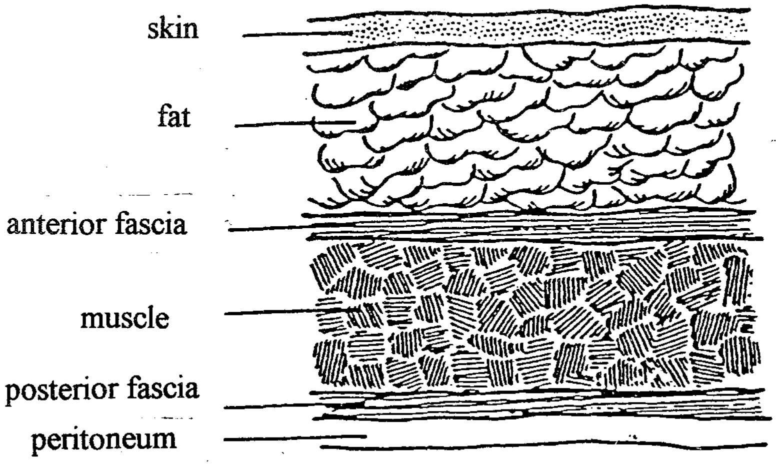


Figure 2.1 Primary tissue layers [52].

Tissue layers are either tough and fibrous, or soft and friable. Those of the abdomen, for example, include: peritoneum (soft), fascia (tough), muscle (soft), fat (soft) and skin (tough). The peritoneum is the soft membrane lining the abdomen. Peritoneum is a tissue that is commonly dealt with in surgery, and may be easily torn; because of this it is often sutured with the fascia layer. The word fascia means band, and it is the band of tough, fibrous connective tissue which surrounds a muscle or group of muscles. Due to its fibrous structure, the fascia is ideally suited for suturing. Muscle is the soft tissue with little holding power for sutures and as a result is seldom sutured except to eliminate dead space and to prevent fluid accumulation in tissue. Because muscle consists of long, thin cells, sutures tend to pull out under moderate tension. Fat is the soft, friable subcutaneous layer which is often sutured to prevent dead space. Skin is the tough, outermost covering of the body. This layer is always sutured, stapled, taped or closed with a combination of these methods.

2.3 Phases of Wound Healing

Healing restores the continuity of tissues after trauma or surgery. It begins at the moment of injury when vessels are divided, blood is shed into the wound, and hemostasis occurs. The blood supply at the injury site is diminished by thrombosis of vessels, thus the area becomes hypoxic and acidotic. Platelets are activated and release series of growth factors which initiate the reparative events. Within few minutes, inflammatory cells enter the wounded area attracted by complement components and degradation products of fibrin and fibrinogen. First, the granulocytes appear; they provide nonspecific immunity to infection. Macrophages which collect in the next few days are critical to repair. Lymphocytes are also capable of exciting reparative reactions. A "unit of healing" is made up of a sequence of cells led by macrophages and followed by capillaries and immature fibroblasts. Synthesis of collagen begins in a few days after wounding. It is deposited in a disorganized pattern until the second week. From about the third week, the collagen network is rebuilt in more organized way and better meets the stresses applied to the scar [53].

The healing process can be divided schematically into three different phases: the inflammatory phase, the fibroplasia phase and the maturation phase.

The inflammatory phase or exudative phase : During which, the biological cleansing of the wound takes place. Chemical and nervous stimuli cause changes in the circulation with increased vascular permeability in the injured area. Cellular and humoral defense mechanisms against bacteria, foreign bodies and damaged cells are therefore activated in the area of injury. During the inflammatory phase, the wound is characterized by the classical signs of inflammation: redness, swelling, pain, increase in temperature and impaired function. This phase lasts for two or three days. It increases in degree and duration with presence of foreign body and contamination by bacteria. When prolonged, it results in retarded development of strength in the wound, as shown in Figure 2.2 [53].

Fibroplasia phase or proliferative phase: During this phase, the defect is bridged by capillaries and fibroblasts growing from wound surfaces. The newly formed tissue is called granulation tissue. The strength of the wound depends mainly on the collagen fibrils and fibers which are formed by the fibroblasts and deposited in the intercellular substance. This phase lasts for several weeks, during which, the signs of inflammation subside. During this phase also, there is a rapid increase in the strength of the wound. Several factors can impair this phase, like a decreased oxygen tension, anemia, hypoproteinemia, advanced age, zinc deficiency. Figure 2.3 shows the importance of impaired fibroplasia in the development of wound strength.

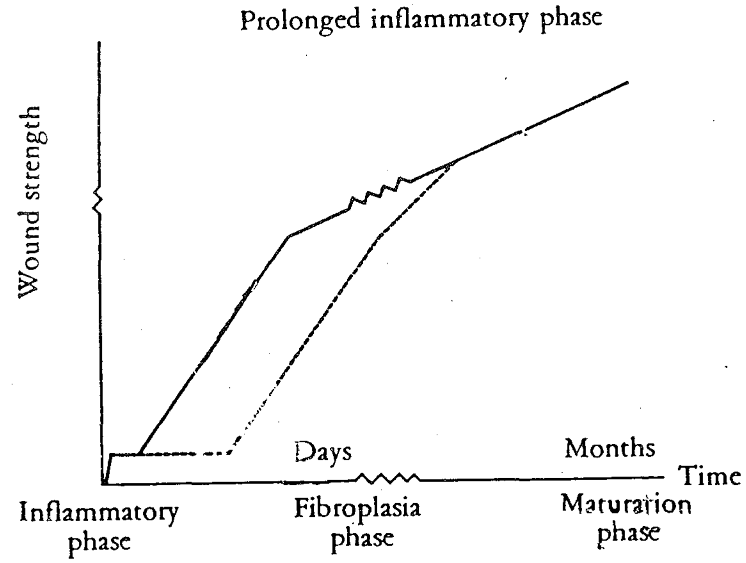


Figure 2.2 Normal strength development in a wound. The broken line shows the case of prolonged inflammatory phase [53]

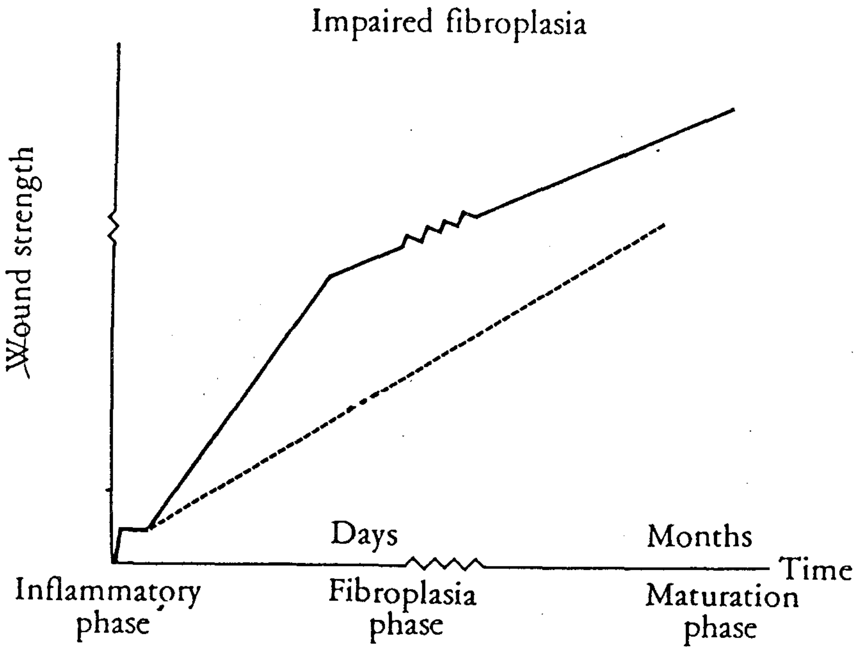


Figure 2.3 Normal strength development in a wound. The broken line shows alteration in impaired fibroplasia [53]

The Maturation phase: During this long-lasting phase, the strength of the scar increases without a concomitant increase in the quantity of collagen. This increase is therefore, due to restructuring of the collagen fiber network. Along with, the scar shrinks and the vascular bed of granulation tissue decreases. The scar line turns into a white streak. Impairment of this phase may occur because of vitamin C deficiency.

2.4 Wound Strength during the Healing Phases

At the beginning, before new fibrous tissue bridges the wound cleft, the strength of the wound is completely dependent on the suture. The suture holding capacity of all sutured tissues decreases during the first three to four days after wounding due to lysis of preexisting collagen. During this time, wound strength diminishes consequently. As fibroplasia proceeds, strength increases and supportive role of sutures becomes progressively smaller. During the first twelve to fourteen days, the rate at which strength increases is almost the same irrespective of the tissue which is injured. During the maturation phase, the increase in strength depends to some extent on tissue and on the stresses that the wound encounters.

2.5 Suture- Related Healing Complications

2.5.1 Wound Dehiscence

The integrity of the wound is completely dependent on the sutures until the reparative tissue has bridged the wound and matured to reach adequate strength. Wound dehiscence occurs when sutures are unable to hold the wound edges together until the wound has acquired sufficient strength of its own through the healing process [53]. This can be due to:

1- Insufficient original suture strength or insecure knots
2- Premature loss of suture strength, or its premature removal
3- Cutting of the suture through the tissue
4- Delayed healing

Improper suture knotting techniques and inadequate selection of knots may be the cause of suture knot failure. Premature loss of strength is a result of suture being absorbed by tissue earlier. Sutures cutting through the tissue is probably the most common cause of wound dehiscence. The causes of which are not fully understood. One important factor is weakening of supportive tissue by lysis of collagen where infection and inflammation are the causes of this weakening process. Sometimes, sutures tied so tightly are responsible too [52, 53].

2.5.2 Wound Infection

The risk of wound infection increases if the contamination of the wound increases. Bacterial contamination is the important factor. Tissue injury and less suitable suture material increase the risk of significant infection by impairing the body's defense against bacteria. Braided, nonabsorbable suture materials seem to involve a distinctly high risk of fostering infection. Coating them may be beneficial.

2.5.3 Suture Granuloma

A suture granuloma is a painful swelling related to buried suture. The cause is a low-grade infection around the suture. It can also be due to mechanical or chemical irritation by the sutures.

2.5.4 Sinus Formation

With more intense inflammatory reaction around a suture there can be a sinus formation in the scar. The suture thread can migrate through the opening-suture extrusion. Such an extrusion can only occur when the tissue supporting the suture has been broken down and is brought to quick healing if the suture is removed.

2.6 Types and Characteristics of Sutures

There are two basic types of sutures: absorbable and nonabsorbable [52, 53]. Absorbable sutures are either digested by body enzymes or broken down by hydrolysis within the body and absorbed by the tissue. Nonabsorbable sutures, on the other hand, remain in the body unless they are removed, although certain types do break down within the body over a prolonged period of time. The two major types of absorbable sutures are: surgical gut and synthetic absorbable sutures. Synthetic absorbable sutures are made from synthetic materials, such as polyglycolic acid (PGA). These sutures are absorbed by the process of hydrolysis, which means that the material is broken down by contact with body fluids.

There are various types of nonabsorbable sutures available ,including: silk, cotton, synthetic nylon, polyester, polypropylene and stainless steel. Nonabsorbable sutures in the skin are normally removed, while those within the body in deeper tissues become "encapsulated" so that tissue grows around the sutures and they remain in the body in this form.

In general, the largest potential users of absorbable sutures are: obstetrics an gynecology, urology, general, orthopedic and plastic surgeons. Nonabsorbable sutures are also used more frequently for particular surgical situations. For example, vascular surgeons use them to anastomose vessel-to-tissue and vessel -to-prosthesis. Plastic surgeons and emergency physicians use nonabsorbable sutures for skin closure, and ophthalmic surgeons for traction sutures, retinal detachment, corneal transplant and cataract surgery. The following Table compares absorbable and nonabsorbable sutures.

Table 2.1
Scheme of types and uses of absorbable and nonabsorbable sutures [53].

Type	Characteristics	Examples	Surgical specialties
Absorbable	Digested by body enzymes or broken down by hydrolysis then absorbed by the tissue	Gut, synthetics	OB/GYN, urology general, orthopedic and plastic surgeons
Nonabsorbable	Remain in the body unless removed	Silk, cotton, nylon, polyester, polypropylene, stainless steel	vascular, general, ophthalmic, plastic emergency physicians neurosurgeons

2.6.1 Suture Sizes

Sutures come in various sizes depending on their usage. The sizes range from 7, the largest to 11-0, the smallest as shown in Table 2.2 [52]. It is depending on the diameter of the suture thread that the correspondent size is given. For example, going from size 11-0 to 7, the diameter gets larger meaning the thread is getting thicker. The diameters of sutures correspond to the type of surgery to be performed. They are standardized by the United States Pharmacopoeia (USP) [52], [53].

Table 2.2
Suture size and usage [53].

	Suture size	Possible surgical use
smallest diameter	11-0, 9-0, 8-0, 7-0, 6-0	Most frequently used in microsurgery/ ophthalmic surgery
	5-0, 4-0, 3-0, 2-0	Most frequently used in plastic/ cuticular surgery
	0, 1, 2, 3, 4	Most frequently used in general closure, obstetric/gynecologic surgery
largest diameter	5, 6,7	Most frequently used in retention

2.6.2 Suture Construction

Sutures are constructed either as monofilaments or as multifilaments. Monofilaments consist of a single suture filament and are known for their smoothness through tissue. Multifilaments, consist of a number of strands braided or twisted together. Multifilament sutures are known for their handling properties.

2.6.2.1 Suture treatments and Coatings

Manufacturers may coat or treat sutures to facilitate their ease of handling, to improve their smoothness as they pass through tissue, or to limit capillary action. Capillarity in this case refers to the ability of infection to travel along the suture strand. Polyglycolic acid suture is treated with polycaprolate, which acts to reduce friction. Silk is treated with silicone or wax. This makes it easier for the suture to pass through tissue.

2.6.2.2 Suture Sterilization

Sutures are sterilized to reduce the possibility of the introduction of infection in the wound. Manufacturers use two methods to sterilize sutures: ethylene oxide and irradiation.

2.6.2.3 Physical Properties of Sutures

The important physical properties of sutures include:

- Tensile strength
- Flex-life
- Handling and flexibility
- Knot security
- Smoothness through tissue

Tensile strength is a measure of how pull a suture can withstand before it will break. The tensile strength of sutures can be measured at various times after the suture has been used to close a wound to determine how long the suture retains its strength in tissue. An in vitro technique involves measuring the tensile strength under dry conditions. An in vivo technique involves placing sutures within lab animals for a specified period of time, then retrieving them and measuring tensile strength.

Flex life of a suture is its ability to withstand repeated bending without breaking. Handling refers to those properties which made a suture easy for surgeon to work with. These properties include: flexibility (ability to bend), elasticity (ability to stretch and rebound), texture (softness to feel), and knot tying characteristics.

Knot security is the ability of a suture to hold a knot without slipping. This is an important characteristic of sutures because complications in wound healing may develop if knot slips.

2.6.2.4 The Perfect Suture

There are currently more than ten different types of suture material in use. None of them are perfect, but some have more of the characteristics which might make up an ideal or "perfect" suture. These characteristics are varied [52, 53].

The ideal material would have excellent handling, meaning be pliable. It would not fray as knots are snugged down and tied and there would be no tissue drag. Knot security would require no extra throws or change in surgeon's technique. It material would have excellent strength. It would cause little or no tissue reactivity. Suture construction would provide no "nooks and crannies" to which necrotic tissue, clots and bacteria could adhere.

The ideal suture should be inert, nonallergenic, sterile and economical. Overall it should be used in contaminated as well as in clean wounds, in every surgical situation and with equal success.

3. TENSILE TESTING OF SURGICAL SILK AND NYLON SUTURES WITH CONSEQUENT EVALUATIONS

3.1 Introduction

In vivo, a suture is constantly subjected to changing tensile loads. It should have the same strength as that of the tissue, otherwise its failure can result in wound dehiscence, which is a serious complication in surgery. It should provide good resistance to the swelling pressure of the wound, keeping its elasticity and ability to elongate without permanent deformation.

To discriminate between suture materials, the study of mechanical properties would provide a good background to assist surgeons in the selection of appropriate materials for specific surgical applications.

Because surgical sutures are considered as fibrous materials, their most important mechanical properties are their tensile properties which determine their behavior under forces and deformations applied along the fiber axis. This chapter studies the mechanical parameters of silk and nylon sutures : apparent modulus of elasticity, maximal strength, yield strength, percentage elongation to the yield point, percentage elongation to maximum and toughness. The sizes chosen were 2/0 and 4/0 as assigned by the USP. The interpretation of the mechanical behavior of the two sutures is done in relation to the microstructural composition of each suture fibers and their organizational arrangement in the corresponding sutures.

3.2 Methods

The Nylon and silk sutures are subjected to tensile test using Zwick 1446 Universal Testing Machine. A gauge length of 40 cm is used for all the suture strands. Around 25 suture samples were tested for each of the corresponding 2/0 and 4/0 USP sizes for nylon and silk sutures respectively. The specimens are clamped using tapered screw-type specimen holders. The specimen is positioned symmetrically in the middle of the jaws and the clamping screws are rotated in equal amounts. The high clamping force required for holding the specimen is automatically achieved by the wedging action of the roller-mounted taper jaws, as the tensile force increases and the jaws move at a constant cross-head speed of 300 cm/min. The stress-strain curves are drawn during the test run and printed thereafter. The mechanical parameters are implemented or extracted from the curves when necessary. One-way Anova test is used for statistical analysis to compare, interpret and evaluate the results.

Young's modulus is measured as the slope of the linear or initial straight line portion of the stress-strain curve, as shown in Figure 3.1.[16], [54]

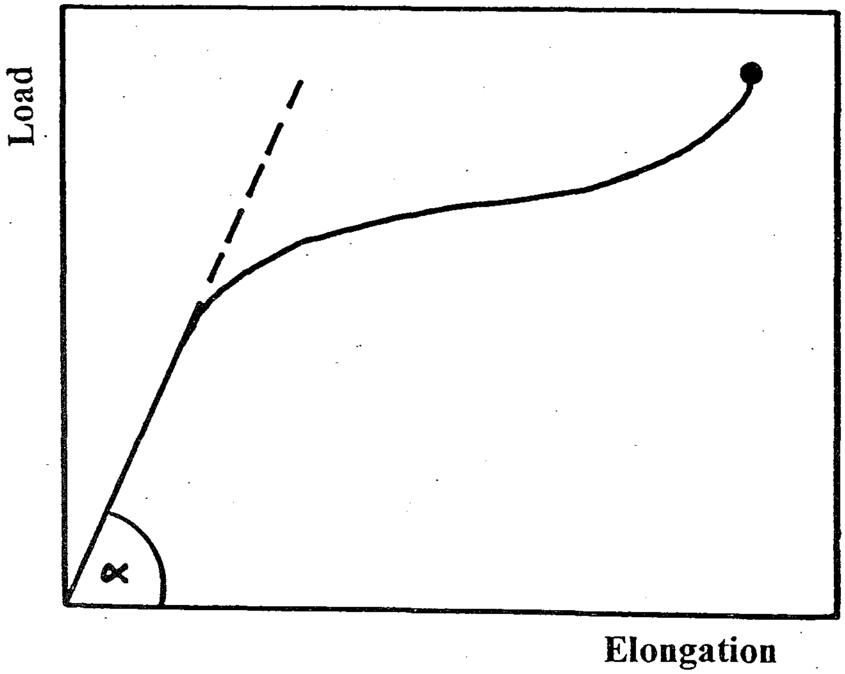


Figure 3.1 E-Modulus measurement.

The yield point is extrapolated using Coplan's method, i.e. taken as the intersection of the tangent at the origin with the tangent having the least slope as shown in Figure 3.2 [16], [54]/

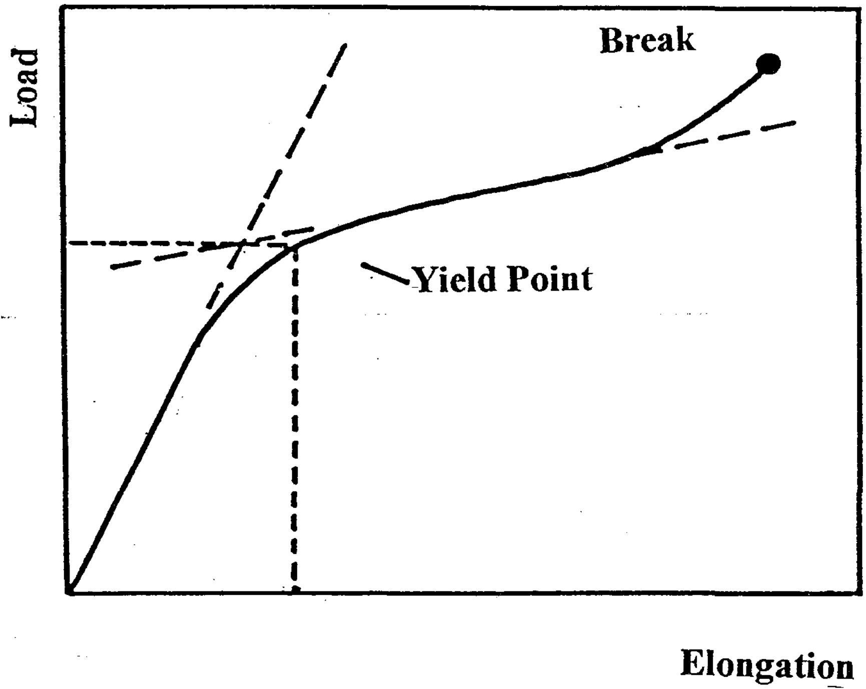


Figure 3.2 Extrapolation of the yield point using Coplan's method [16], [54].

Toughness or work of rupture is defined as the energy needed to break the specimen, or the energy absorbed by the specimen before rupture and is calculated as the area under the load- displacement curve (integral), (see Figure 3.3) [54].

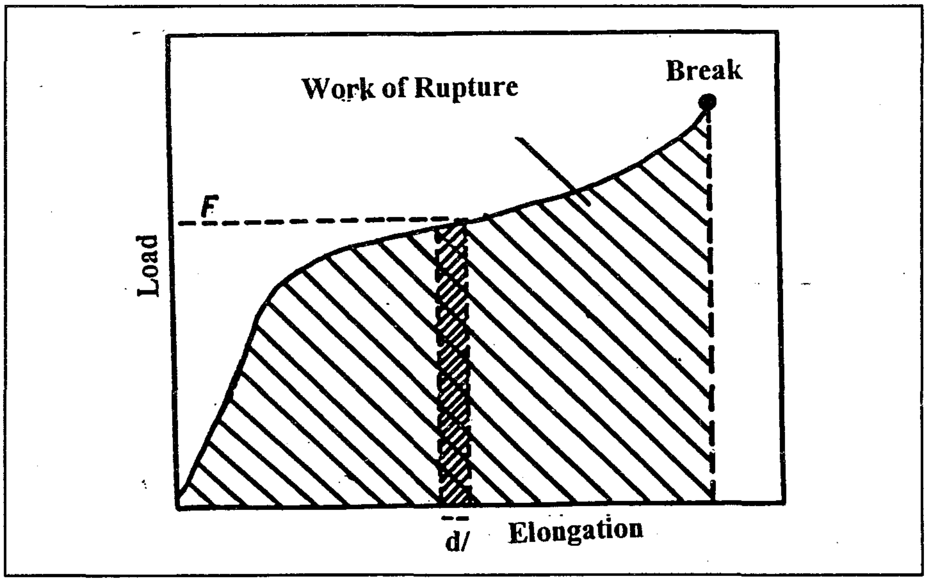


Figure 3.3 Scheme of toughness or work of rupture, being equal to the area under the load-displacement curve [54].

3.3 Results

The different mechanical parameters studied are shown in Table 3.1. Statistical analysis of the results is done using one-way Anova, followed by multiple range test:Tukey-HSD test with significance level 0.05).

Table 3.1
Mechanical Properties of Silk and Nylon Sutures of 2/0 and 4/0 Sizes

Size	E-Mod N/mm^2	σ_{MAX} N/mm^2	σ_{YIELD} N/mm^2	EL$_{MAX}$ %	EL$_{YIELD}$ %	Toughness N. mm
2/0 Silk	7344±1396	212± 1	98.7 ± 0.6	28±4	2.4±0.5	973±152
2/0 Nylon	1484±465	245±3	123±1	85±14	24±2	3563±730
4/0 Silk	6269±1037	242±1	121±1	23±4	3.5±1	342±72
4/0 Nylon	1777±577	338±2	206±1	66±11	20±3	1135±279

Figures are expressed as means ±SD.

Elastic modulus evaluation revealed that suture material behaved according to suture design, i.e. the braided silk suture demonstrated less compliance (higher E-mod. values) than monofilament nylon suture for both 2/0 and 4/0 sizes (the difference is statistically different). Silk demonstrated different modulus values between the 2/0 and 4/0 sizes ($p<0.05$), unlike nylon whose modulus values for both sizes were not significantly different.

According to Holmund [12], the relation between the stress and the strain is defined by the modulus of elasticity. Because the deformation most commonly considered is that of longitudinal tensile stress, the modulus that describe this is called Young's constant E.

$$E = \frac{Applied\ \ load\ \ per\ unit\ area}{Increase\ in\ length\ per\ unit\ length}$$

Increase in length per unit length is called elongation. He approximated the cross area to be similar for all the 3-0 suture. Then, the formula can be written

$$E = \frac{Applied\ \ load}{e} K$$

If the load is kept constant = 1 Kp

$$E = \frac{1}{e} K$$

That is , $1 / e$ can be used to describe the modulus of elasticity of different 3-0 suture materials. He recorded values for silk between 2 and 4, for nylon the values were between 59-81. Chu, [16], measured the modulus of elasticity in grams per denier (GPD), which described the amount of weight required to break a fiber or yarn of one denier (a denier is defined as the weight of a fiber or yarn 9,000 meters long). The modulus of elasticity for silk was 78.5 GPD, for nylon 19 GPD. These values are in agreement with the recorded values in this study since silk had a higher modulus than that of nylon.

The modulus of elasticity is a measure of the resistance to stretch or elongation before the yield point. Its reciprocal is called compliance. For nylon suture, it is calculated as the slope of the load displacement curve at the origin. For the braided silk suture, however, the origin is different from the nominal origin.

Due to the braided geometry, the silk suture shows the unique characteristic of braid called "crimp", which is the small curve region in the early beginning of the load-displacement curve indicated by OA, as shown in Figure 3.4, [54]. Sutures of monofilament form do not have this unique region. The crimp is usually removed by a small tension, therefore the modulus of elasticity is determined after the removal of any crimp.

Strength which is considered as a measure of the steady force necessary to break the specimen, is given experimentally by the maximum strength developed in a tensile test [54]. The maximum strength of nylon suture was higher than that of silk suture for both sizes, the difference is found to be statistically significant. as shown in Table 3.1. This agrees with the literature values. Chu [16], recorded stresses of 3.4 GPD for silk, and 6.25 GPD for nylon.

The elongation necessary to break the specimen is expressed as breaking or maximum elongation and is a useful quantity. Nylon was able to undergo more elongation than silk suture, with the ability to elongate decreasing with size, i.e. 4/0 size demonstrated less elongation than 2/0 (the difference being statistically different). While, silk suture's

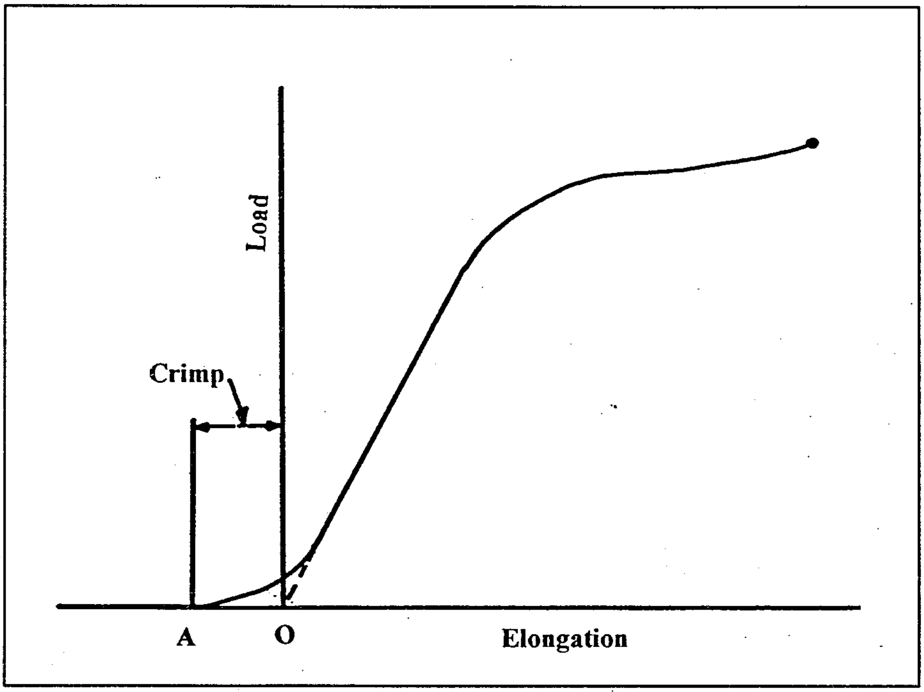


Figure 3.4 Representation of the crimp, small region indicated by OA [54].

ability to undergo elongation till maximum was not different among the two sizes, as shown in Table 3.1. Holmund [12], recorded the percent elongation to the elastic limit values for silk took from different manufacturers ranging from 2 to 4 %. He recorded higher values for nylon: 18, 35%.

The yield point is determined according to Coplan's method, as illustrated in Figure 3.2, and is important because for most materials, elastic recovery, which is good up to the yield point becomes less complete for higher strains due to plastic deformation. In practice, the point at which the permanent or plastic deformation starts to take place may be as important as the point at which breakage occurs [54]. Nylon suture showed higher yield strength than silk suture with the difference statistically significant, Table 3.1. Holmund results [12], agree well with ours, he recorded values of the elastic limit for silk : 1.0 Kp and for nylon : 2.1 Kp.

The degree of elongation undergone to the yield point was higher for nylon suture than the silk one, and only 2/0 nylon showed more elongation till yield than 4/0 nylon, with no statistical significance between the 2/0 and 4/0 silk sutures. Chu [16], gave similar results for silk. He recorded values of 1.9 % compared to 2. 4% . For nylon, he recorded a value of

2.2% which was too low compared with 20.7%. However, Holmund [12] gave a closer value of elongation % for nylon, 18%. For silk, he recorded values ranging between 2 and 4 %.

The toughness termed also the work of rupture gives the ability of the material to withstand sudden shocks of given energy. It represents the amount of energy absorbed till rupture, given as the area under the load-displacement curve, as shown in Figure 3.3 [54]. There was a wide difference in toughness values between the two sutures, nylon absorbed more than twice the energy absorbed by the silk suture, size 2/0, as shown in Table 3.1, the difference is statistically different. The 4/0 nylon suture showed less than half the energy absorbed by the 2/0 nylon suture, while the silk suture 2/0, absorbed a similar amount of energy to that absorbed by 4/0 nylon suture (the difference was not significant). The 4/0 silk suture toughness value was less than that of the 2/0's value. Chu [16], recorded the values of specific work of rupture in (N/Tex) x 10^{-2} as 2.36 for silk and 8.96 for nylon.

3.4 Discussion

Force- Displacement relationships are easily measured. The resulting curve represent basic mechanical relationships. From this curve, we can derive the parameters of strength, toughness, strain at yield, force at yield, strain at maximum or failure, maximal load as well as the elastic modulus. So, the true mechanical parameters of the single strands of sutures are represented. This may not represent the same parameters of the suture in vivo because, a suture used clinically may behave differently not only because it is subjected to knotting, but also because it is placed under tension.

Differences in the mechanical parameters between nylon and silk suture can be explained on the basis of the material composition and the microstructural organization of the fibers consisting each suture.

3.4.1 Suture Material Composition (Fiber Structure)

The silk suture is made up of silk fibers consisting of the two proteins Sericin and Fibroin. The sericin represents the amorphous or the noncrystalline part (random-chain protein cross-linked acting as a supporting matrix) while fibroin represents the crystalline fibers. The protein chains are fully extended with their axes parallel to the fiber axis as shown in Figure 3.5 [55].

At intervals chains will fold back upon themselves so that adjacent chains run in opposite directions (antiparallel). In this configuration, hydrogen bonds form easily between the peptide groups of adjacent chains, and several antiparallel chains may be bound together to form a parallel "pleated sheet" These sheets then stack one a top another to form crystallite. Sometimes, the axes of the extended chain lie perpendicular to the fiber axis instead of parallel to it Figure 3.6 . This is known as cross pleated sheet configuration [55].

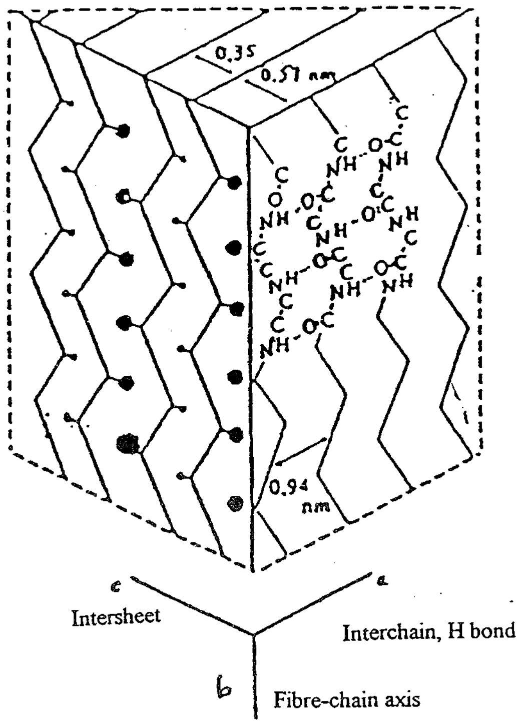


Figure 3.5 The parallel pleated sheet configuration, chain axis being parallel to fiber axis [55].

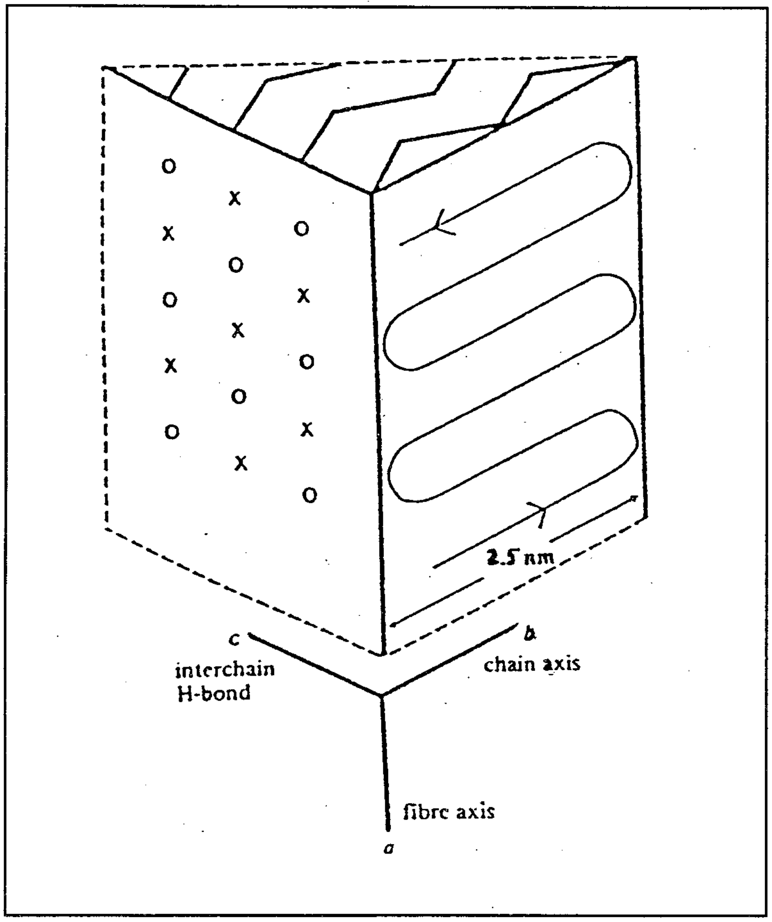
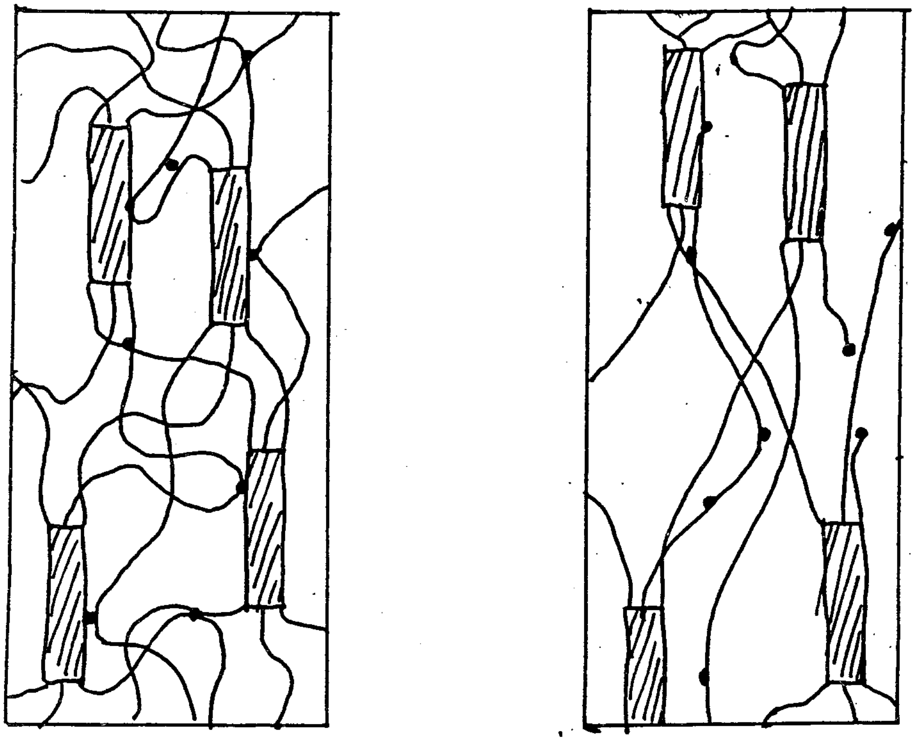


Figure 3.6 The cross configuration, chain axes are perpendicular to fiber axis [55].

Tension along the fiber axis is placed directly on the bonds holding chains together and can cause this cross configuration to unfold. This unfolding process involves a considerable extension of the crystallite along the fiber axis and this is reflected in the high extensibility of the material as a whole .

From these molecular studies a reasonably clear picture of the basis for the mechanical behavior of silk can be drawn: silk is partially crystalline polymeric material. Crystalline regions are accurately aligned with the fiber axis and are interspersed with regions of randomly arranged chains. As tension is applied to a silk fiber, the material deforms on a molecular scale and this deformation occurs primarily in the amorphous region rather than the highly aligned, stiff crystallites as indicated in Figure 3.7 [55]



(a) (b)

Figure 3.7 Representation of molecular events accompanying extension in silk
(a) unstressed silk, random chain bound to themselves and to crystalline regions.
(b) extended silk, chains extended and bonds are broken [55].

The elastic modulus and the viscosity of the material during this initial extension depends upon the extent to which the amorphous chains are bond to each other, the interaction of amorphous chains with the crystallites and the proportion of the amorphous material in the fiber. Bonds between chains and short chains connecting crystallites are broken before the bulk of the amorphous chains may extend. The strength of these bonds accounts for the high initial modulus of silk. Once these bonds have been broken, the amorphous regions will deform easily. As the material is further extended, the amorphous regions become aligned such that it may be possible for them to crystallize, hence producing permanent plastic deformation [55].

The nylon suture is made up of polyamides which are macromolecules whose structural units are interlinked by the amide linkage - NHCO-. The crystal structure of polyamides results from the conformation of the macromolecules. There exist intermolecular forces that are both van der waals' bonds and hydrogen bonds. The latter, entailing the NH and CO moities, cause the formation of sheet-like arrangements between adjacent chains. The stacking of these hydrogen bonded sheets controls the size and shape of unit cell. The general conformation of the chain segments in the unit cell and mode of packing are basis for classification of various polyamides. In nylon 6,6 and nylon 6, the most important for commercial fiber production, the chain segments are fully extended, and this is referred to as the alpha-structure, as shown in Figure 3.8 [56]

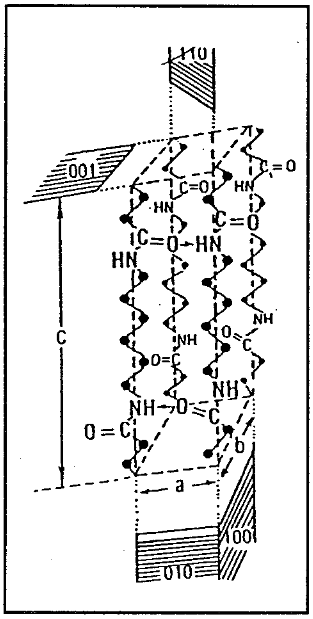


Figure 3.8 Unit cell of nylon 6,6 [56]

The alpha structure is comprised of stacks of sheets of planar hydrogen-bonded extended -chain segments. These sheets are characterized in nylon 6,6 by a parallel alignment of the adjacent extended molecules, which are spaced with a perpendicular chain-to chain distance and which are successively displaced in chain direction by a distance corresponding to one chain atom, as shown in Figure 3.8 [56]

The behavior of polyamides fibers is best explained by the models that assume a regular alternation of crystalline and amorphous layers within a microfibrillar structure, Figure 3.9

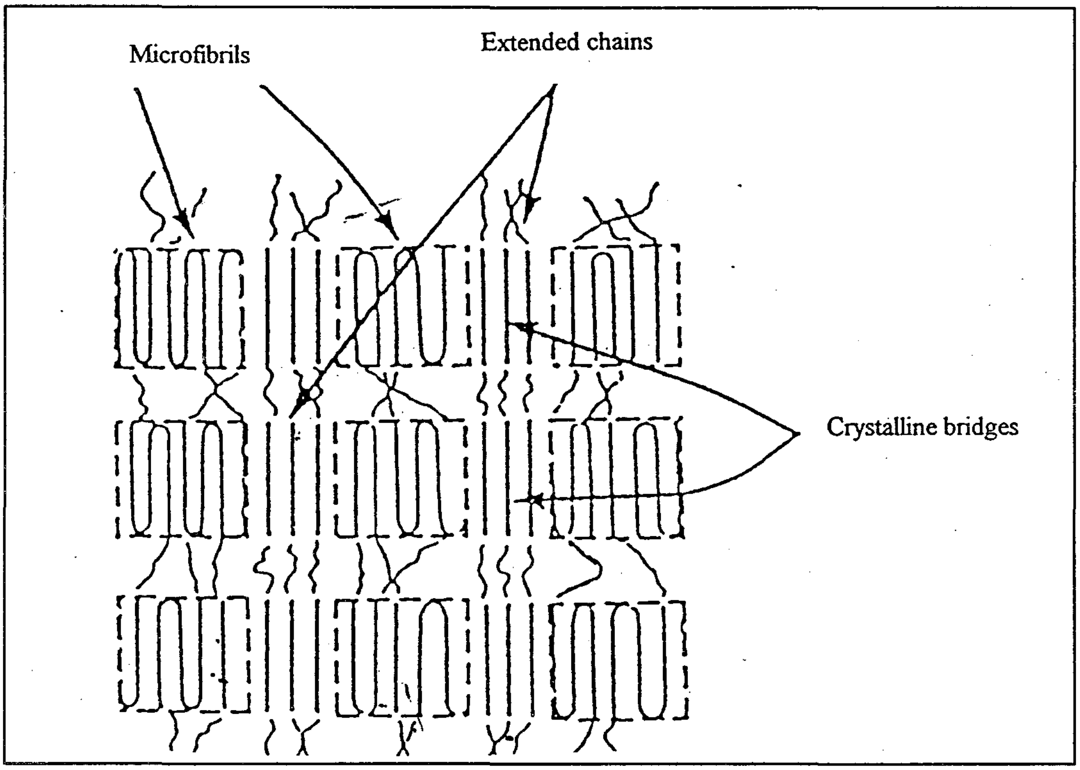


Figure 3.9 Microfibrillar structure with noncrystalline extended-chain molecules [56].

The microfibrils in turn are bound tight together into macrofibrils. The crystal blocks are connected by both intrafibrillar and interfibrillar tie molecules. Plastic deformation of the fiber causes unfolding of chains at the outer boundaries of the crystal blocks and results in an increase of the fraction of taut interfibrillar tie molecules. Since this process entails shearing displacement of adjacent microfibrils, it eventually effects full extension of the interfibrillar tie molecules, and is responsible for the high extensibility of nylon. A shematic presentation of the resulting structure is shown in Figure 3.9 [56].

The increase of the fraction of these extended molecules is proportional to the draw ratio. At high draw ratios, such extended molecules can form crystalline bridges between folded chain blocks, Figure 3.9 [56]

Tensile failure is indicated by rupture of taut tie molecule. Such rupture results in formation of microcracks that by growth can reach critical dimension and therefore cause catastrophic failure. Radial growth results in rupture of tie molecules bridging amorphous layers, whereas axial growth separates adjacent microfibrils and filament failure is then a consequence of rupture of tie molecules connecting microfibrils on opposite sides of the crack [56].

The nylon fibers are characterized by an initial modulus which is proportional to the ratio at which the fibers were drawn. This initial modulus is related to the mobility of the chain segments in the amorphous regions.

The stress-induced deformation (elongation) results in arrangement of chains in the axial direction. Further increase of strain results in the formation of both extended interfibrillar tie molecules due to shearing displacement of adjacent microfibrils, and new crystalline bridges in the amorphous layers. This may be affected by the length and extensibility of the intrafibrillar tie molecules [56].

3.4.2 Suture Structural Organization (Fiber Arrangement)

The physical structure of the silk suture is a braided one where each strand is twisted on the other leading to the formation of empty microspaces between multifilaments of a strand or more or less between fibers falling around the yarn cross-section thus, making the arrays formed by these fibers unsymmetric in the yarn cross-section. It follows that the concentric helices fibers form are not arranged all with the same pitch. So, upon loading, tension is not uniformily distributed, load is carried in proportion between fibers lying at nonsymmetric arrays along the same circumference, friction is reduced due to presence of microspaces, less amount of energy is absorbed by fibers due to the lack of its transfer among the fibers and to its dissipation through the microspaces.

Another consequence of this braided structure the "Crimp" formation, a characteristic of braid. A crimp interchange phenomenon occurs when braided silk fibers are elongated that accounts for different fiber extensions as a result of straightening of one set of yarns and increasing the bend of the other set [54]. This may affect the degree of deformations in the subsequent fibers of each yarn and the resulting breakage of the bonds between chains leading to the achieved crystallinity of the amorphous regions. This accounts for smaller elongation for silk than for nylon suture.

As for the nylon suture, the fibers are longitudinally aligned to form a continuous strand, twisted, fixing the fibers in an essentially helical geometry in the yarn structure and thus providing for force transfer from fiber to fiber through friction. As silk suture is an assemblage of braided silk strands or yarns, the nylon suture turns out to be an assemblage of twisted yarns, which behavior is more or less easier to be analyzed (see Figure 3.10 a,b).

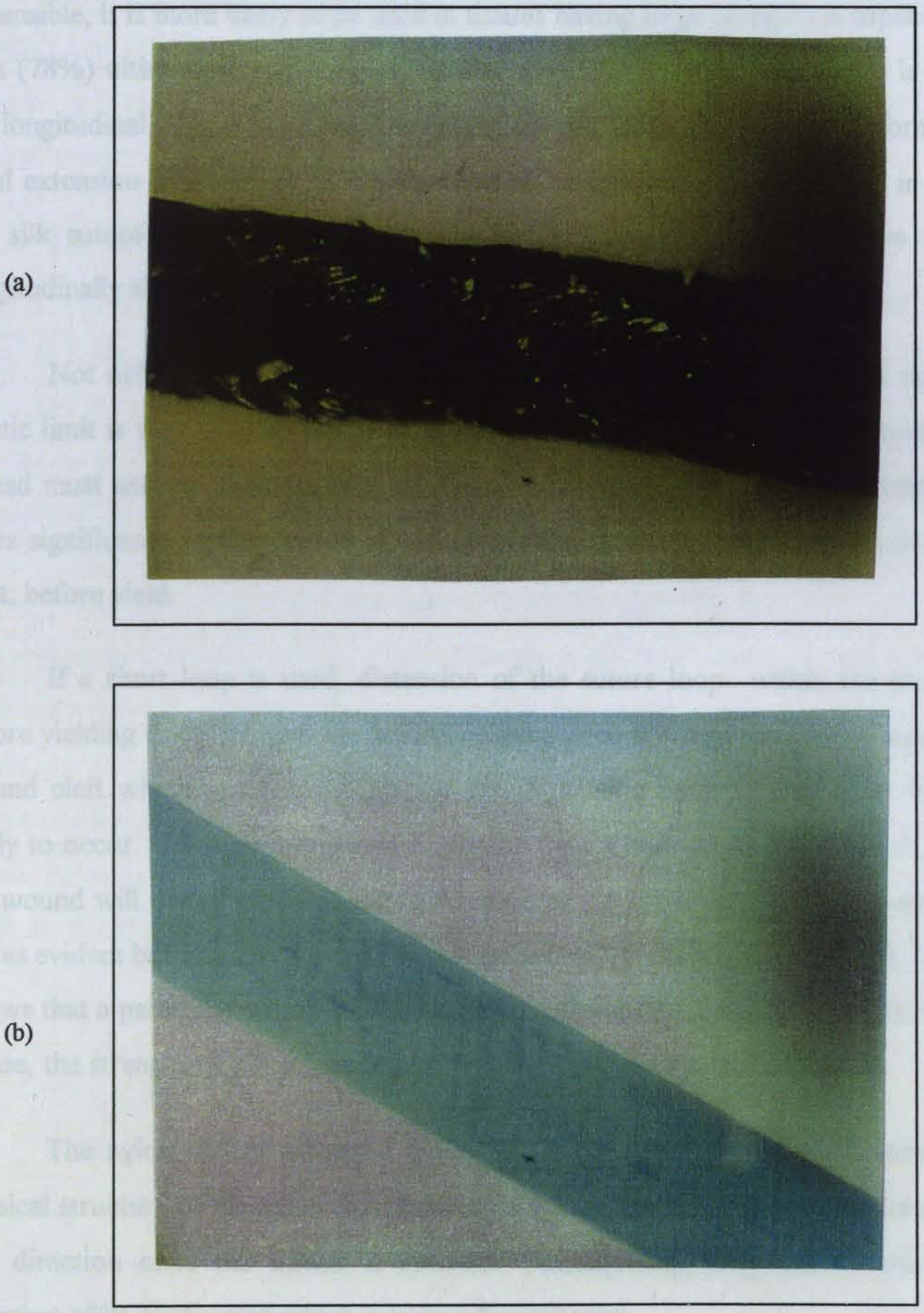


Figure 3.10 Photographs at 67.5x magnification of: (a) braided multifilament silk suture (b) monofilament nylon suture .

When closing wounds with large loops, the use of a stiff suture is required, while with small loops and when ligating small blood vessels, use of flexible and easy bending suture is required. Here, arises the significance of a property which has, as well, an outcome on surgical applications, the percentage elongation or strain Since nylon suture is more distensible, it is more likely to be used in tissues having large elongation capabilities such as skin (78%) ultimate elongation) and cardiac muscle (63.8 %). This ability is attributed to the longitudinal alignment of the fibers along the fiber axis in a way that deformation causes axial extension of the fibers in the direction of its application. While fibers in a braid as in the silk suture are loaded in proportion and at different directions since they are not longitudinally aligned across the suture long axis.

Not only ultimate elongation has a relevance but also the ability to elongate in the elastic limit is well as important. For, when the swelling of the wound recesses, the suture thread must assume again its original dimensions and approximate the wound edges. This gives significance to the degree of elongation the suture material undergoes in the elastic limit, before yield.

If a short loop is used, distension of the suture loop within the elastic limit i.e. before yielding does not limit i.e. before yielding, does not result in a wide separation of the wound cleft which is advantageous for slowly healing wounds and when hemorrhage is likely to occur. Consequently, when a surgeon uses a fiber which is too weak, a rupture of the wound will occur; when he uses a fiber which distends too much the consequences are not as evident but one must assume that it influences the wound healing [16]. If the surgeon knows that a particular suture material is easily distended he may use a thicker fiber. In such a case, the strength of a fiber is not used but its resistance against distension.

The nylon suture withstand higher loads than silk. This may be attributed to the physical structure of the nylon suture. Being a monofilament suture, all fibers are aligned in one direction once the thread is stressed. Consequently, they sustain the load in the direction of its application which is quite advantageous.

Because silk suture has a braided structure as shown in Figure 3.10, not all or most of the fibers are stressed as in the nylon suture, the fact that explains lower maximum load. As fibers are braided to form yarns, they are differently oriented; so, only the ones aligned in the direction of the load applied will loaded. Thus, probably, the number of loaded fibers in a nylon suture exceeds that in a braided silk suture.

Finally it is worth noting the effect of strain rate. At low strain rate, the material has more time to undergo more elongation until rupture therefore, absorbs energy that will be dissipated at failure when fibers rupture in a serial mechanism. At high strain rates, the material is strained rapidly in such a way failure occurs in a sudden when all fibers rupture altogether simultaneously.

The experimental values did not differ widely from the literature ones. There were uncontrolled experimental errors like holding the specimen suture tighter or lighter in the jaws, as it has an effect on how much fibers are grasped and stressed under tension, the more are held, the higher is the recorded stress or strain values. Local failure does contribute in biasing the experimental results in such a way that whether it occurs at midpoint of the suture length or somewhere upper or lower or even at the grip site.

As a conclusion, knowing such mechanical parameters can be of utmost validity to the surgical applications. On the behalf of the discrimination between suture materials, when knowing a special property of such materials, the surgeon can correctly vary his suture technique. Suture materials having good elongation capability or wide range of elongation can be tied tightly, so that even under continuous loading, when distension occurs, neither plastic deformation (yielding of the material) nor loosening of the knot (the material becomes slack) results.

4. MECHANICAL PROPERTIES OF ALTERNATING SLIDING KNOTS

4.1 Introduction

In the daily routine of the operating theater, the prevalence of the sliding knot in actual use is high. The reason is that when surgeons fail to cross their hands in tying the flat knot, keeping tension on one strand to prevent the first throw from loosening, they unconsciously make a sliding knot. In spite of this, systematic studies on the physical qualities of the various sliding knots are lacking. Besides the fact that they have not been more closely examined, the few studies have some short comings, one of them is they were performed using suture materials in which the type of knot only minimally affects relative knot security [22].

Comparison of the different suture materials revealed major differences in knot holding ability. Trimbos and associates [17], [18], studied the knot strength of different sliding knots (the identical, nonidentical and parallel). In their study, they indicated that knot strength is dependent on both the type of the knot and the type of suture material. As far as the sliding knots are concerned, there is the type of knot where the strand alternates using a pattern other than with every throw[28].

In addition, the studies performed used different synthetic absorbable suture materials like polyglycolic acid, polydioxanone, polyglactin and polyglyconate. Unfortunately, it is questionable how well the findings can be expected to apply to other suture materials [28]. This chapter presents the results of an experimental study of the mechanical performance of various sliding knots in comparison between silk sutures and nylon sutures.

4.2 Materials and Methods

Silk and nylon sutures of 2/0 USP size are used. Loops of the suture material were tied around two cylindrical rods attached to a board. The knots were tied under closed observation and the type of the knot was snugged tight. The diameter of the rod was 1 cm and the distance between them was 16 cm [17]. The knots were tied by the same person to minimize variability and snugged down by moderate force. The loops, then were mounted on the Zwick 1446 universal testing machine, using special clamps to hold the loop at both sides so that the knot will be placed halfway between them. The loop method is followed as it is more convenient, as shown in Figure 4.1 [28]. The loops are pulled at a rate of 50 mm/min.

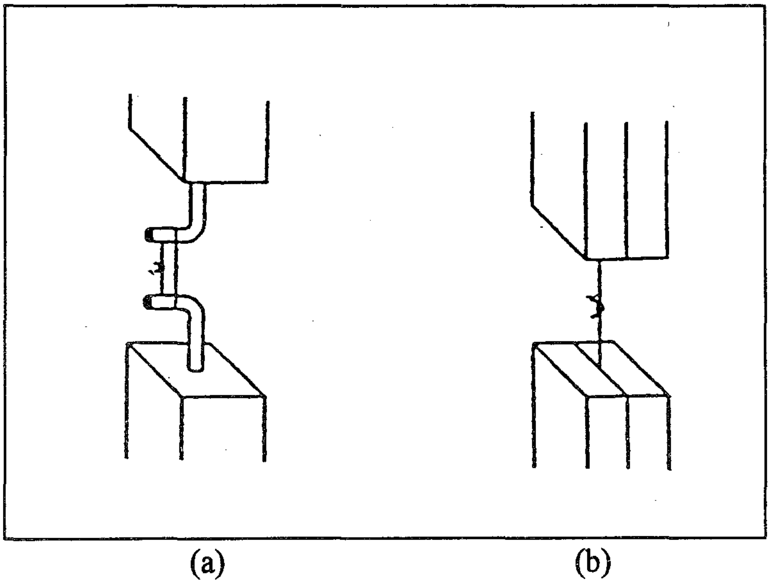


Figure 4.1 Tensile testing of sutures by: (a) Loop method (b) Single strand method.

To identify the knots, a unified nomenclature is used in the description of the knots. Using this nomenclature, the "=" sign means that the subsequent throw turns in the same direction as the one preceding it. The "x" means that the subsequent throw turns in the opposite direction from the one preceding it. Numerals are used to designate the number of turns in each throw. The "S" is used to show that the knot is a sliding one. For sliding knots in which the axial strand alternates, the "//" symbol is used to show the change when the following throw turns in the same direction. A "# " sign is used when the axial strand changes and the following throw turns in the opposite direction. This nomenclature is illustrated in Figure (4.2 a: as taken from literature [28]; b: as visualized under the stereoscope)

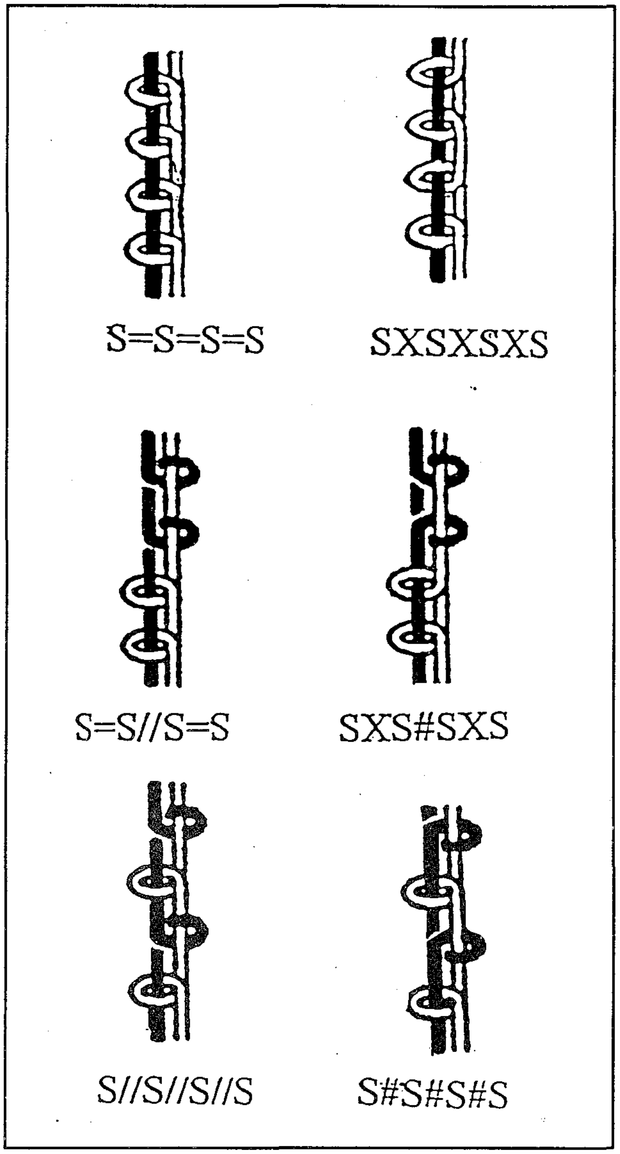
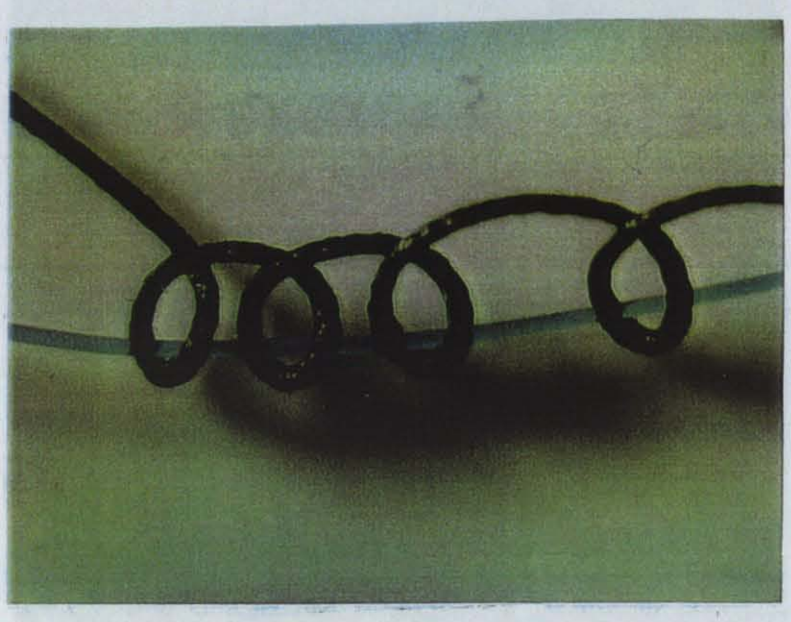


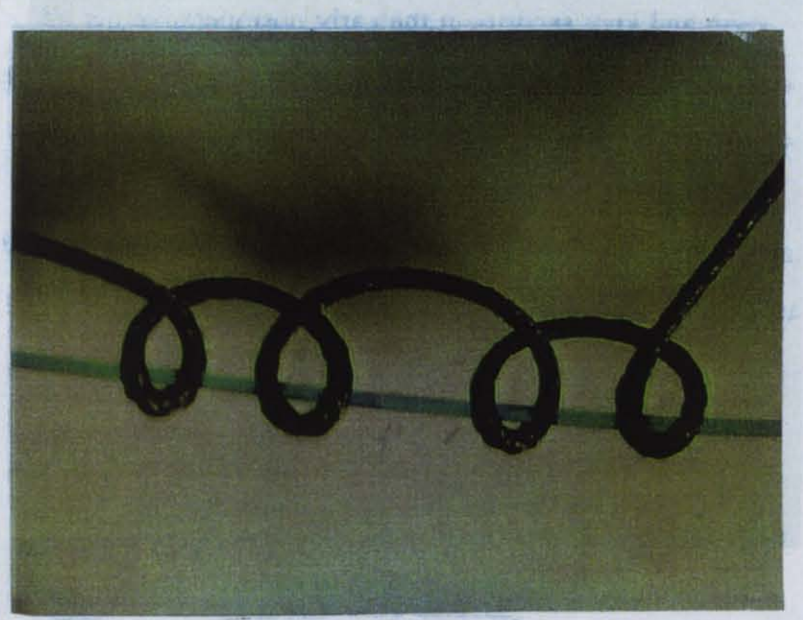
Figure 4.2 a Structure and codes of the tested knots [28].

Four different sliding knots are tested for both silk and nylon sutures. Around fifteen experiments were run for every knot of each suture of the gauge size 2/0 USP.

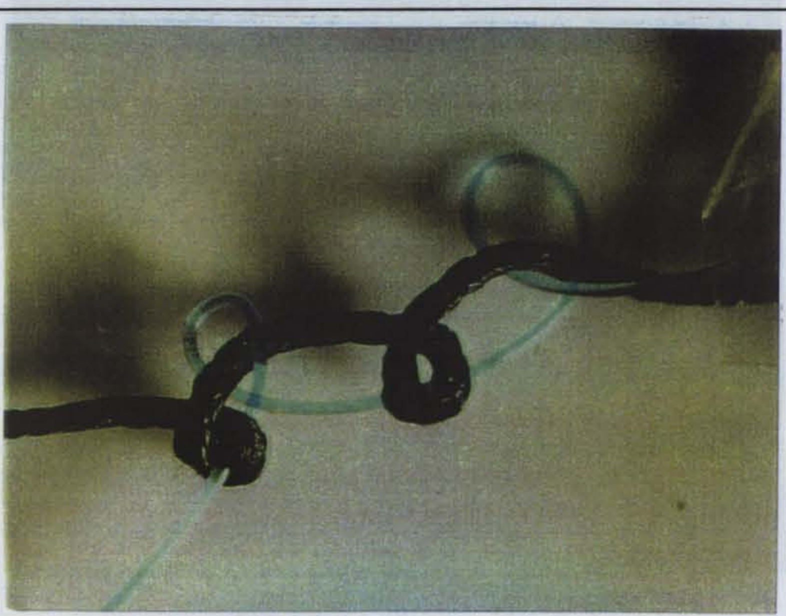
The loop holding capacity was defined as the maximum force calculated from the load-elongation curve required to break the tied suture loop, by loading the part of the suture that forms the loop. Knot failure is defined as the break of the knot or its slippage exceeding two mm. used as standard [17]. The knot holding capacity is half the value of the loop holding capacity and may be used to compare the loop holding capacity with the breaking strength of the unknotted thread. Separate experiments were run to determine the single thread's tensile strength of the two sutures. Photographs of the tested knots, obtained using a stereoscope are presented below in Figure 4.2b.



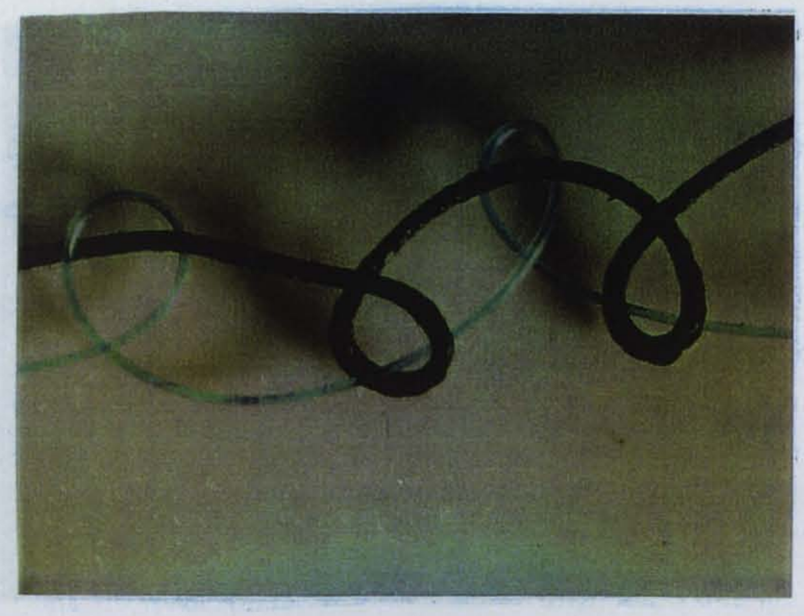
S=S=S=S



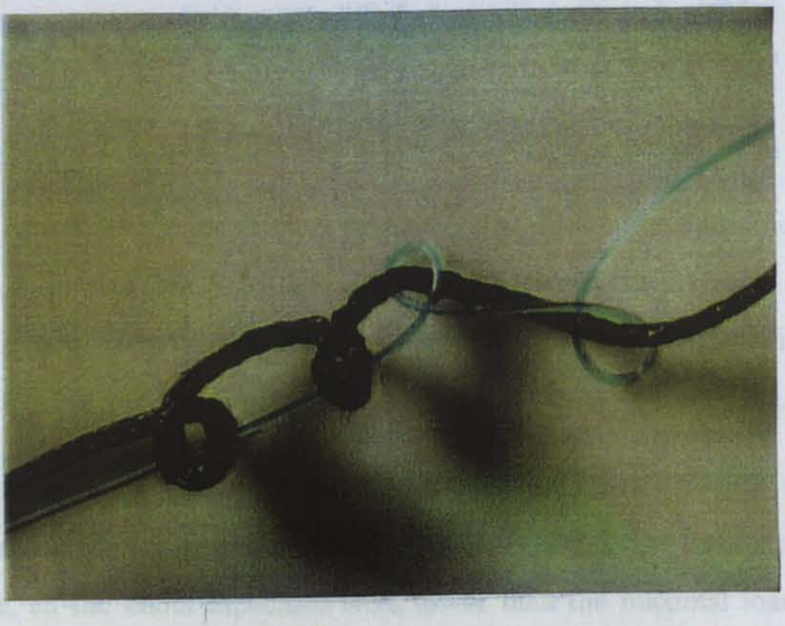
SXSXSXS



S//S//S//S



S#S#S#S



S=S//S=S

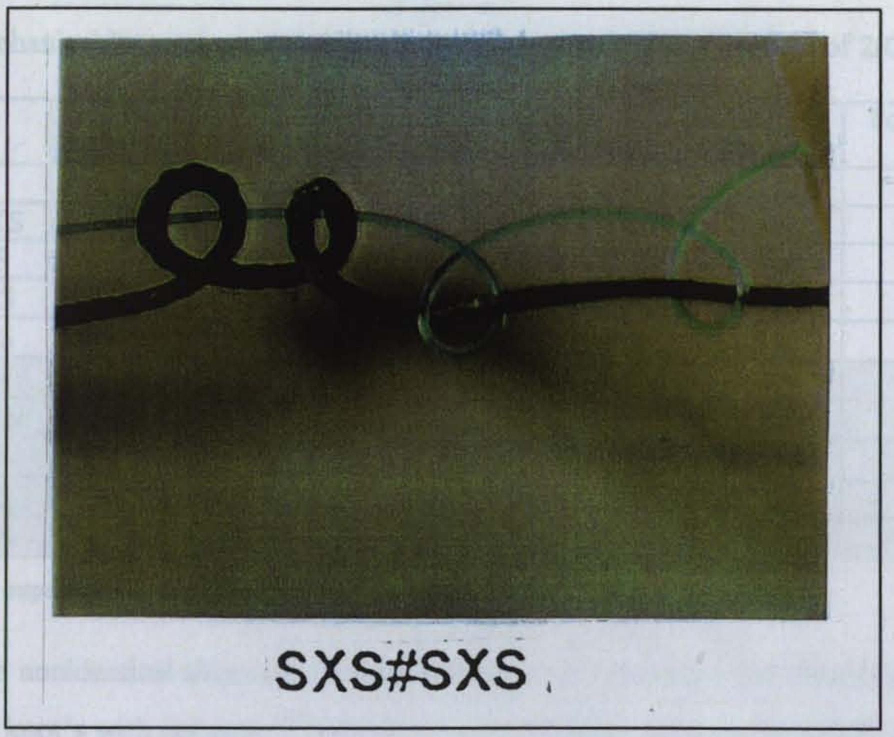


Figure 4.2 b: The tested knots as visualized under the stereoscope (x 67.5)

4.3 Results

Table 4.1 presents the mechanical parameters of different sliding knots of silk and nylon sutures of USP size 2/0. Statistical differences between results were determined by one way ANOVA test followed by multiple range test: Tukey's-HSD test with significance level 0.05.

Differences in knot holding capacities between the knots of both nylon and silk suture were found to vary independently. Since knotting decreases the tensile strength of the single suture, all the knots capacities were lower than the maximal loads of both the single nylon and single silk sutures.

For every material, the differences were various; i.e. for nylon suture , the parallel alternating sliding knot holding capacity (S//S//S//S) was found to be significantly stronger than the nonidentical alternating sliding (S#S#S#S), as shown in Table 4.1.

Table 4.1
Mechanical Properties of Knotted Loops of Silk and Nylon Sutures of 2/0 Size

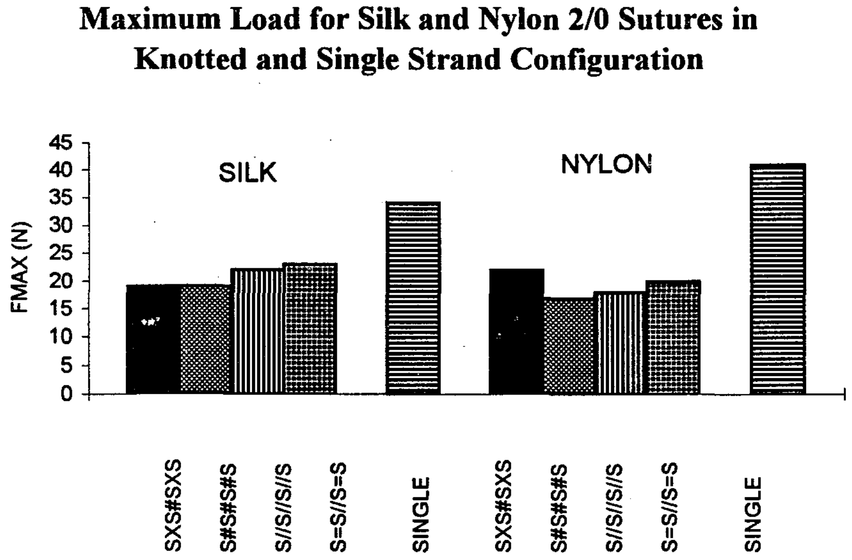
	KHC N	FYIELD N	ELMAX %	ELYIELD %	Toughness N. mm
Silk					
SXS#SXS	19±1	9.7±0.7	15±2.7	2.5±0.2	304±53
S#S#S#S	19±1	11±2	14±1.7	2.3±0.4	284±33
S//S//S//S	22±2	10±1	16±3	2.2±0.6	377±116
S=S//S=S	23±1	12±1	19±2	3.4±0.7	471±70
Nylon					
SXS#SXS	22±3	17±3	30±4	10±2	725 ±208
S#S#S#S	17±6	15±2	18±6	9±0.9	370±245
S//S//S//S	18±3	15±3	20±5	9±1	383±157
S=S//S=S	20±6	16±2	30±8	12±2	694±343

Figures are expressed as average value with standard deviation.

The nonidentical alternating knot's with different patterns (SXS#SXS) and parallel alternating knot's with patterns (S=S//S=S) had higher KHC than the (S#S#S#S) knot.

Comparing the knots' capacities between both nylon and silk suture, it shows up that the parallel alternating with different patterns (S=S//S=S) of nylon is stronger than the nonidentical alternating knot (S#S#S#S) of silk suture. Both the parallel alternating (S//S//S//S) and parallel alternating with different patterns (S=S//S=S) of silk suture have significantly higher KHCs than that of the nonidentical alternating (S#S#S#S)of nylon suture, with no significance in the differences between the other knots of both sutures.

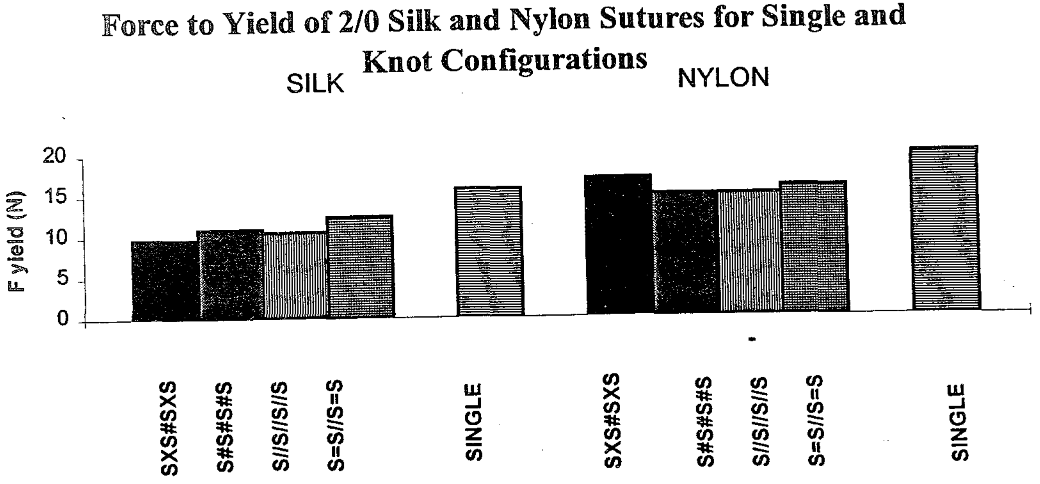
The silk suture's knots varied in knot holding capacity among themselves, the parallel alternating knot (S//S//S//S) and the parallel alternating with different patterns (S=S//S=S) have significantly higher KHC's than the nonidentical alternating and nonidentical alternating with different patterns knots (S#S#S#S and SXS#SXS) of the corresponding suture, as shown in Figure 4.3a.



Maximum Load for Silk and Nylon 2/0 Sutures in Knotted and Single Strand Configuration

Figure 4.3a Knot holding capacity or maximal load of sliding knots of silk and nylon sutures 2/0.

The force to the yield point did not differ significantly between the knot configurations of each suture. All the knots of nylon suture had similar values, the silk suture as well. There was statistically a significant difference between the two materials, with the nylon suture's values being higher. The loads at yield differed also from those obtained for the single strand of both sutures, as shown in Figure 4.3b.



Force to Yield of 2/0 Silk and Nylon Sutures for Single and Knot Configurations

Figure 4.3,b Force to the yield point for the tested knots of silk and nylon sutures

As for the elongation to maximum, a significant difference was found between the knots of both materials with nylon knots undergoing higher elongations for the following knots: the alternating sliding knots of different patterns. No significance was found comparing the other knots. However, only the nonidentical alternating knot of silk had less elongation when compared to the alternating knots of nylon, as shown in Figure 4.3c.The percentage elongation to maximum for the knots of silk was less than that of the single suture. The same value for the knots of nylon was even less than half the value of the single suture.

Percentage Elongation to Maximum for 2/0 Silk and Nylon Sutures for Single and Knot Configurations

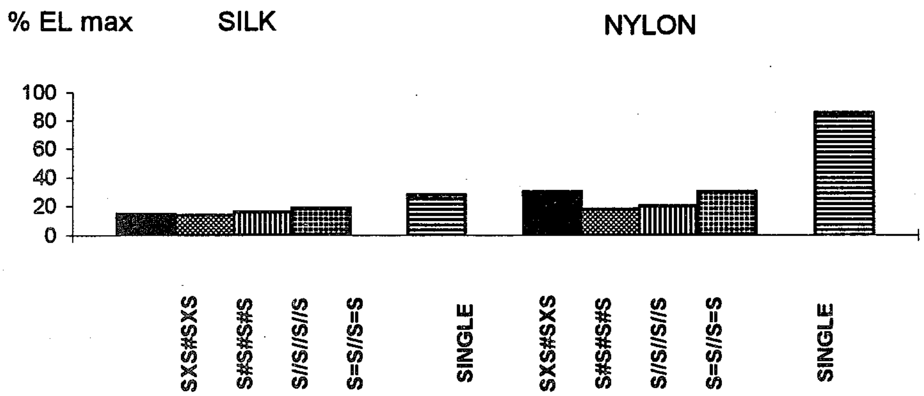


Figure 4.3c The percentage elongation to maximum of the sliding knots of both silk and nylon sutures.

Comparing the elongation to the yield point, for nylon suture, the parallel alternating knot with different patterns (S=S//S=S) showed a significantly higher value than that of both the parallel alternating knot (S//S//S//S) and the nonidentical alternating knot (S#S#S#S), with all values belonging to knots of nylon being significantly higher than those belonging to knots of silk suture. The single strand of silk had similar values to the knots values, but the nylon thread had relatively a higher value than that of the knots, as shown in Figure 4.3d. The percentage elongation to the yield point for the knots of nylon was nearly about half that of the single suture.

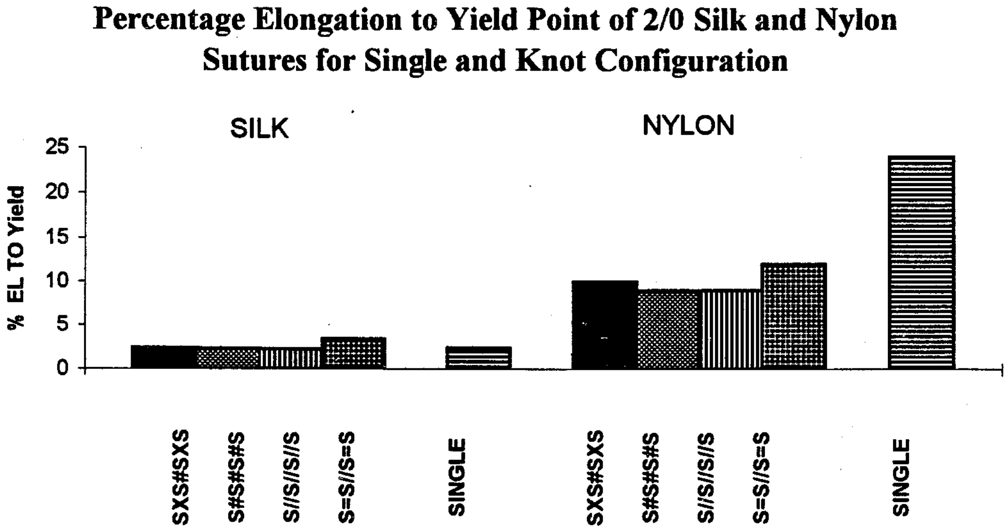


Figure 4.3d Percent elongation till yield for sliding knots of silk and nylon sutures.

The toughness values did differ among the knots of the silk suture, but it differed among the nylon knots. The alternating sliding with different patterns recorded higher values than the others. When compared together, the toughness values of the single thread of silk and nylon were higher than those of the respective knot materials, as illustrated in Figure 4.3e.

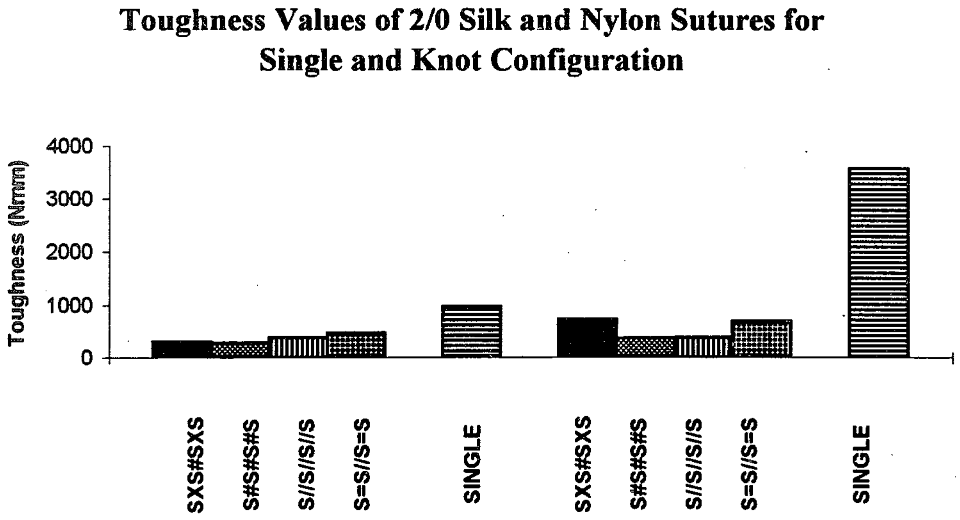


Figure 4.3 e Toughness values for sliding knots of 2/0 silk and nylon sutures.

4.4 Discussion

Although the importance of physical characteristics of surgical suture knots has been recognized, few studies on these aspects have been published. Comparison of the results between investigators is hampered by the fact that experimental design has not been standardized [18]. In this chapter, knotted suture loops were tested. An alternate method is to cut the loop and attach the loop to a tension bench. The loop method was used because it offers some advantages: 1) it eliminates the difficulty of fastening the suture thread in the machine, 2) it excludes any risk of slippage at the points of attachment, and 3) it mimics the in vivo conditions more closely [17].

Although the properties of a variety of different knots have been reported by several authors, a systematic analysis of sliding knots has not been reported, in the English literature, in spite of recent evidence that sliding knots are more commonly used by gynecologic surgeons [18].

In this chapter, it was found that the knot holding capacities did not vary much between the two sutures. Even, the sliding knots of different patterns holding capacity did not differ much from that of other alternating sliding knots. In nylon, both the alternating knots with different patterns (SXS#SXS and S=S//S=S) had similar knot capacities while in silk, the parallel alternating with different patterns (S=S//S=S) has higher capacity than its nonidentical one (SXS#SXS). The force to the yield point was the same for all knots as stated in the previous section, which is evident since the knots are all of the same material.

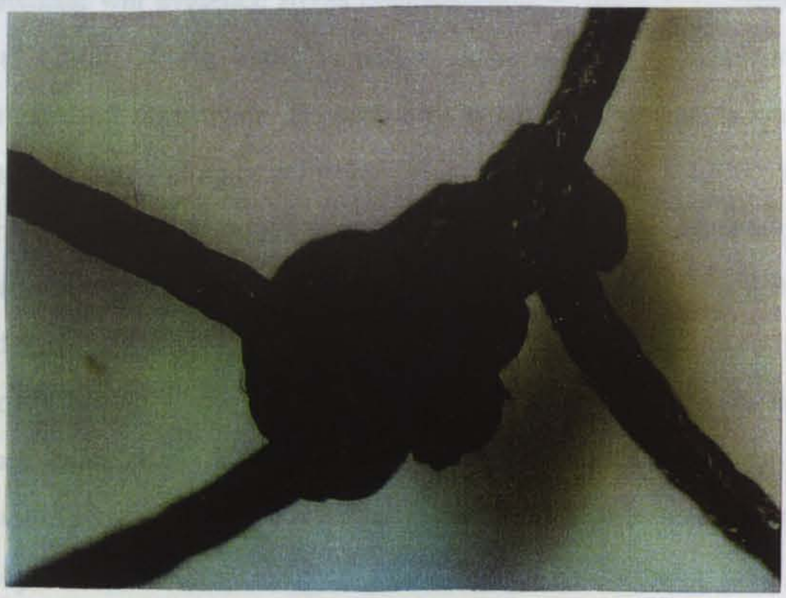
The difference in the results between the two sutures is related to the effect of the material composing the suture, the structural composition and fiber arrangement (mono- versus multifilament) of the two sutures. This was explained in more details in the previous chapter.

In this chapter, the braided silk suture demonstrated less compliance i.e. greater modulus of elasticity than the monofilament nylon suture. However, the monofilament nylon suture was more extensilble (greater strain) than the braided silk suture. Our data agreed well with those of Greenwald [26]. He, too, showed that suture modulus of elasticity is determined by suture design as well as suture type.

The importance of the yield point for suture knots lies in the fact that after this point there is a permanent deformation of the suture material, which leads to the distortion of the suture loop, giving the way for the wound edges to separate progressively until rupture, resulting in dehiscence of the wound. Therefore, knowing the corresponding load and elongation at that point, ensures the surgeon that stressing the suture loop to this limit hence called the elastic limit the suture loop won't be deformed and consequently, the wound is safely closed with the proper tying tension. Evidently, since the knot's capacity was lower than that of the suture single thread, the corresponding parameters to the yield point would rather be lower too. Therefore, knotting not only decreases the suture tensile strength but also, the yield strength.

Knot properties should be considered among other properties (Distensibility, surface friction, flexibility etc...) that are of great interest to the surgeon. In the United States Pharmacopoeia (USP) the strength and cross-diameter are the only physical data that are used to specify surgical materials [12]. Figure 4.4 shows sliding knots of a: Silk and b: Nylon observed and photographed under the stereoscope.

(a)



(b)

Figure 4.4 Sliding knots of: (a)silk, (b) nylon as observed under stereoscope (x 67.5)

Any material that is strained to permanent deformation, whether by creasing, crushing or stretching will evidence stress concentration at the site of damage, leading to reduced loads at failure. Tying knots in suture materials imposes such strains, and the results can be measured objectively. Beyond the strains imposed when tying knots, knot technique and suture type influence other measurable mechanical properties [26]. Besides, when enough force is applied to a tied suture to result in breakage, the site of disruption of the suture is always the knot [14]. This is illustrated in Figure 4.5. This force necessary to break a knotted suture is lower than that required to break an untied suture. The tensile forces exerted on a tied suture are converted into shear forces by the configuration of the knot and rupture the suture. The magnitude of the forces necessary to produce knot breakage vary with the size of the suture loop, the type of suture material and the diameter of the suture [14]. The tissue in which the suture is implanted may also have an influence on the knot strength of sutures.

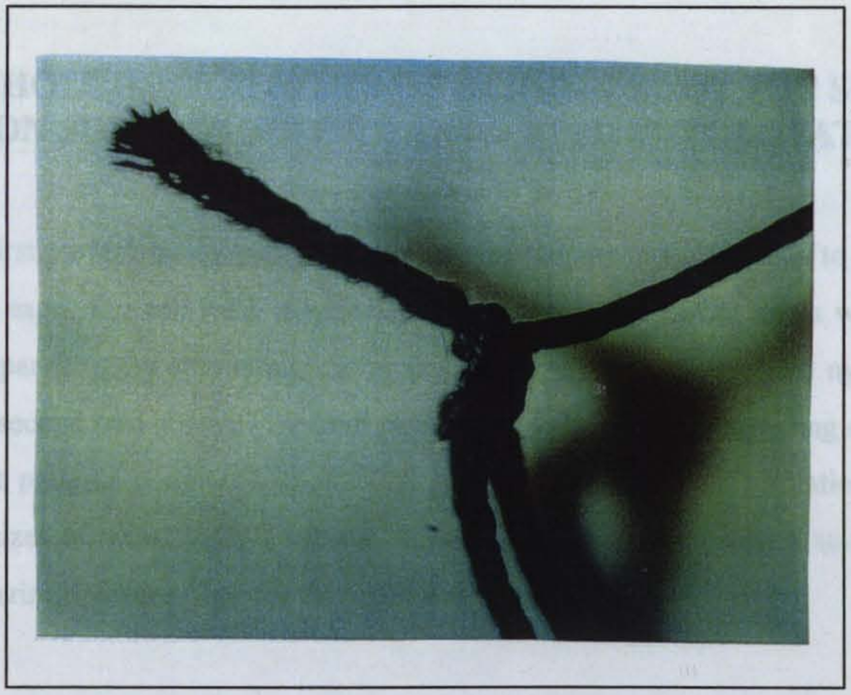


Figure 4.5 A photograph of a silk knot ruptured at the site of or near the knot as taken under stereoscope (x 67.5)

Furthermore, knot properties should be taken into consideration because in clinical practice, the knots are used. One cause of their failure is untying of the knot resulting from its slippage. Actually, knot slippage is counteracted by frictional forces of the knot [14]. The degree to which knots slip can be influenced by a variety of factors, including the coefficient of friction of the suture material, suture diameter, type of knot, the length of the cut ears of the knot and moisture [14].

Accordingly, in this chapter we focused on the knot under <u>in vitro conditions</u> to proceed with testings under <u>in vivo conditions</u> in the following chapter.

5. KNOT HOLDING CAPACITY OF SLIDING KNOTS OF SILK AND NYLON SUTURES BEFORE AND AFTER IMPLANTATION

The first part of this chapter presents experiments that were designed to evaluate the knot holding capacities and knot efficiencies of the sliding alternating knots with different patterns compared to the alternating sliding in the dry state for 2/0 and 4/0 nylon and silk sutures. The second part presents the knot holding capacities of the alternating sliding knots with different patterns compared to alternating sliding knots after implantation, again for 2/0 and 4/0 sizes of nylon and silk sutures. It also, adds the tissue reaction to these knots. Separate experiments were done for this purpose.

5.1 Knot Holding Capacity of Sliding Knots Under In Vitro Conditions

5.1.1 Introduction

Comparison of the different suture materials revealed major differences in knot holding ability. Trimbos and associates [18], studied the knot strength of different sliding knots. In their study, they indicated that knot strength is dependent on both the type of the knot and the type of suture material. As far as the sliding knots are concerned, there is the type of knot where the strand alternates using a pattern other than with every throw. In this chapter, this type i.e. the alternating sliding knots with different patterns will be compared to the alternating and the simple sliding knots. It is questionable then, how well the performance of all these knots will vary among themselves, in relation to the suture material and the suture size. In this chapter, also, we wanted to test the following hypothesis : does the knots performance depend on the degree of their complexity? The alternating knots with different patterns were considered the most complex, followed by the alternating and the simple sliding knots.

5.1.2 Materials and Methods

As described in chapter IV, the loops of the suture material were tied around two cylindrical rods attached to a board. The knots were tied under closed observation and the type of the knot was snugged tight. The diameter of the rod was 1 cm and the distance between them was 16 cm [17]. The knots were tied and snugged down by moderate force. The loops, then were mounted on the Zwick 1446 universal testing machine, using special clamps to hold the loop at both sides so that the knot will be placed halfway between them. Again, the loop method is followed. The loops were pulled at an extension rate of 50 mm/min. Six different alternating sliding knots are tested for both silk and nylon sutures. Around fifteen experiments were run for every knot of each suture of both gauge sizes 2/0 and 4/0 USP The codes and configurations of the tested knots are given in Figure 5.1 [28]. They were illustrated previoulsy in Figures 4.2 a and b.

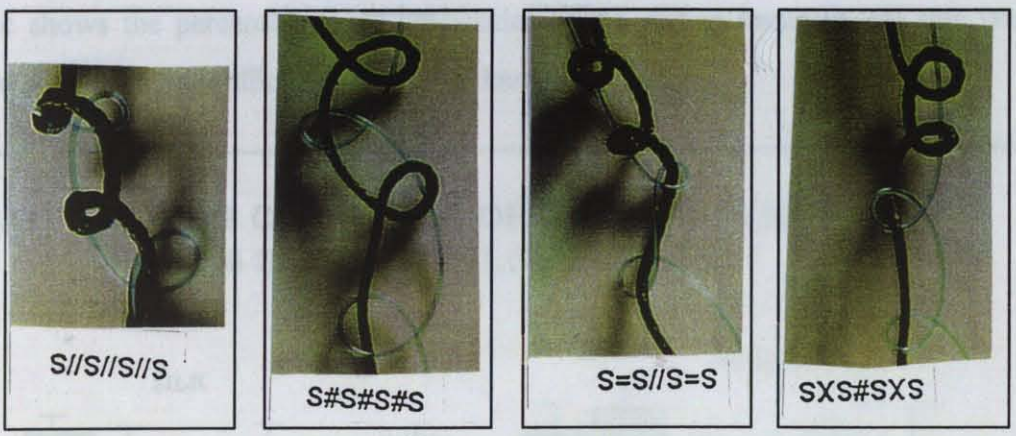


Figure 5.1 Codes and configurations of sliding knots (x 67.5) under the stereoscope.

The loop holding capacity was defined as the maximum force calculated from the load-elongation curve required to break the tied suture loop, by loading the part of the suture that forms the loop [17]. Knot failure is defined as the break of the knot or its slippage exceeding two mm. used as standard. The knot holding capacity is the force that a specified suture can sustain without failing either through the suture breaking or the knot slipping. The formula for determining the KHC varies according to the testing method, KHC equals one-half the force required for rupture i.e. the loop holding capacity. For the single strand method, KHC equals the force required for rupture [28].

Knot efficiency KE, termed also relative knot security is defined as the KHC expressed as a percentage of the tensile strength of the single strand of the suture. This value has the benefit of not being affected by the testing method or by the size or strength of the suture being tested [28]. Secure knots were defined as knots that failed by breaking in at least nine of ten cases. Knots that did not meet this criterion were defined as insecure knots. The mean knot holding capacities were compared by one-way analysis of variance followed by multiple range test: Tukey's-HSD test with significance level 0.05 [57].

5.1.3 Results

The results of the experiments are expressed as knot holding capacities with standard deviations and knot efficiencies. Figure 5.2a shows the knot holding capacities of the tested sliding knots for silk and nylon sutures of 2/0 USP size. Figure 5.2b shows the knot holding capacities of the six sliding knots of silk and nylon sutures of 4/0 U.S.P size. Figure 5.2c shows the percentage knot efficiencies of the sliding knots of 2/0 size while Figure 5.2d shows the knot efficiencies of these knots of 4/0 size.

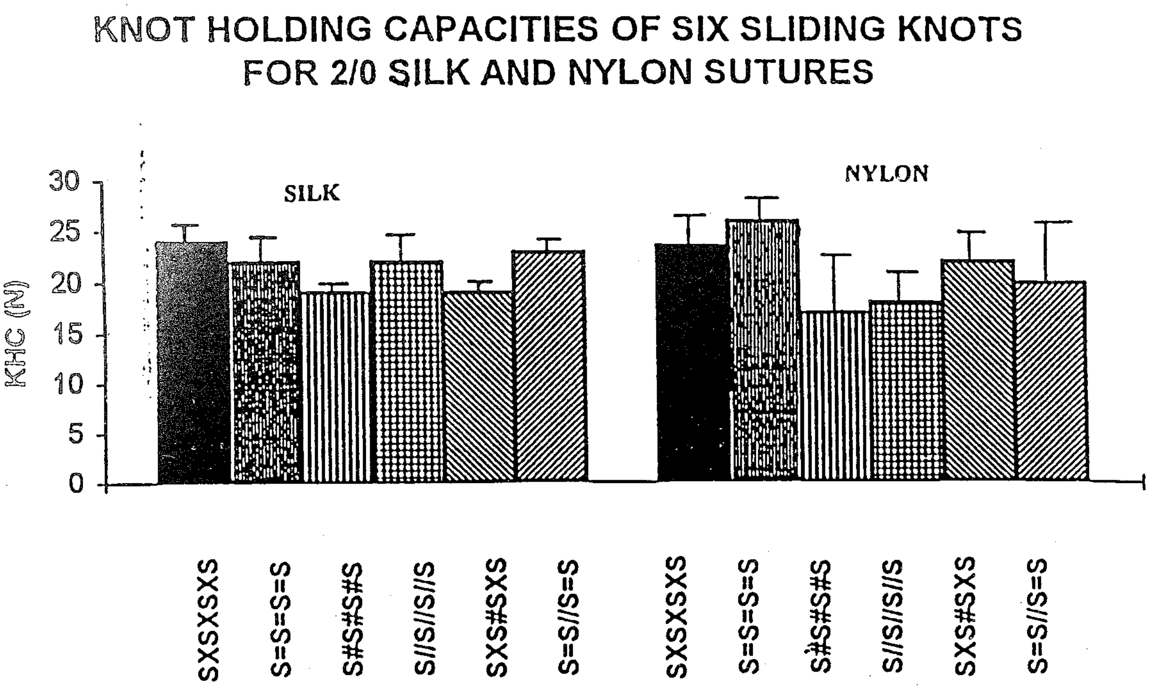


Figure 5.2 a Knot holding capacities of silk and nylon sliding knots 2/0.

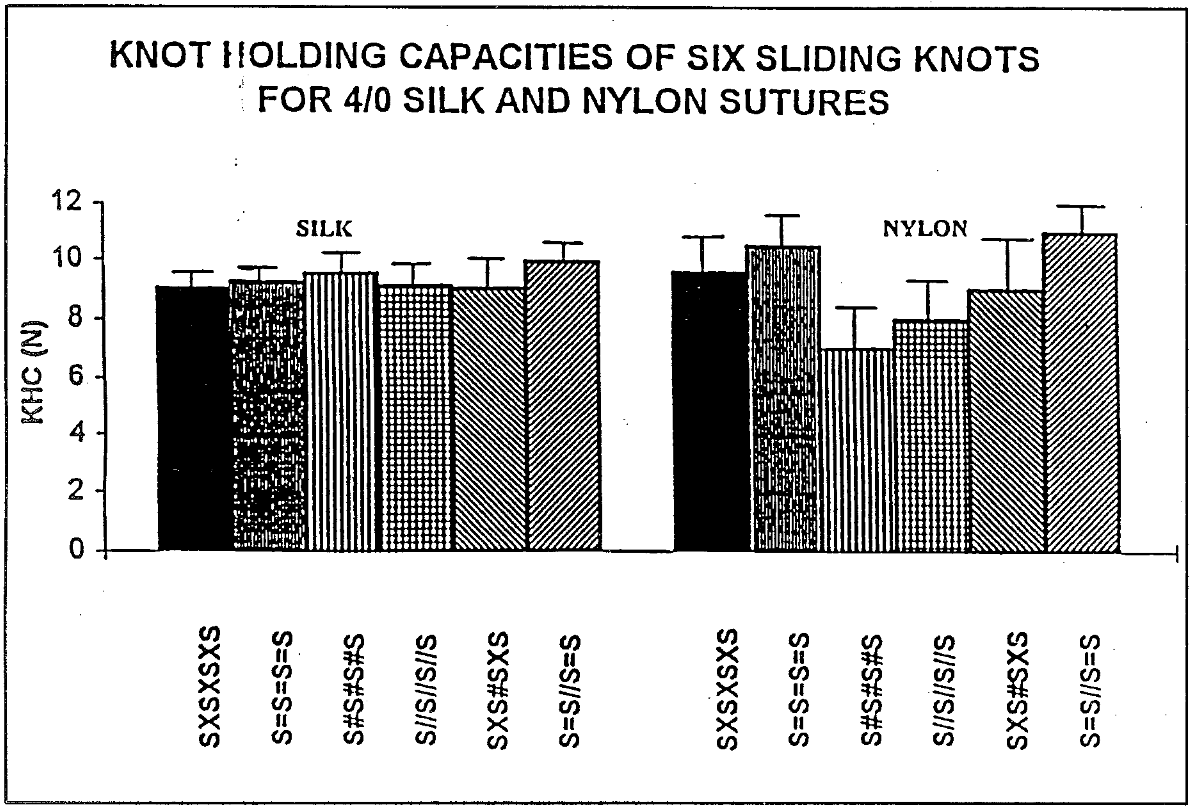


Figure 5.2 b Knot holding capacities of silk and nylon sliding knots 4/0.

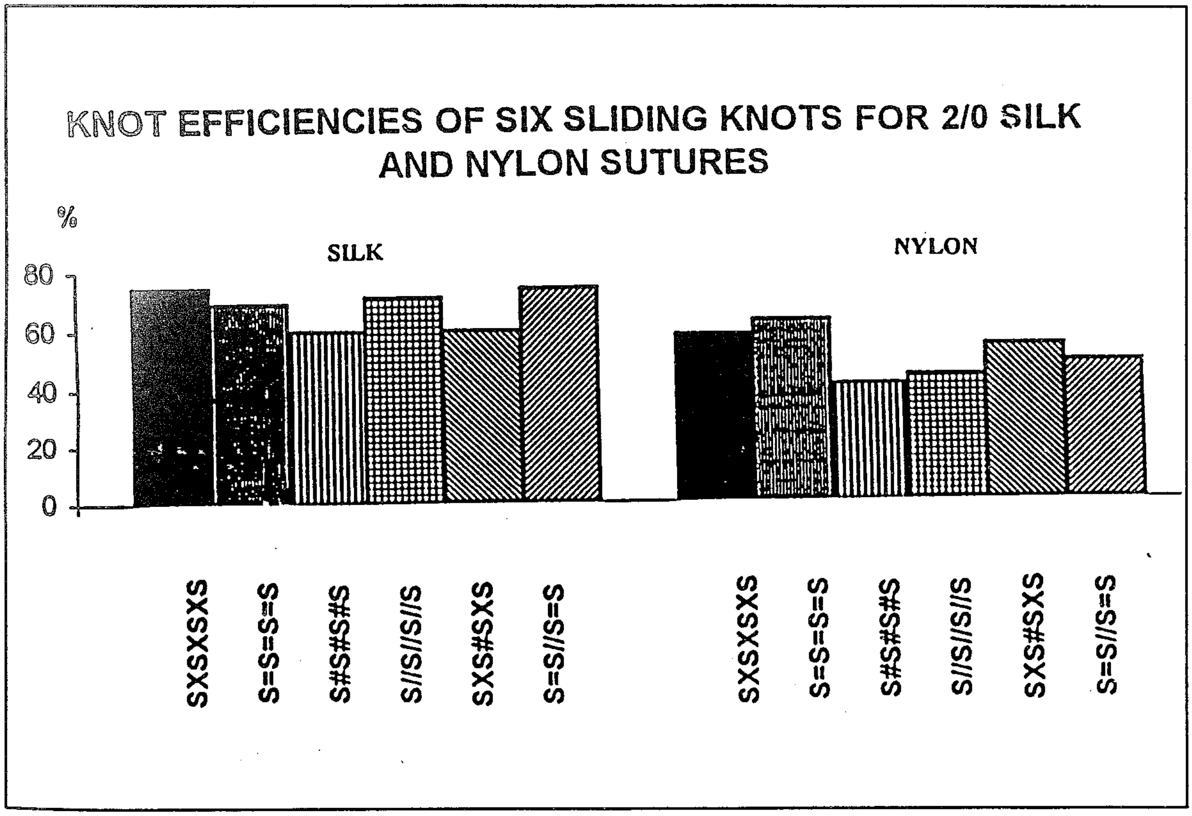


Figure 5.2 c Percentage knot efficiency of silk and nylon knots 2/0.

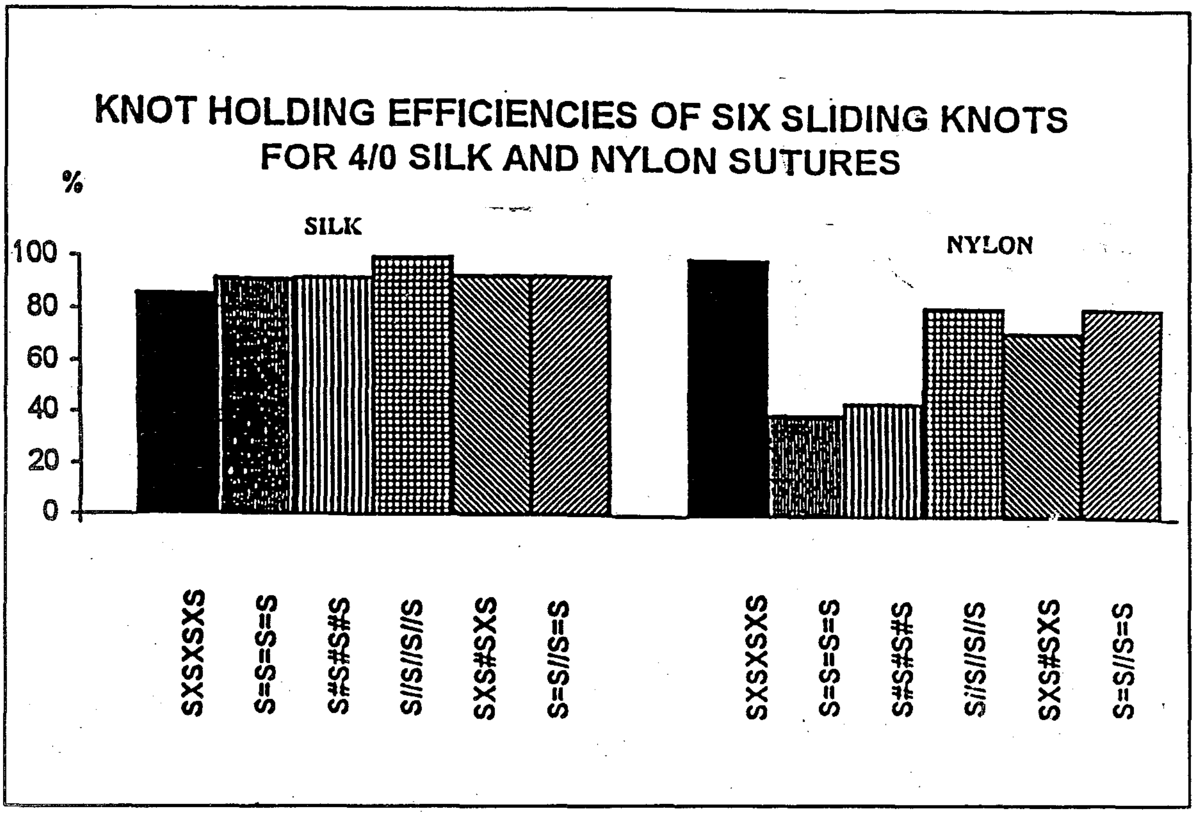


Figure 5.2 d Percentage knot efficiency of silk and nylon knots 4/0.

5.1.3.1 Effect of Suture Material

Both silk and nylon showed similar KHC for the simple sliding knots 2/0 and 4/0. Only, the parallel knot (S=S=S=S) of 2/0 size was superior in KHC with nylon, as shown in Figures 5.2a,b. The difference is statistically significant and shown in Table 5.1.

Table 5.1
Statistical analysis of differences in knot holding capacities of the sliding knots between the silk and nylon sutures of both sizes.

Knot Code	2/0	4/0
SXSXSXS	NS	NS
S=S=S=S	P<0.05	NS
S#S#S#S	NS	P<0.05
S//S//S//S	P<0.05	NS
SXS#SXS	P<0.05	NS
S=S//S=S	P<0.05	P<0.05

NS=not significant
Statistical differences are determined by one way ANOVA test: Tukey-HSD test with significance level 0,05.

For the alternating knots, the parallel alternating knot (S//S//S//S) of size 2/0 was superior in KHC using the silk suture. Whereas, for size 4/0, the nonidentical alternating knot (S#S#S#S) was superior with the silk suture. This is illustrated in Figures 5.2a,b and Table 5.1

As far as the alternating knots with different patterns and referring to the same Figures and Table, the nonidentical knot (SXS#SXS) showed higher KHC using nylon 2/0. Whereas, the parallel knot (S=S//S=S) showed higher KHC with silk 2/0 over nylon 2/0 and with nylon 4/0 over silk 4/0.

5.1.3.2 Effect of Suture Size

As for every material, the knot profiles varied differently in relation to the suture size. Using the silk suture, of 2/0 size, the alternating knots with different patterns did not differ in KHC from the alternating knots. For size 4/0, the parallel alternating knot with different patterns (S=S//S=S) showed higher KHC than the parallel alternating (S//S//S//S). This is shown in Figures 5.2a, b and Table 5.2.

Table 5.2
Statistical analysis of differences in knot holding capacities between the different sliding knots using the silk and nylon sutures with two sizes.

Knot Code	**Silk**	**Nylon**
2/0 USP		
SXSXSXS versus S=S=S=S	NS	NS
S#S#S#S versus S//S//S//S	$P<0.05$	NS
SXS#SXS versus S=S//S=S	$P<0.05$	NS
S#S#S#S versus SXS#SXS	NS	$P< 0.05$
S//S//S//S versus S=S//S=S	NS	NS
SXSXSXS versus SXS#SXS	$P< 0.05$	NS
S=S=S=S versus S//S=S//S	NS	NS
4/0 USP		
SXSXSXS versus S=S=S=S	NS	NS
S#S#S#S versus S//S//S//S	NS	NS
SXS#SXS versus S=S//S=S	$P<0.05$	$P<0.05$
S#S#S#S versus SXS#SXS	NS	$P< 0.05$
S//S//S//S versus S=S//S=S	$P<0.05$	$P<0.05$
SXSXSXS versus SXS#SXS	NS	NS
S=S=S=S versus S//S=S//S	$P<0.05$	NS

NS= not significant
Statistical significance is determined by two-tailed student's test for paired samples.

Referring to the same Figures and Table, the parallel alternating knot with different patterns (S=S//S=S) was superior in KHC to the nonidentical alternating knot with different patterns (SXS#SXS) for both sizes in silk. The simple sliding knots had higher KHC with 2/0 size (where the nonidentical knot (SXSXSXS) had high KHC than with 4/0 size.

Using the nylon suture, the alternating knots with different patterns were comparable to the simple sliding knots for both sizes. When compared to the alternating knots, of 2/0 size, the SXS#SXS knot was superior in KHC to the S#S#S#S knot. Whereas for the 4/0 size, the alternating knots with different patterns were superior in KHC to the alternating knots.

Compared to each other, the parallel allternating knot with different patterns (S=S//S=S) was superior in KHC to the alternating nonidentical knot with different patterns (SXS#SXS) for nylon, 4/0 size. They were similar in KHC for nylon 2/0 size. This is shown in Figures 5.2a,b and Table 5.2

5.1.3.3 Effect of Knot Configuration

The introduction of the alternating knots with different patterns seemed to be beneficial. Though they were comparable to the alternating knots in silk for the 2/0 size, the S=S//S=S knot was superior in KHC to the S//S//S//S knot for the 4/0 size. Likewise, in nylon 4/0, the two knots with different patterns were superior in KHC to the alternating knots, as shown in Figures 5.2a,b and Table 5.2. Also in nylon 2/0, the SXS#SXS knot was superior in KHC to the S#S#S#S knot.

5.1.3.4 Knot Efficiency

The knot efficiencies as shown in 5.2c,d did not vary much among the knots. The values, in general, ranged between 40 % to 74.5%. The S#S#S#S and the S//S//S//S knots of nylon 2/0 and 4/0 recorded low percentages, whereas the S=S//S=S knot of the same suture of 4/0 was the most efficient (66.67%). The knots of the silk suture had quite good efficiencies with no great variations among them.

5.1.3.5 Knot Breakage

The percentage of knots that reached failure by breakage is shown in Table 5.3 All the knots were considered secure when silk suture of both sizes was used. With nylon suture, however, the nonidentical alternating knot (S#S#S#S) and the simple parallel knot (S=S=S=S) were considered insecure for both 2/0 and 4/0 sizes, along with the alternating parallel (S//S//S//S) for size 2/0.

Table 5.3 Percentage of knots that reach knot failure by breakage of sliding knots

Knot Code	KNOT BREAKING (%)			
	Nylon		Silk	
	2/0	4/0	2/0	4/0
SXS#SXS	80	73	93	93
S#S#S#S	42	44	77	92
S//S//S//S	42	82	100	100
S//S=S//S	85	82	100	93
SXSXSXS	100	100	93	86
S=S=S=S	45	40	100	92

5.1.4 Discussion

Although the properties of a variety of different sliding knots have been reported by several authors, there is still a lack in the systematic analysis of sliding knots and especially the alternating sliding knots with different patterns [28]. This chapter compared the knot holding capacities of these knots in comparison to the alternating sliding and simple sliding knots (nonidentical vs identical). Nylon and silk sutures were used and two gauge sizes 2/0 and 4/0 USP The values of KHC of tested knots varied between the two materials and the two sizes. There was a 100% increase in knot holding capacities of loops when size 4/0 was compared to 2/0.Tera and Aberg [17], reported this same increase when size 2 was compared to size 0. Such information, together with sufficient knowledge about the physical properties of a knot, may provide a rationale basis for the selection of an adequately strong suture material and an adequately strong knot.

Secure knots, as stated by definition, fail by knot breakage in a percentage of 90. They break at a higher mechanical load than knots that fail by slippage [58]. This was proved in nylon. The S=S//S=S knot failed by breaking at high percentages (85 for 2/0 and 82 for 4/0). It had higher breaking loads (KHC: 20 N for 2/0 and 11 N for 4/0) than the S#S#S#S knot (KHC 17 N for 2/0 and 7 for 4/0) that failed at low breaking percentages (42 for 2/0 and 44 for 4/0) as illustrated in Table 5.3 and Figures 5.2 a,b. The same applies to the alternating identical (S//S//S//S) knot of 2/0 size.

However, there was an exception: the simple parallel sliding knot (S=S=S=S) of 2/0 and 4/0 size. Though it had a low breaking percentage (45 for 2/0 and 40 for 4/0 as shown in Table 5.3), it broke at a similar load (26 N for 2/0 and 10.5 N for 4/0) to the knots having high breaking percentage for example, the simple nonidentical sliding knot (SXSXSXS) (breaking percentage:100% for both sizes and KHC : 24 N for 2/0 and 9.6 for 4/0) as shown in Figures 5.2a, b and Table 5.3. For the S=S=S=S knot, to have such a high KHC along with a low breaking percentage, is quite surprising. It is wondered then, whether this high KHC value encourages the practical use of this knot.

Insecure knots are judged as knots that fail by slippage in nearly a 50:50 distribution [51]. Accordingly, taking the parallel sliding knot of nylon (S=S=S=S, size 2/0): out of 12 experiments, 5 knots broke and 7 slipped which accounts for a proportion higher than 50% of slippage. The same can be stated for the alternating nonidentical knot (S#S#S#S) of nylon 2/0 , where 7 slipped out of 12, and for size 4/0, where 5 slipped out of 9. Therefore, these knots are considered as insecure knots. The fact that reflects their unreliability, indicating the risk of extrapolating the findings of laboratory study to clinical practice. Even if the simple parallel sliding knot (S=S=S=S) of nylon size 2/0 failed at a high load (26.2 N) which might be clinically adequate, its breaking percentage is low (45 as shown in Table 5.3), meaning that this knot can fail by slippage or breakage in a 50 % probability. Therefore, it should be avoided in clinical use where the mechanical reliability of the suture prevails.

The alternating sliding knots with different patterns (SXS#SXS and S=S//S=S) were secure knots for both materials, denoting their possible use in clinical situations. The superiority of one over the other depended on the material used and the gauge size.

Smaller gauges of a monofilament suture have higher pliability than larger gauges because the diameter is smaller [58]. Therefore, the knot can be more likely to be tightly tied leaving no empty space between the alternating strands, hence, being stronger than that tied with a multifilament material of the same gauge. In addition, it is often thought that a monofilament suture has a much lower coefficient of friction than a multifilament suture [58]. Because of this, being a monofilament suture, it was easier to tie tightly the S=S//S=S nylon knot than a similar silk knot of the same gauge size. This brings out the comparison of both sutures, being monofilament (nylon), and multifilament braided (silk). However, it is worth noting here that nowadays, all braided sutures are coated to reduce their tissue drag, improve their knot run down and to make them softer on the surgeon's hands. This is the case with the silk suture since it is coated with silicone. Consequently, this leads us to conclude that braided sutures are expected to be comparable to monofilament sutures in knotting abilities. The experimental results confirm this conclusion.

In this chapter, only knots of four throws were tested for both materials. Even though, there were still some knots considered as insecure but only for nylon suture, the knots of silk sutures were all reliable. The parallel sliding knot (S=S=S=S) was one of insecure knots found in this chapter, a finding that confirms that of Trimbos [17]. Trimbos [18] demonstrated in an earlier study that knots consisting of nonidentical throws (SXS), and alternating parallel knots (S//S) are considered stronger than knots consisting of identical throws around same suture (S=S) with no significant difference between the nonidentical and parallel knots. He demonstrated this conclusion on three and five throws sliding knots using monofilament sutures like Prolene, PDS and Maxon, and multiflament sutures like Vicryl and Dexon-S. It is questionable how well this statement applies on nylon and silk sutures knots of four throws and two gauge sizes : 2/0 ansd 4/0 USP. Accordingly, comparing the SXSXSXS and S=S=S=S knots of both materials, no significant difference was found between them for both sizes of the two materials. Therefore, our results did not agree with those of Trimbos [18].

The study demonstrated the superiority in KHC of the nonidenical knot with different patterns (SXS#SXS) over the alternating nonidentical (S#S#S#S) of nylon 4/0 and the superiority in KHC of the parallel alternating knot with different patterns (S=S//S=S) over the alternating parallel knot (S//S//S//S) of nylon 4/0 too. From this perspective, if we consider that the newly introduced knots (SXS#SXS and S=S//S=S), are more complex than the alternating knots, the study shows that the strength of the knot increases with its complexity i.e. as the knots become more complex, they tend to be stronger.

As for the silk suture of size 4/0, it demonstrated the superiority of the S=S//S=S knot over the S//S//S//S knot supporting this hypothesis. As far as the size 2/0 is concerned, silk suture showed no significant differences among the knots that may relate their strength to the complexity of their structure. The nylon suture of size 2/0 confirmed the hypothesis denoting the superiority in KHC of SXS#SXS knot over the S#S#S#S knot.

However, there were exceptions, which we could not account for, like the high KHC of the SXSXSXS knot in silk 2/0 and nylon 4/0. This led us to the following conclusion: the comparison of the three categories of sliding knots: simple, alternating and alternating with different patterns using nylon as the monofilament suture and silk as the braided suture and the two gauge sizes 2/0 and 4/0 USP does not confirm the hypothesis that the strength of these knots increases with the degree of complexity (the complexity of the knots is considered in the assumption previously made).

The present study evaluated the performance of the alternating sliding knots with different patterns under dry conditions. But, sufficient knowledge about these knot configurations and their strength seem to be important. Accordingly, more research will be done that account for the behavior of these knots under in vivo conditions, where factors such as tissue strength, inflammatory reactions, body fluids and their effect on tensile strength upon implantation, resistance to infection and bacterial invitation will be further investigated.

5.2 Changes In Knot Holding Capacity of Sliding Knots Under In Vivo Conditions and Tissue Reaction

5.2.1 Introduction

Few investigations on the holding power of knots in vivo have been published. In 1971, Herrman found no significant difference in knot holding power after 24 hours soaking in plasma in synthetic suture materials, metallic sutures and silk whereas catgut showed reduction. He confirmed these findings, in 1973 in a series of experiments on rats and rabbits [11]. The knot type tested was the three square throw.

Tera and Aberg [59] have reported the strength of 12 types of suture thread in combination with 12 types of knots outside the living organism. The results obtained indicated that the type of knot was crucial to the holding power of the knotted thread. Trimbos and associates [18], studied the knot strength of different sliding knots. They indicated that the knot strength is dependent on both the type of the knot and type suture material. In an earlier study, his findings [17], revealed that the properties of various knots used in surgical practice differ considerably. Therefore, he declared that sufficient knowledge about knot configuration and knot strength seems to be an important aspect of surgical handicraft.

Maguillian et al. [60], showed that, in general, with synthetics, a degree of knot security approaching four squared throws can be achieved by two slip and three squared throws. His study compared knot security of various synthetic suture materials using silk as a standard. In their study, Rijssel et al, [61], concluded that the outcome of the comparison of square and sliding knots depends on knot configuration, suture material and suture size. Brown [23], examined three groups of knots to identify the most effective and efficient knotting techniques (square, surgeon' s and double throw knot). He used different suture materials (absorbable versus nonabsorbable) (monofilament versus braided), to determine their LHC, loop holding capacity and breaking force.

Shimi et al [62], evaluated the holding and tensile characteristics of five extracorporeal slip knots in relation to type and size of ligature materials. Lately, Trimbos et al [58], compared the three-throw, four-throw non-identical sliding knots and the three-throw square knot properties among five synthetic absorbable sutures.

All of these stated studies presented the in vitro tensile strength or the knot holding capacity and knot security of currently available suture materials. Perhaps, of more importance to the surgeon are the changes that may occur in these important suture knots properties during the postoperative period.

Seid et al [63], used an animal model for measurement of incision strength after suture repair in an attempt to compare the interrupted and continuous mass closure for abdominal incisions. Herrmann [11], designed experiments to evaluate the changes in breaking strength and knot security in the early postoperative period.

The reason that there should be concern in the changes in tensile strength of the sutures after being implanted in animals, arises of the fact that the in vivo environment is thought to alter the physical properties of sutures whether it is in strand or knot configuration.

As far as the sliding knots are concerned, (as previously stated), there is the type of knot where the strand alternates using a pattern other than with every throw. They are known as the sliding alternating knots with different patterns [28]. In this chapter, this type i.e. the alternating sliding knots with different patterns will be compared to the alternating knots. The question, here, is how well these knot configurations will behave with regard to the knot holding capacity under in vivo conditions.

Besides that the suture must be able to maintain an adequate tensile strength to make certain that the knot does not slip, the surgeons demand a suture which does not produce tissue reactions that considerably weaken the holding power of the wound and delays healing.

Many studies have been done to assess different tissue reactions to various suture materials. Recently, more attention is devoted to the knot site of the surgical loop. Rijssel et al. [43], studied the tissue reaction to the simple nonidentical sliding knot using polyglactin 910 and polyglyconate and correlated the degree of tissue reaction surrounding the knot with the suture size, number of throws added and knot volume.

The aim of this chapter is to evaluate the changes in knot holding capacity or strength, knot efficiency and knot security in vivo of a newly introduced sliding knots :the alternating sliding knots with different patterns [28]. It is also, to compare them to the alternating sliding knots after several periods of implantation subcutaneously in the abdominal wall of rats. Silk and nylon sutures of two USP sizes: 2/0 and 4/0 are used. In addition, it assesses the tissue reaction to these knots using Sewell's scoring system [33]. Throughout the study, the knot configuration, knot volume and gauge size are considered. The study, also provides a comparison of the knot holding capacities and efficiencies of these knots in vivo to the values obtained in vitro (performed in earlier experiments).

5.2.2 Materials and Methods

Silk and nylon sutures of both USP sizes:2/0 and 4/0 were used for this chapter. They are chosen as multifilament (silk) and monofilament (nylon) non-absorbable sutures. Four knot configurations were used: the parallel alternating sliding knot (S//S//S//S), the nonidentical alternating sliding knot (S#S#S#S), the parallel alternating with different patterns (S=S//S=S) and the nonidentical alternating with different patterns (SXS#SXS). They are illustrated in Figure 5.1 [28].

Wistar rats weighing about 230 g each were used. Animal care was taken so that to confirm to the NIH regulations. A total of one hundred and twelve rats were used for the combination of every suture material, size, and the four knot configurations taken at different intervals of time: fourth, seventh, eleventh and twentieth day postoperatively. One rat was used for every four knot configurations. Seven rats were used for every material and size (silk 2/0 (7), nylon 2/0 (7), silk 4/0 (7) and nylon 4/0 (7)). Accordingly, at the fourth day, groups of twenty eight rats were sacrified. The same was true for the seventh, eleventh and twentieth days to end up with a total of 112 rats.

Experiment I

For tensile strength measurement, the loops of the suture material were tied around two cylindrical rods (made up of wood) attached to a board [4]. The knots were tied and the type of the knot was snugged tight. The diameter of each rod was 1 cm and the distance between them was 8 cm to fit for implantation. The knots were tied by the same person to minimize variability and snugged down by moderate force.

The alternating sliding knots were tied with alternating right and left hand throws, the standing part and the hitch alternating strands with every successive throw. Alternating the hitches is most easily accomplished by alternating the hand that snugs down the throw, with the standing part held in the other hand Figure 5.3 [19]. With the alternating knots of different patterns, two throws were made on a single standing part and then, alternating the standing part with hand to perform the other two throws on the new standing part.

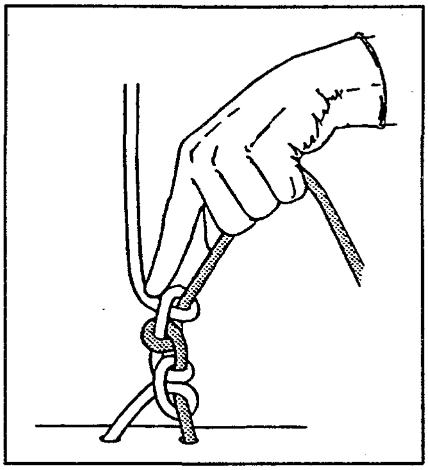


Figure 5.3 The alternating sliding knot [19].

The rats were anesthetized with an intramuscular ketamin HCL injection (80 mg/kg). The skin was undermined (midline incision) so as to create four subcutaneous pouches, two on each side as shown in Figures 5.4 and 5.5. Every loop corresponding to a different knot configuration was implanted in one of these pouches giving a total of four knots implanted in one rat. The incision was then closed with a running suture. For every suture material and every size, a group of seven rats was used (N=7).

The rats were then sacrified at the specified day, and the loops were dissected free from surrounding tissue, placed in saline solution to prevent drying, then mounted on Zwick 1446 Universal machine and distracted at a rate of 50 mm/min. The loop method is followed [28].

Experiment 2

For tissue reaction, the rats were anesthesied with an intramuscular ketamin HCL injection (80 mg/kg). A vertical skin incision in the midline of abdominal wall was performed under aseptic conditions. An incision of around 3 cm is made through the musculo-aponeurotic (linea alba) along which the four sliding knots were tied with moderate force. This is illustrated in Figure 5.6 a, b. End edges were cut to 2 mm. The knots were separated from each other at around 1 cm intervals and from the incision edge at 0.5 cm [53, 64]. All operative procedures were aseptically performed with sterile materials to avoid contamination.

At the desired day, rats were sacrificed by ether asphyxiation. Knots were taken along with the surrounding tissue being excised, fixed in 10% formalin, and embedded in paraffin as shown in Figure 5.7a and b. For histological examination, sections of 4 to 5 μ were taken, stained with hematoxylin-eosin and examined under light microscopy.

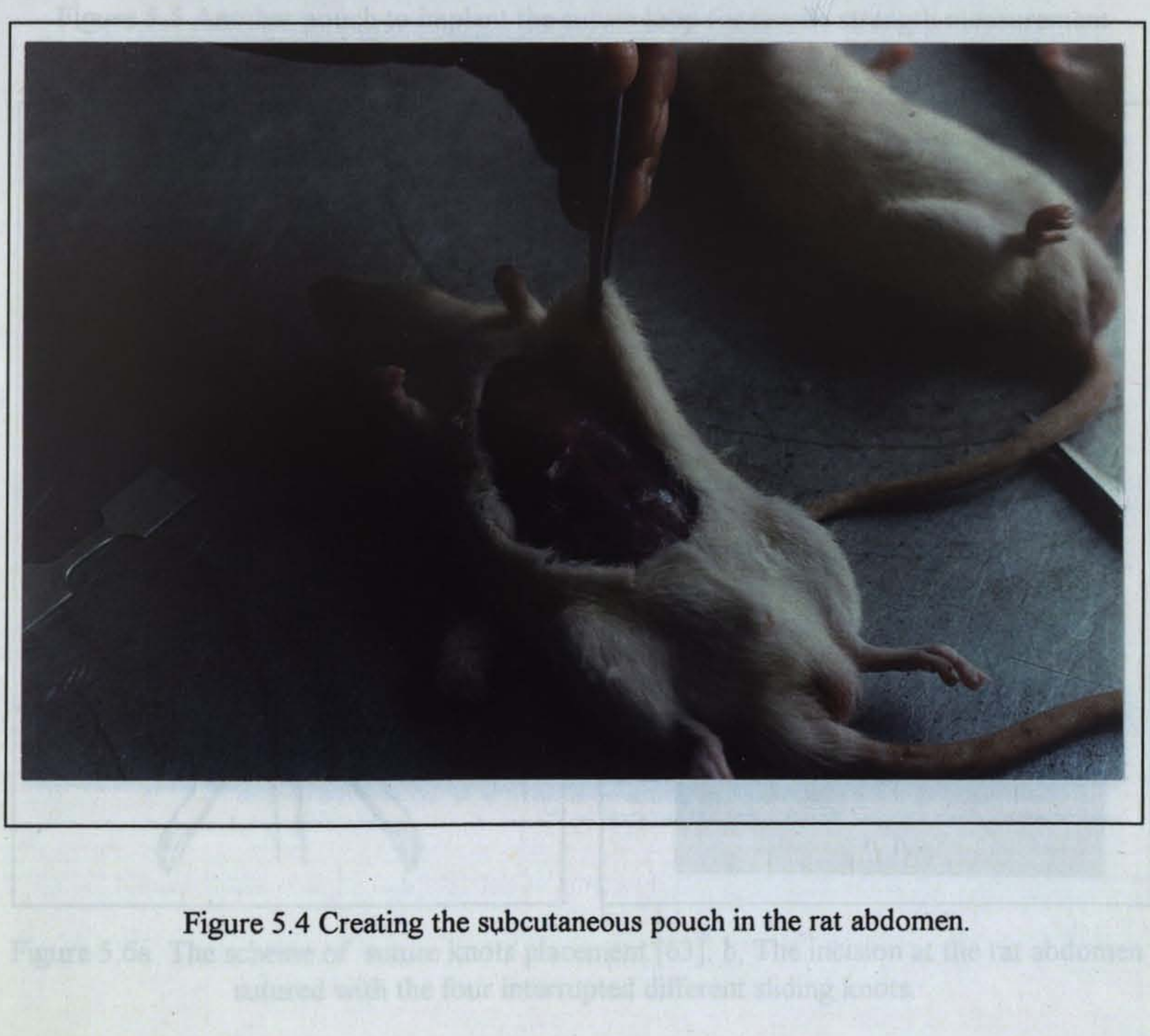


Figure 5.4 Creating the subcutaneous pouch in the rat abdomen.

Figure 5.5 Another pouch to implant the suture loop for tensile strength measurement

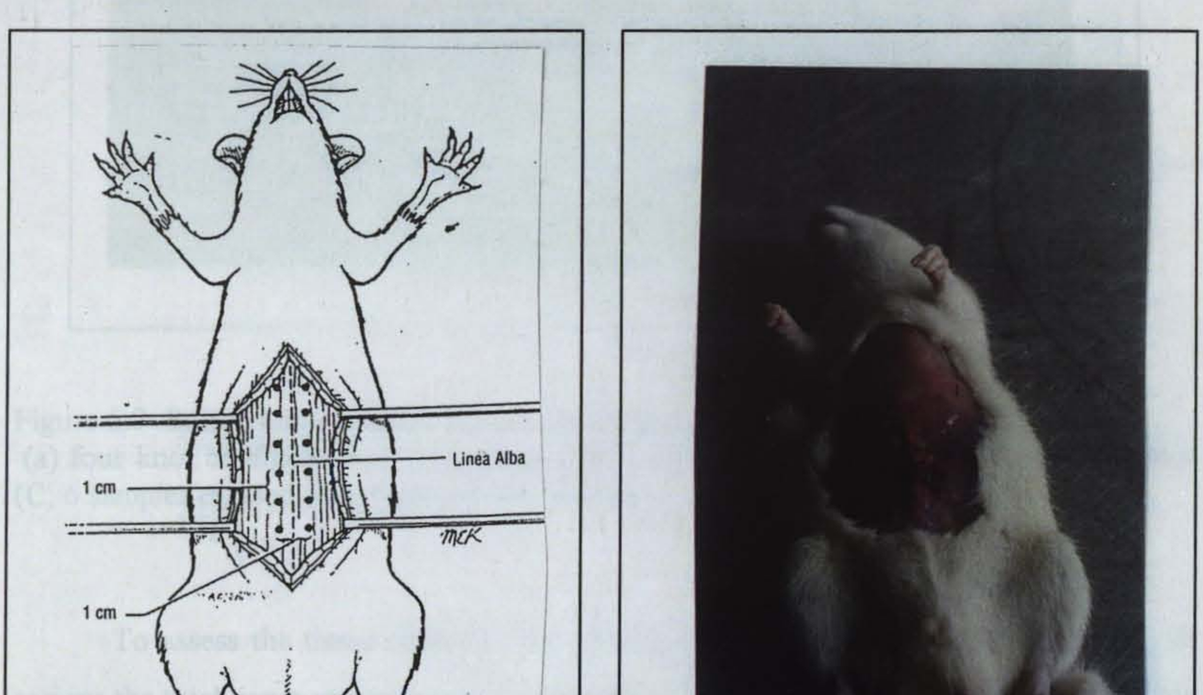


Figure 5.6a The scheme of suture knots placement [63]. b, The incision at the rat abdomen sutured with the four interrupted different sliding knots.

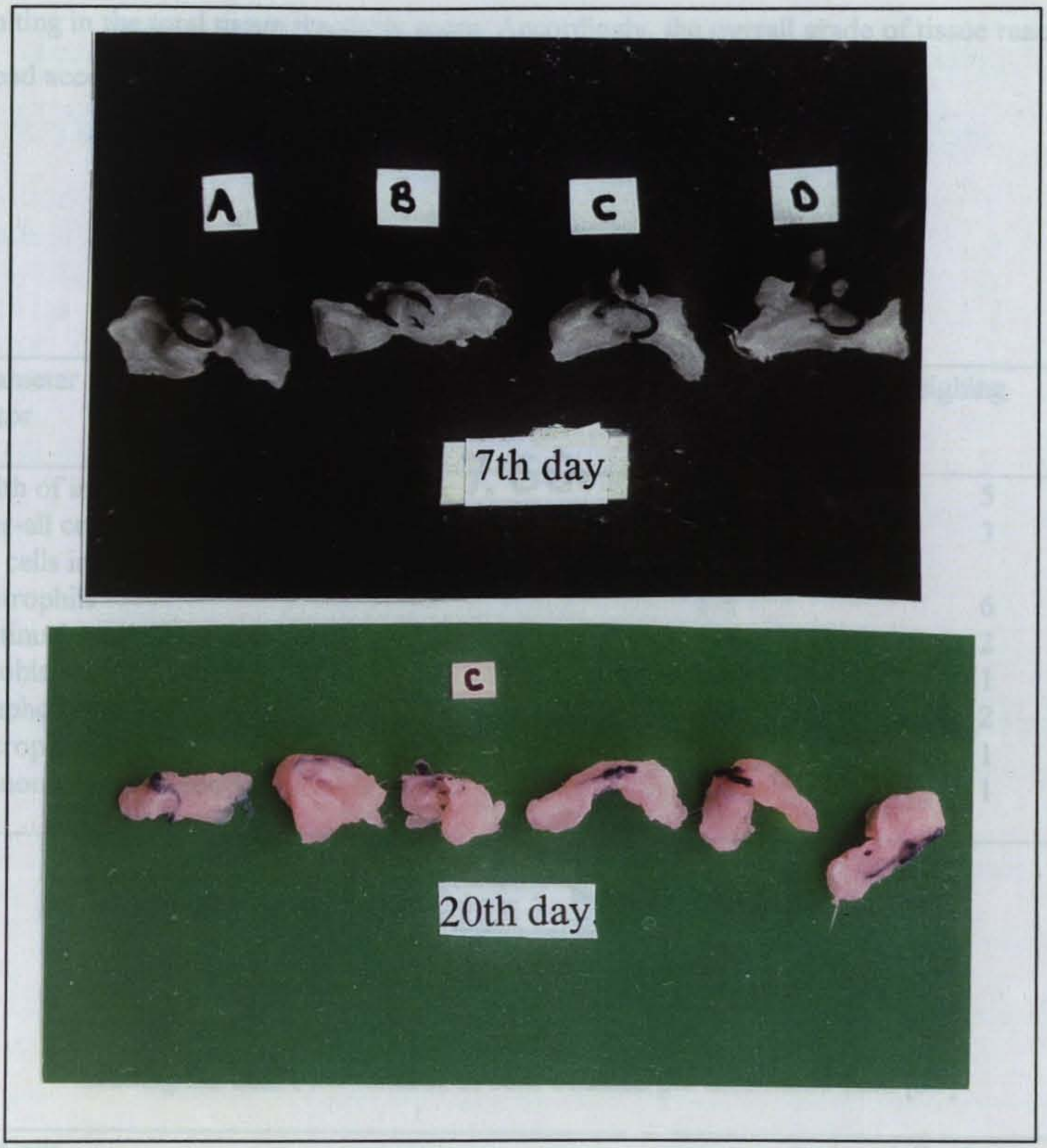


Figure 5.7 Suture knots excised with surrounding tissue for histological studies. (a) four knot configurations designed as A;B,C, and D at the 7th day. (b) S#S#S#S knot (C; 6 samples excised from 6 rats) at the 20th day.

To assess the tissue reaction , the scoring system of Sewell was adopted [33]. It assigns the total tissue reactivity score composed of eight parameters, as illustrated in Table 5.4.

The average values are calculated for each parameter, graded as shown in Table (5.5a and b), multiplied by a specific weighing factor, as shown in Table 5.4 and summated resulting in the total tissue reactivity score. Accordingly, the overall grade of tissue reaction is read according to Table 5.6 [33].

Table 5.4
Histological Parameters used to assess tissue reaction surrounding the knots and weighing factors [33].

Parameter Factor	Grade	Weighing
Width of inflammation zone (mm)	1-8	5
Over-all cell density	1-8	3
No. cells in inflammatory zone		
Neutrophils	1-8	6
Multinucleated Giant cells	1-8	2
Fibroblasts	1-8	1
Lymphocytes	1-8	2
Macrophages	1-8	1
Mononuclear Phagocytes	1-8	1

Table 5.5a
Grading the cells ; the number of cells counted per immersion field [33]

No. Cells	Assigned Grade
1-5	1
6-15	2
16-20	3
21-35	4
36-50	5
51-100	6
101-150	7
greater than 150	8

Table 5.5b
Grading the depth of inflammatory zone (diameter in high power field x 430) [33]

Depth of reaction zone	Assigned Grade
0.01-0.25	1
0.26-0.33	2
0.34- 0.50	3
0.51- 1.0	4
1.1-2.0	5
2.1-3.0	6
3.1-4.0	7
4.1 and above	8

Table 5.6
The assignment of the over-all grade of tissue reaction [33]

Total points	Grade of tissue reaction	Name assigned to grade
0-16	1	very slight
17-32	2	Slight
33-48	3	Slight to Moderate
49-64	4	Moderate
65-80	5	Moderate to Marked
81-96	6	Marked
97-112	7	Marked to Extensive
greater than 112	8	Extensive

The volume of each knot was determined as follows: the diameter and the height of the knot were determined using a micrometer. From these values, the content of the cylinder fitting the particular knot is calculated. Therefore, the volume of knot cylinder is calculated mathematically ($V = \Pi R^2 h$) [49]

By measuring, under the microscopy, the width and height of the inflammatory reaction surrounding the knot, it was possible to calculate the volume of the inflammatory cylindrical tissue reaction surrounding the knot. The difference between the volume of the inflammatory cylinder and that of the knot cylinder is calculated to represent the amount of tissue-reaction-sheath surrounding the knot [49]. Knot volume cylinder is $V_1 = \Pi R_1^2 h_1$.

The tissue cylinder volume is $V_2 = \Pi R_2^{\,2} h_2$. The tissue reaction sheath is $V_2 - V_1$. This is illustrated in Figure 5.8 [49]

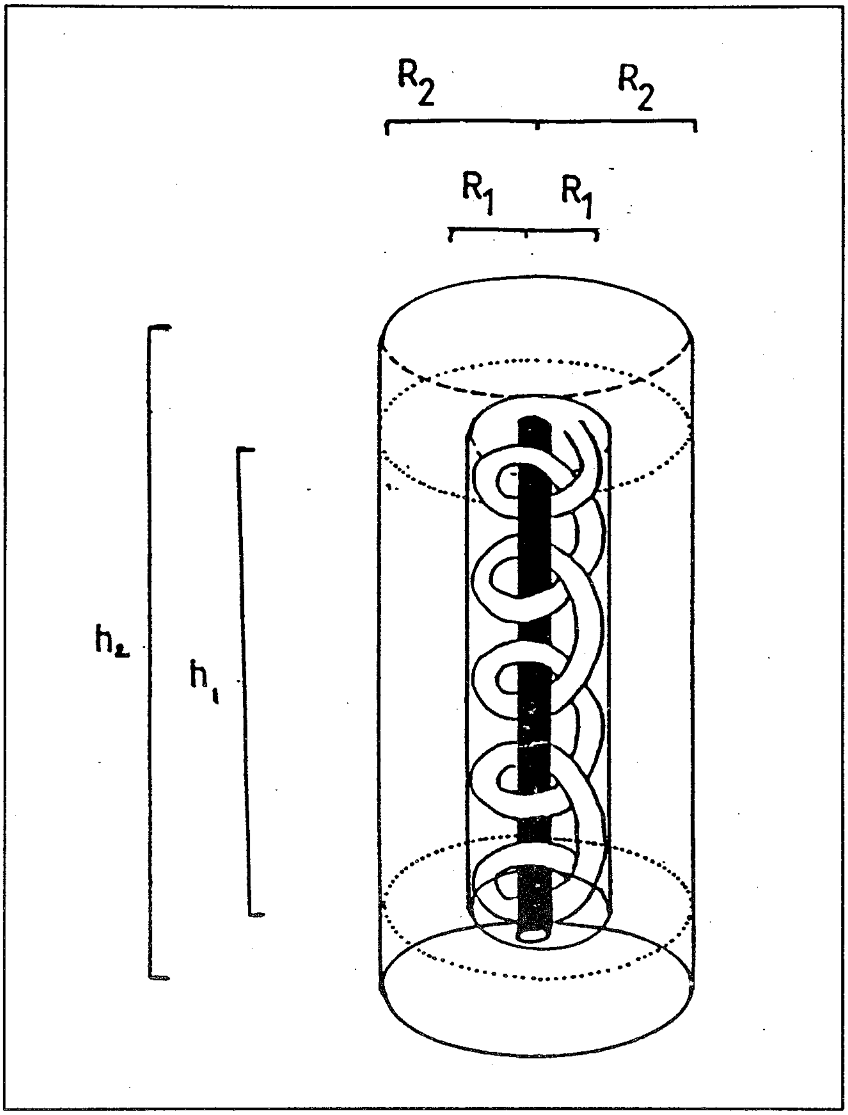


Figure 5.8 Knot Volume $V_1 = \Pi R_1^{\,2} h_1$, tissue reaction volume $V_2 = \Pi R_2^{\,2} h_2$ and inflammatory sheath volume $V_2 - V_1$

5.2.3 Results

Experiment I: Changes in Knot Holding Capacity Throughout Twenty Days

The loop holding capacity was defined as the maximum force calculated from the load-elongation curve required to break the tied suture loop, by loading the part of the suture that forms the loop. Knot failure is defined as the break of the knot or its slippage exceeding two mm used as standard [18]. The knot holding capacity (KHC), is the force that a specified suture can sustain without failing either through the suture breaking or the knot slipping. The formula for determining the KHC varies according to the testing method, KHC equals one-half the force required for rupture i.e. the loop holding capacity. For the single strand method, KHC equals the force required for rupture [28].

Figures 5.9 and 5.10 show the changes in tensile strength or knot holding capacity (KHC) in newtons for silk and nylon sliding knots of 2/0 and 4/0 respectively.

The mean KHCs of the knots are recorded at the zero, fourth, seventh, eleventh and twentieth days postoperatively. Statistical differences between the KHC values are determined by a multivariate ANOVA test with significance level 0.05.

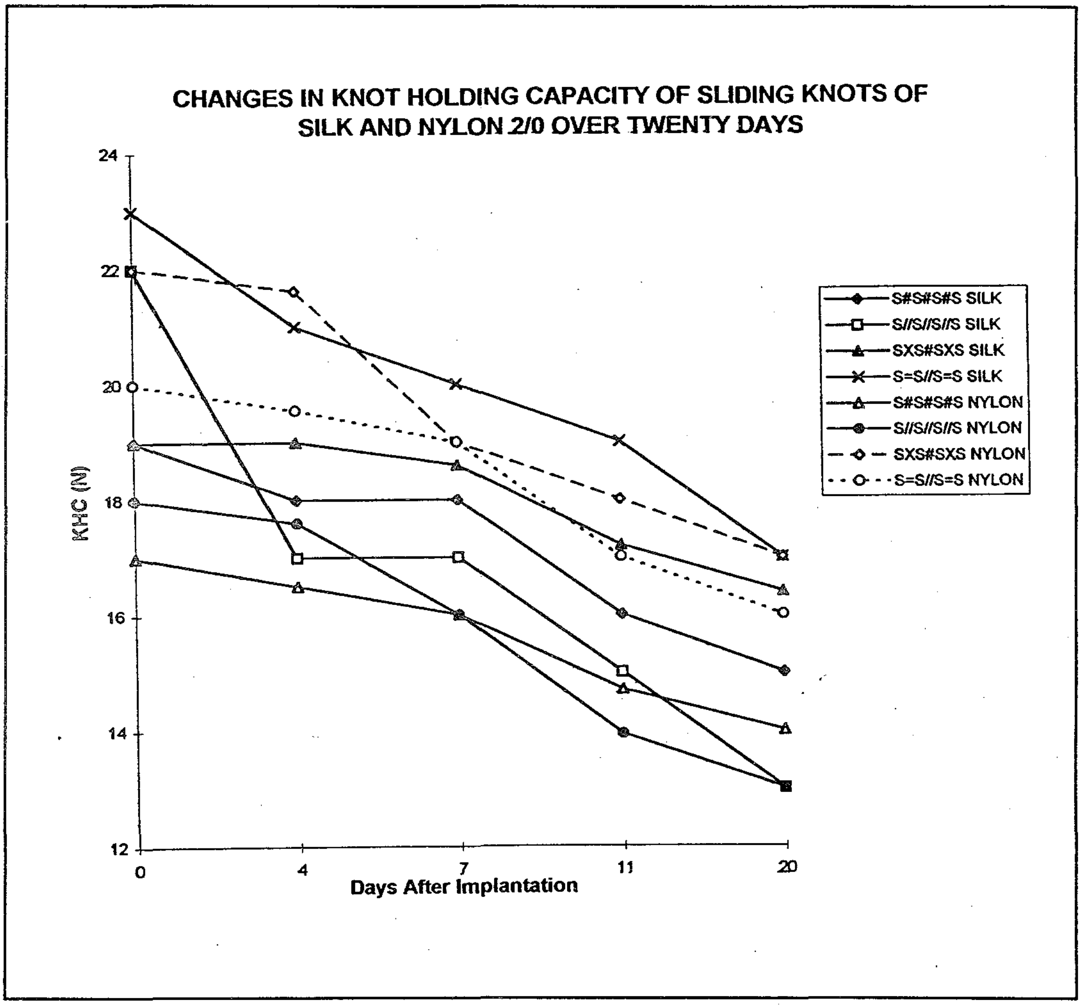


Figure 5.9 Changes in KHC of sliding knots of silk and nylon 2/0 over twenty days of implantation.

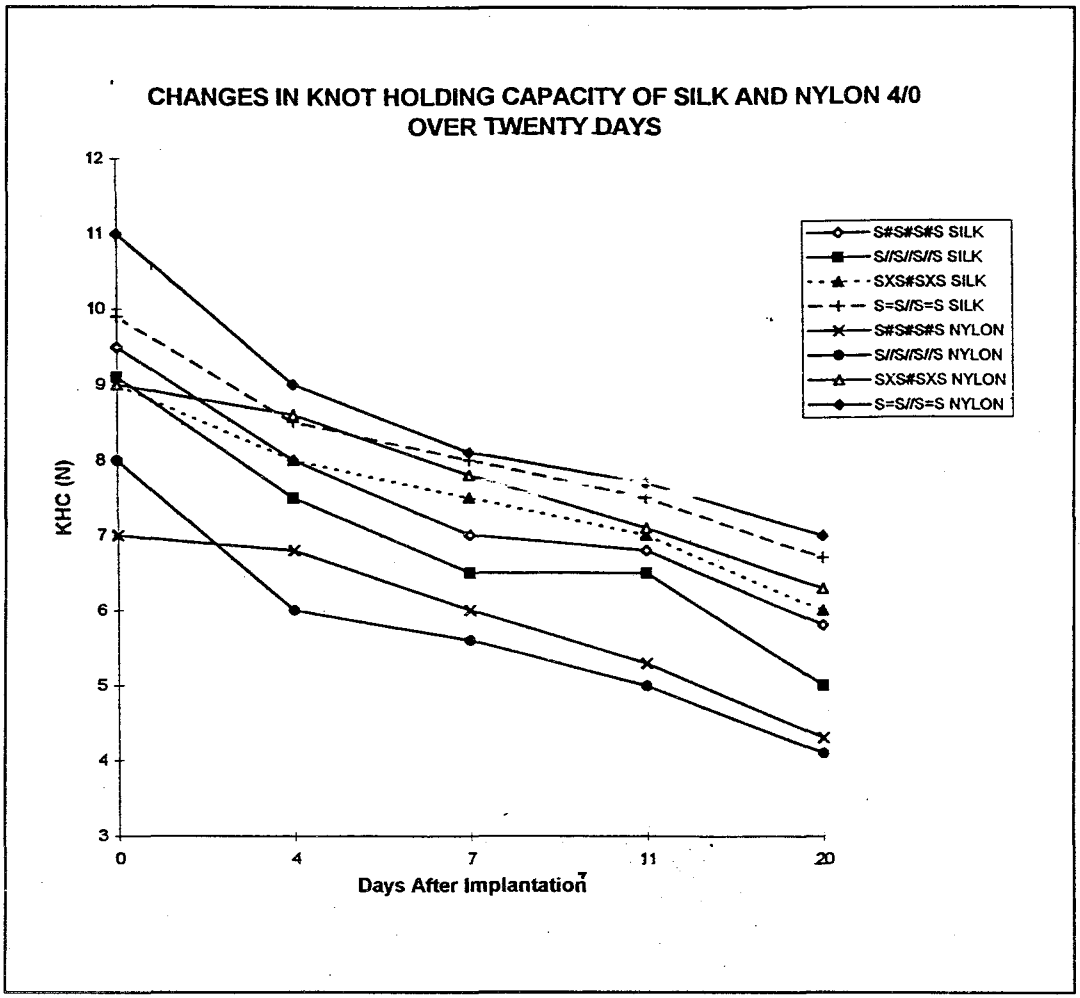


Figure 5.10 Changes in KHC of silk and nylon 4/0 over twenty days of implantation.

For loops of both sizes, the knot configuration, postoperative day and suture material were all important factors in determining the KHC, (p = 0.000) (see Table 5.7). The knot configuration along with the suture material seemed to have some significant effect on KHC (p = 0.002 for 2/0 loops and 0.000 for 4/0 loops). The factor of both the suture material and the postoperative days was not significant as well as the interaction of all the three factors together (knot configuration, postoperative days and suture material) as shown in Table 5.7.

Table 5.7
Multivariate analysis of knot configuration, postoperative days and suture material as possible factors determining the suture knot holding capacity

Analysis of variance	MS	DF	F-Ratio	P
Size : 2/0				
Knot configurations	126.0	3	31.0	0.000
Postoperative days	145.35	3	36.0	0.000
Suture material	51.5	1	13.0	0.000
Knots x days	1.6	9	0.4	0.038
Knots x material	20.4	3	5.0	0.002
Days x material	2.7	3	0.6	0.575
Knot x days x material	1.5	9	0.4	0.953
Size : 4/0				
Knot configurations	25.4	3	43	0.000
Postoperative days	45.2	3	76	0.000
Suture material	36.5	1	61.3	0.000
Knots x days	0.2	9	0.4	0.937
Knots x material	24.0	3	40.0	0.000
Days x material	0.4	3	0.74	0.527
Knot x days x material	0.1	9	0.25	0.986

MS = Mean -square; DF = degree of freedom, P = significance. N.B.= the data sheet for KHC values is given in Appendix A.

As illustrated in the Figures 5.9 and 5.10, all the knots of both materials and both sizes showed a decrease in KHC over the twenty days of implantation. The percent decrease in KHC is shown in Table 5.8. The alternating nonidentical knot of different patterns (SXS#SXS) recorded a decrease in KHC ranging from (14.5 to 33.5) for silk and nylon of both sizes. The alternating parallel knot with identical patterns (S=S//S=S) range was: (20-38.5)%, the alternating nonidentical (S#S#S#S): (17.6-40)%, and the alternating parallel (S//S//S//S): (28-49)%, as shown in Table 5.8. Obviously, the parallel alternating knot (S//S//S//S) presented the highest range of decrease in KHC among all the knots and the highest percent value i.e. the most decrease in KHC: 49 % corresponding to 4/0 nylon.

Table 5.8
The percent decrease in KHC of the sliding knots of silk and nylon sutures for 2/0 and 4/0 sizes throughout twenty days of implantation.

	Percent Decrease in Strength (%)			
Suture Material	Silk		Nylon	
USP	2/0	4/0	2/0	4/0
Knot Code				
S#S#S#S	18.0	33.5	17.6	39.0
S//S//S//S	38.0	30.0	28.0	49.0
SXS#SXS	14.5	33.4	22.6	33.0
S=S//S=S	25.0	32.0	20.0	38.5

All the knots reported a higher percent decrease in KHC for 4/0 gauge size than for 2/0, referring to Table 5.8 with the exception of S//S//S//S knot in silk. The multivariate analysis of variance, Table 5.9, revealed that the knot configuration as well as the suture size were important significant factors in determining the percent decrease in KHC. The relatively small sample size (N=7) might have influenced the level of significance corresponding to the factor of suture (p =0.057). So, it is probable that a greater number of experiment would change it. The interactions of knot configuration, and suture size as well as the suture material and size and all of the knot configuration, suture material and size were found to be significant, as shown in Table 5.9.

Table 5.9
Multivariate analysis of knot configuration, suture material and suture size as possible factors determining the suture knot holding capacity percentage decrease.

Analysis of variance	MS	DF	F-Ratio	P
Knot configurations	585.7	3	9.8	0.000
Suture material	220.8	1	3.7	0.057
Suture size	4496.88	1	75.8	0.000
Knots x material	16.4	3	0.2	0.842
Knots x size	161.3	3	2.7	0.049
material x size	599.8	1	10.11	0.002
Knot x material x size	421.1	3	7.1	0.000

The data sheet is given in Appendix B

Changes in Knot Efficiency Throughout Twenty Days

Knot efficiency is the knot holding capacity expressed as the percentage of the breaking strength (in vivo) of the unknotted thread. The in vivo tensile strength of the single strand is taken from separate experiments and is shown in Figure 5.11. The in vivo knot efficiencies of the tested knots are shown in Figures 5.12 and 5.13 for silk and nylon knots of 2/0 and 4/0, respectively throughout the postoperative day.

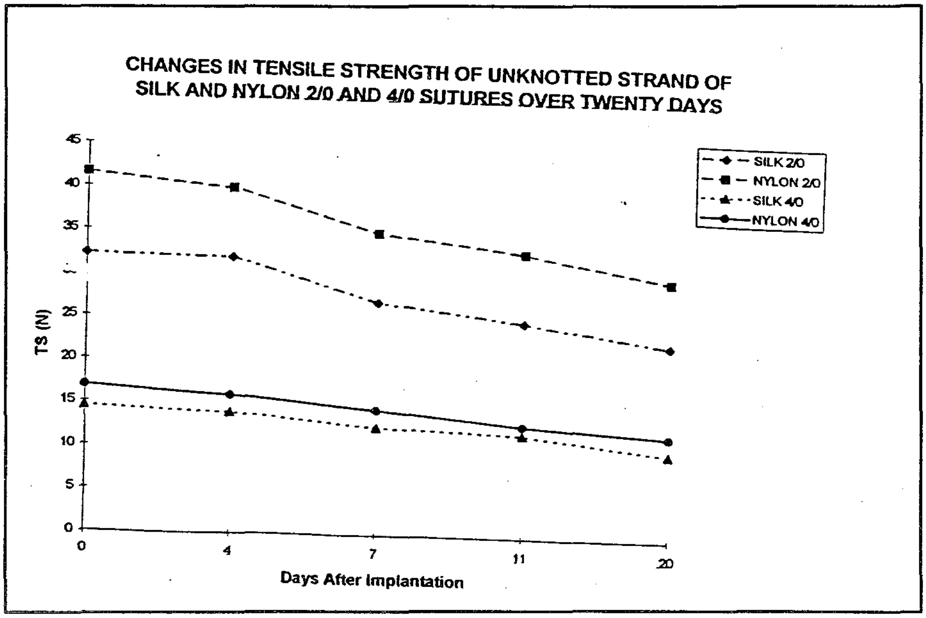


Figure 5.11 In vivo tensile strength of the single strand of silk and nylon.

Most of the in vivo knot efficiency values were higher throughout the post-implantation days (i.e. going from day 4 to 7 to 11 up to the 21st) since the suture looses strength in the tissue. Among the silk knots, the highest in vivo knot efficiency value (KE) belonged to the alternating parallel knot with different patterns (S=S//S=S) at the eleventh day. For 2/0 size, it recorded a 79 % value compared to the in vitro value of 74 % which was also the highest among the in vitro KEs of the silk 2/0. For 4/0 size, the S=S//S=S knot recorded a 67.4 % value compared to the in vitro value, 68.8% which was also the highest among the 4/0 knot efficiencies.

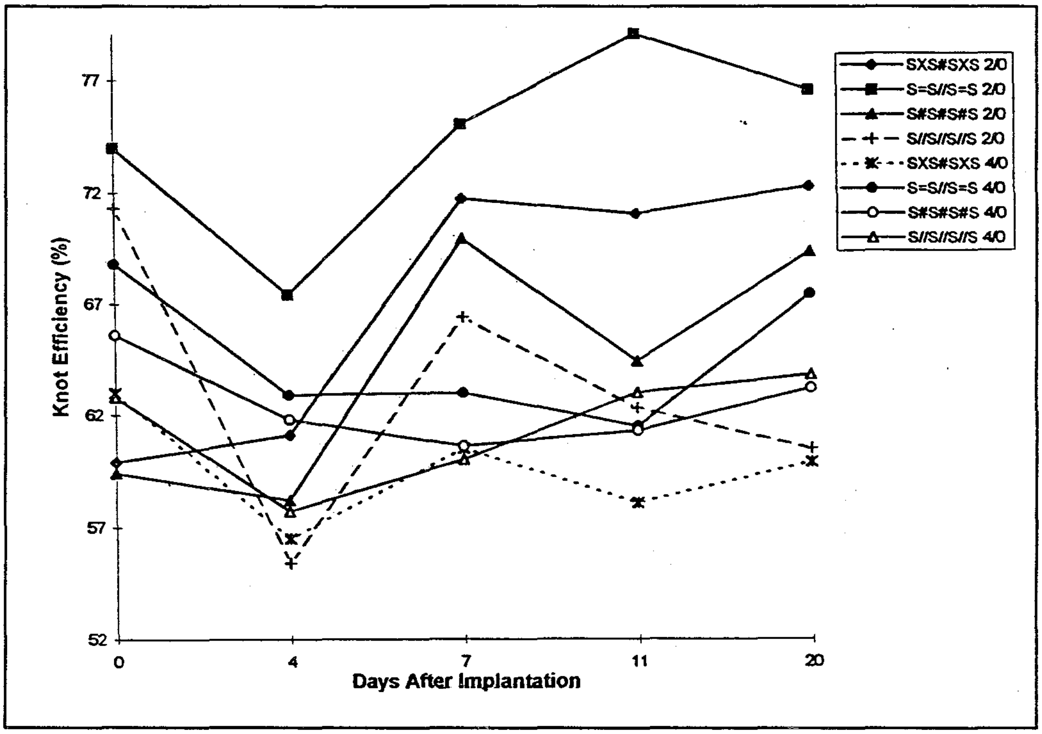


Figure 5.12 The changes in knot efficiency of the sliding knots of silk 2/0 and 4/0 .

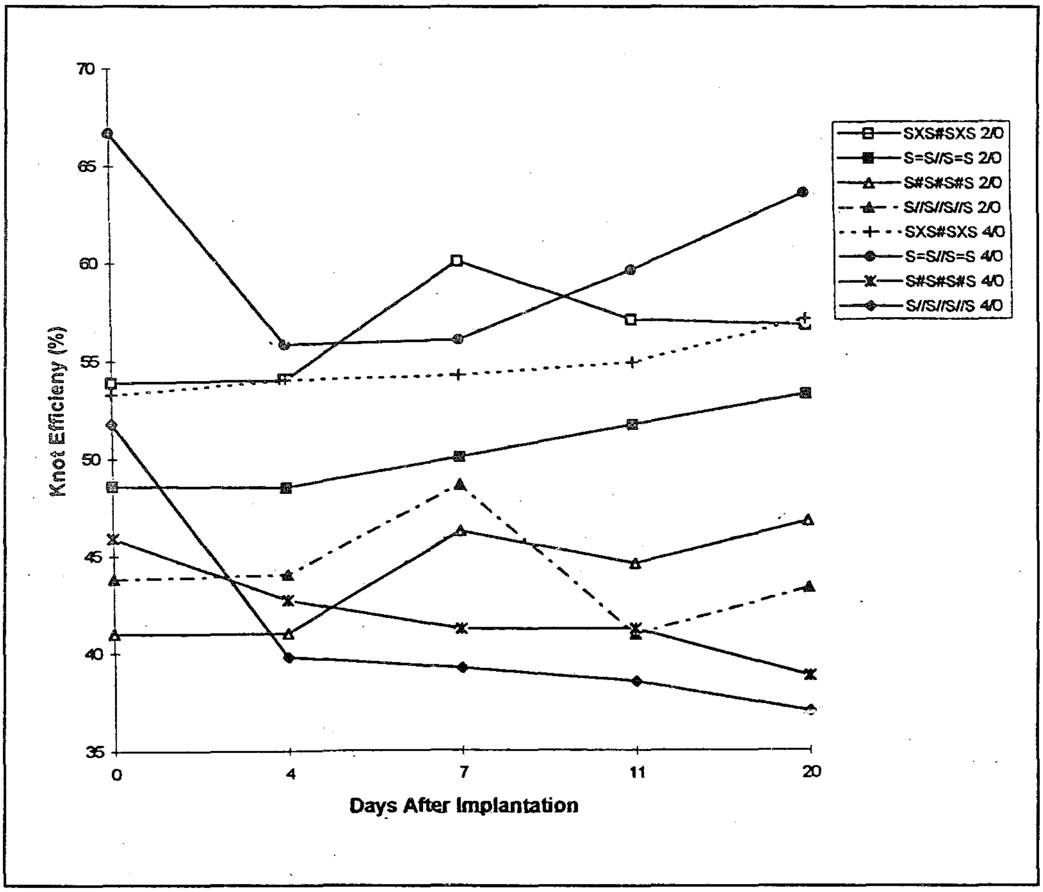


Figure 5.13 The changes in knot efficiency of the sliding knots of nylon 2/0 and 4/0 .

Among the nylon knots, of 2/0 size, and referring to the same Figures, the alternating nonidentical knot with different patterns (SXS#SXS) recorded the highest in vivo KE value, again corresponding to the seventh day (60 %) compared to the in vitro value (53.9%). For the 4/0 nylon knots, the alternating parallel knot with different patterns (S=S//S=S) recorded the highest in vivo value (63.4%) at the twentieth day, compared to the in vitro value (66.67%). As shown in Figures 5.12 and 5.13, among the silk 2/0 and 4/0 knots, the alternating parallel knot with different patterns had the highest KE at the twentieth day. Among the nylon 2/0 knots, the nonidentical knot with different patterns had the highest KE value. The parallel one with different patterns showed the highest KE value among the nylon 4/0 knots again at the twentieth day.

Knot Slipping

Knots that slipped more than 2 mm were considered to fail by slippage [28]. The most slipping was seen with the parallel alternating knot (S//S//S//S). Referring to Table 5.10, in silk 2/0, this knot slipped at the eleventh day (1 out of 6 at 11N) and at the twentieth day (1out of 7 at 11.3N). In nylon 2/0, it slipped again at the eleventh and twentieth day (1of 6 at 13.3 N and 2 of 6 at 12 N respectively). In nylon 4/0, the S//S//S//S knot slipped at the fourth day (1 of 7 at 2N) and at the twentieth day (2 of 7 at 3.6 and 3.5 N). Slippage was commoner in nylon knots than in silk knots. The nonidentical alternating knot slipped in silk 2/0 and nylon 4/0, Table 5.10. None of the knots slipped using silk 4/0. The alternating knots with different patterns showed no slippage for both silk and nylon of both sizes. This is illustrated in Table 5.10.

Table 5.10
The KHC (N) of knots that slipped with regard to knot configuration, postoperative day and suture material

Days	S#S#S#S	S//S//S//S	SXS#SXS	S=S//S=S
Silk 2/0				
11th	12 (1,7)	11 (1,6)	----	----
20th	12.75 (1,7)	11.3 (1,6)	----	----
Nylon 2/0				
11th	----	13.3 (1,6)	----	----
20th	----	12 (2,6	----	----
Nylon 4/0				
11th	----	2 (1, 7)	----	----
20th	3 (1, 7)	3.6, 3.5 (2, 7)	----	----

N.B: The numbers in the parentheses indicate e.g.(1, 7) that out of seven knots, one has failed by slippage.

Experiment II: Tissue reaction to knots

Figure 5.14 presents the mean over-all grade of tissue reaction for the sliding knots of silk and nylon sutures of both sizes throughout the twenty days of implantation. The over-all grades reaction varied between moderate, moderate to marked, marked and marked to extensive. There were no great variations between the grades among the knots of the silk and nylon materials, for both sizes. Only, the knots of nylon 2/0 showed the highest over-all grade at the fourth day, as shown in Figure 5.14. These knots were alternating parallel knots with different patterns (S=S//S=S) and the alternating nonidentical one (S#S#S#S). The grade was 8, corresponding to an extensive reaction. Of lower grade were the alternating nonidentical knots with different patterns (SXS#SXS) and the parallel alternating one (S//S//S//S) of nylon 2/0, of grade 7, corresponding to a marked to extensive reaction. The rest of knots showed reactions alternating from (marked) to (moderate to marked).

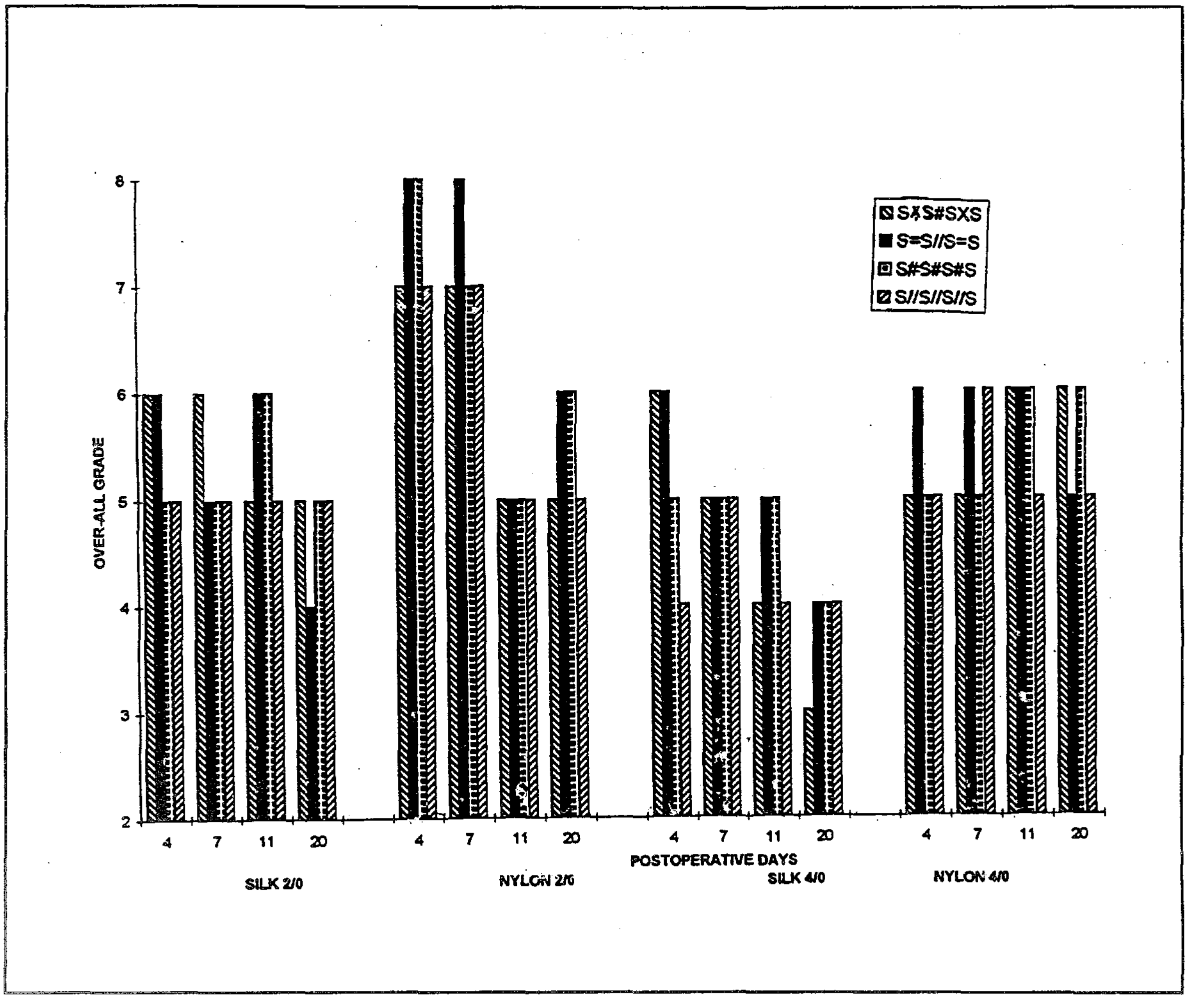


Figure 5.14 Tissue reaction for the silk and nylon throughout the implantation time.

All the knots showed a decrease in over-all tissue reaction from the fourth day to the twentieth. With regard to the twentieth day, the lowest grade belonged to the alternating nonidentical with different patterns (SXS#SXS) for silk 4/0, as shown in Figure 5.14. The highest grade belonged to the alternating parallel with different patterns (S=S//S=S) of nylon 2/0 and the alternating nonidentical (S#S#S#S) one for nylon of both sizes.

On microscopic examination, it was adequate to concentrate upon the tissue reactions on the fourth day when a maximum of exudation is found in the wound and on the twentieth day where the degree of exudation decreases and fibroplasia is normally predominant. In addition, statistical analysis of the average values of histological parameters among the postoperative days revealed a significant difference in these values between the fourth and the twentieth day for all the knots of the two materials and two sizes as well.

Histologically, the tissue reaction to the suture knots consisted of edema, a formation of inflammatory zone around the knot. The diameter and depth of the inflammatory zone varied throughout the postoperative period. The reaction is mainly a neutrophilic infiltration accompanied by a monophagocytic invasion that decreased all the way to the twentieth day postoperatively, as shown in Figures (5.15 - 5.18),. Lymphocytes, eosinophils and giant cells varied in both occurrence and number among the knots and throughout the postoperative period. Fibroblast formation began at the seventh day and predominated at the twentieth day.

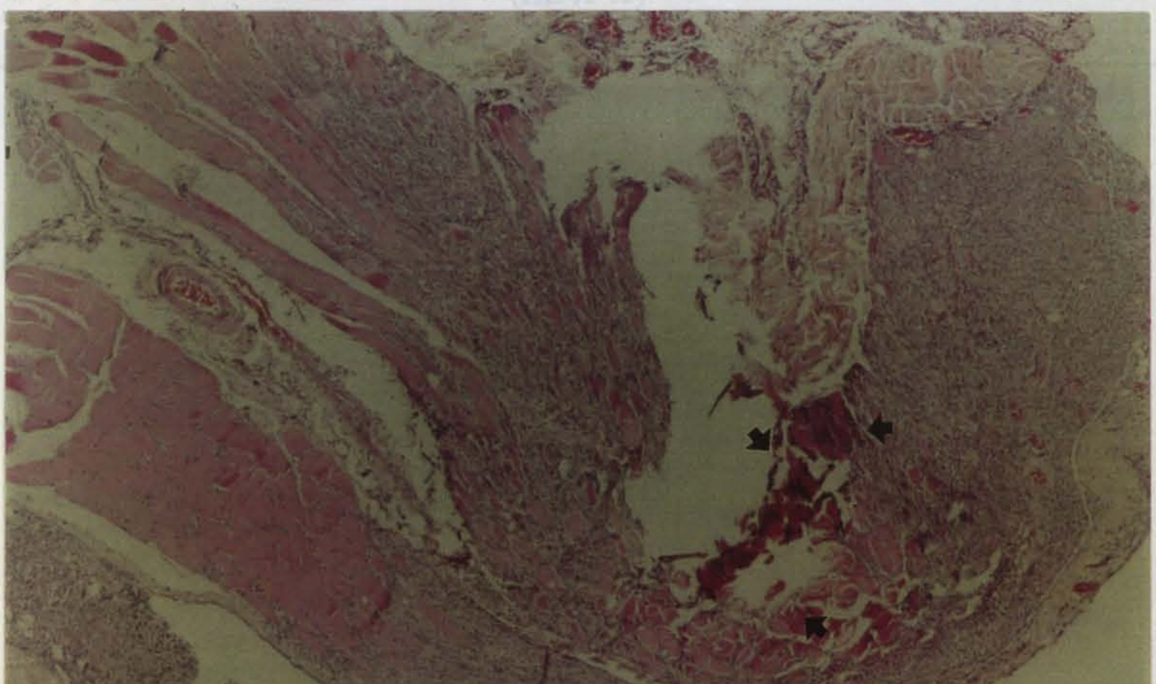


Figure 5.15 Tissue reaction surrounding the SXS#SXS knot in silk 2/0, at the 4th day. Note the inflammatory infiltration zone around knot area. (HEX 40)

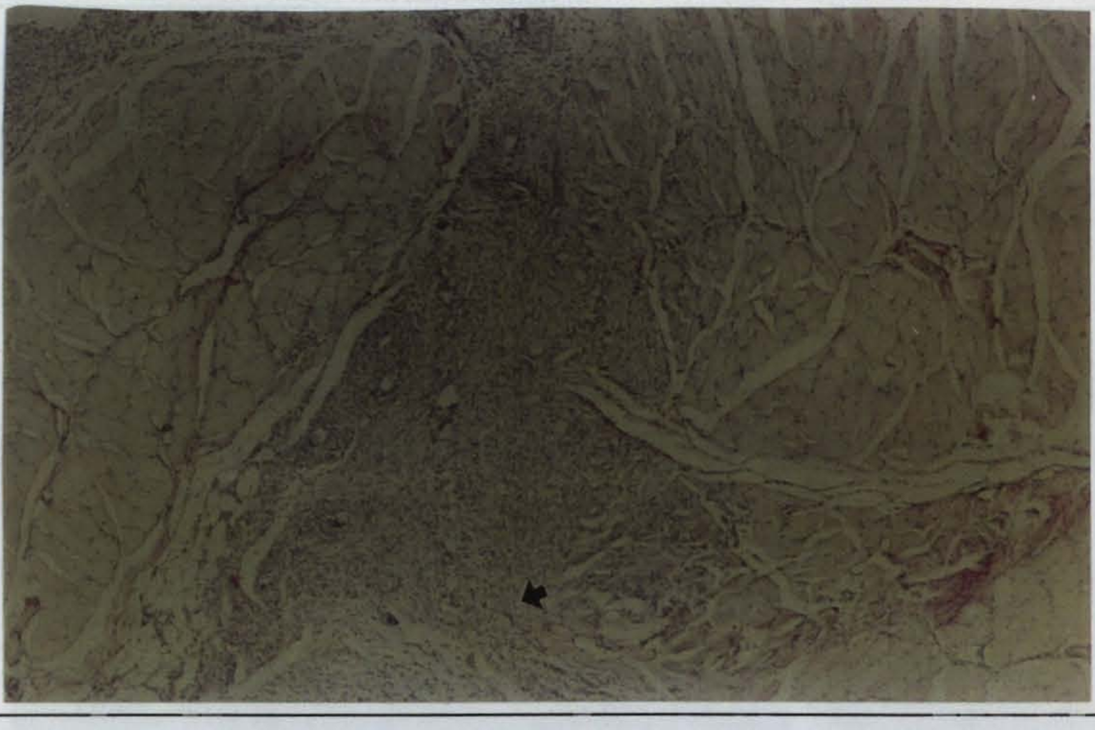


Figure 5.16 Tissue reaction surrounding the SXS#SXS knot in silk 2/0 at the 20th day. (HE X 40).

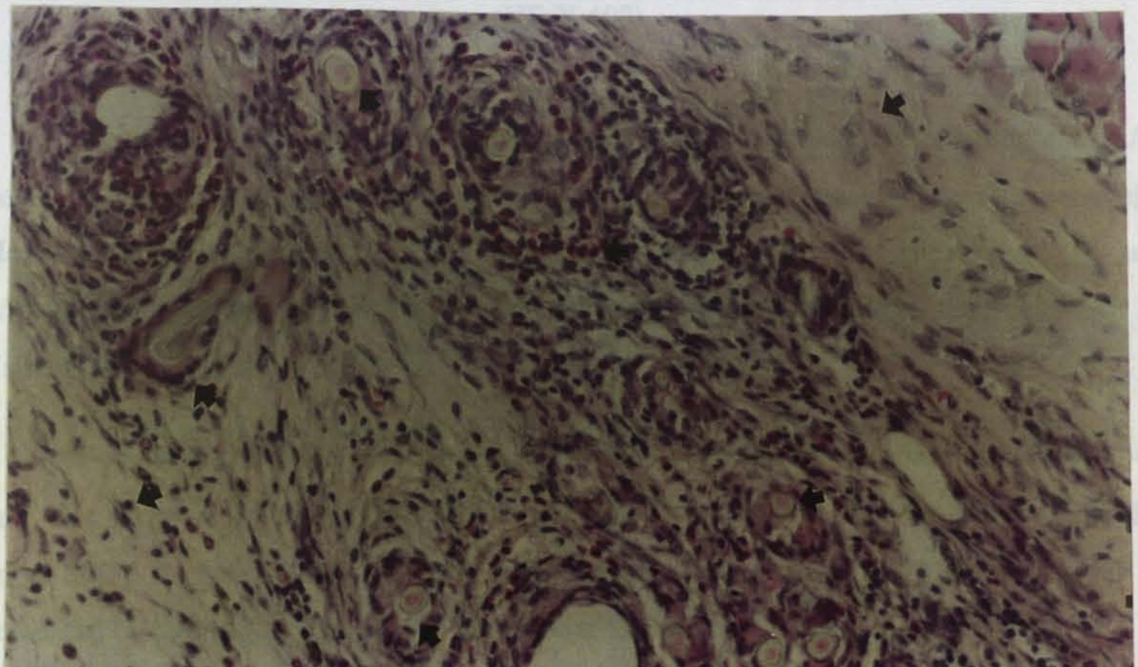


Figure 5.17 Tissue reaction surrounding the SXS#SXS nylon 2/0 at the 4th day. (HE X 400).

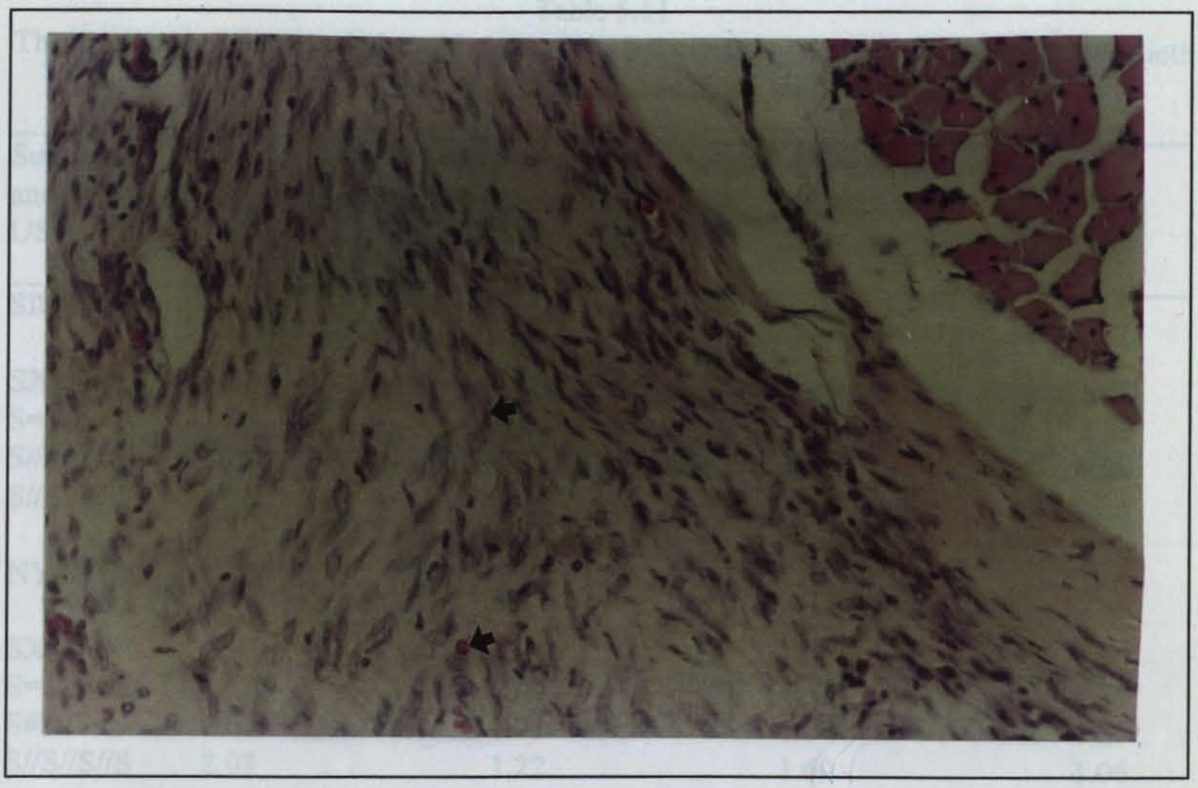


Figure 5.18 Tissue reaction surrounding the SXS#SXS knot in nylon 2/0 at the 20th day. (HE X 400).

There were no great variations in the cell density among the silk knots of 2/0; neither between the 2/0 and 4/0 knots at the fourth day and twentieth days respectively. However, the depth and width of the reaction zone varied between the knots of silk 2/0 and 4/0. At the twentieth day, the values decreased with the smaller gauge size, as shown in Table 5.11.

For the knots of 2/0, the range of the width of inflammatory zone was (1.30-1.60) mm. The knots of 4/0 silk showed a lower range (0.75-0.96) mm. As to the depth range, the knots of 2/0 recorded (1.26-1.77) while the 4/0 knots range was (1.09-1.36) mm which is presented by the height of inflammatory cylinder as shown in Table 5.11

Table 5.11
The mean experimental width and height of the inflammatory tissue cylinder at the twentieth day for the sliding knots of silk and nylon with both sizes.

Suture material and Knot code	width of inflammatory zone (mm)		height of inflammatory cylinder (mm)	
USP	2/0	4/0	2/0	4/0
SILK				
SXS#SXS	1.54	0.80	1.26	1.22
S=S//S=S	1.40	0.80	1.70	1.20
S#S#S#S	1.60	0.91	1.70	1.09
S//S//S//S	1.30	0.75	1.75	1.36
NYLON				
SXS#SXS	2.20	1.27	1.35	1.20
S=S//S=S	1.52	1.09	1.53	1.29
S#S#S#S	2.03	1.71	1.87	1.27
S//S//S//S	2.03	1.22	1.60	1.06

Similarly, as shown in Table 5.11, the 2/0 nylon knots recorded higher width and depth values than the 4/0 knots.

Table 5.12 gives the knot volume V_1, tissue cylinder volume V_2 and tissue reaction sheath volume $V_2 - V_1$ for the corresponding knots of silk and nylon of both sizes. In the dry state, all knots of silk 2/0 had similar volume (being consisted of four throws). Those of 4/0 size had the same volume. Also, this applied on nylon knots. In the dry state, nylon knots of 2/0 had higher volume values than silk knots of the same size, (3.62 ml compared to 2.28 ml). However, the difference in volume values was slighter between knots of nylon and silk of 4/0 size as shown in Table 5.12.

Table 5.12
Knot, inflammatory tissue cylinder and tissue reaction sheath volumes for the sliding knots of silk and nylon sutures with both sizes at the twentieth day postoperatively.

Suture material Knot Code	Suture size USP	Knot volume (ml)V_1	Tissue Cylinder volume (ml)V_2	Tissue reaction sheath volume (ml) $V_2 - V_1$
SILK				
SXS#SXS	2/0	2.28	2.35	0.07
S=S//S=S	2/0	2.28	2.62	0.34
S#S#S#S	2/0	2.28	3.42	1.14
S//S//S//S	2/0	2.28	2.32	0.04
SXS#SXS	4/0	0.56	0.61	0.05
S=S//S=S	4/0	0.56	0.60	0.04
S#S#S#S	4/0	0.56	0.71	0.15
S//S//S//S	4/0	0.56	0.60	0.04
NYLON				
SXS#SXS	2/0	3.62	5.13	1.50
S=S//S=S	2/0	3.62	4.59	0.97
S#S#S#S	2/0	3.62	6.06	2.44
S//S//S//S	2/0	3.62	5.18	1.56
SXS#SXS	4/0	0.77	1.60	0.83
S=S//S=S	4/0	0.77	1.23	0.46
S#S#S#S	4/0	0.77	2.88	2.11
S//S//S//S	4/0	0.77	1.25	0.48

When comparing the tissue reaction cylinder volume V_2, nylon knots demonstrated higher values than silk knots for both sizes. There was a decrease in the tissue reaction cylinder volume when the gauge size was smaller. In silk, the percent volume decrease was in a range of 62.6 to 79.2 %. Nylon showed a percent decrease range of 52.5 to 76 % as deduced from Table 5.12.

The tissue reaction sheath volumes are shown in the last column in Table 5.12. Among nylon knots, for both sizes, the alternating knots showed higher sheath volumes than the alternating with different patterns, the difference being slight for some of them.

Among silk knots, the alternating nonidentical knot showed the highest sheath volume for both sizes. Referring to Table 5.12, it is shown that the alternating nonidentical knot had the highest volume in both silk and nylon and both sizes.

5.2.4 Discussion

In clinical practice the sliding knots are considered as unreliable but, in spite of this the prevalence of their use is high due to convenience [60]. However, systematic studies on the physical qualities of the various sliding knots are still lacking. As far as the knot configuration is concerned, there is the type of sliding knot where the strand alternates using a pattern other than with every throw [28]. These are the alternating knots with different patterns. We were interested in comparing them to the alternating knots under the in vitro conditions. In this chapter, we are interested in the applicability of earlier in vitro findings under in vivo conditions. Therefore, the knot holding capacity of these knots was tested under in vivo conditions and compared to the in vitro values. Moreover, the tissue reaction to these knots was examined. The knot volume, tissue reaction volume and inflammatory sheath volume were measured accordingly.

A decrease in the KHC of all the knots was observed throughout the twenty days of implantation. Herrmann reported a decrease of 30% in strength for silk loops (three squared throws) of 2/0 size during two weeks of implantation [11]. We recorded slightly higher ranges of strength decrease for the four throwed sliding knots over twenty days of implantation.

In his study, Rijssel [49] found that the increase of suture size by two steps (2/0 versus 4/0) resulted in improved knot strength by more than 100%. The results of this study agree with Rijssel's findings.

In nylon, the knots with different patterns showed higher in vivo efficiency than the alternating ones for both sizes at the twentieth day. This superiority was also found comparing the in vitro efficiency values of these knots. For silk, the 2/0 knots showed this superiority. However, among the 4/0 knots, only the alternating parallel with different patterns showed the highest in vivo efficiency. The high efficiency of the alternating sliding with different patterns is well advantageous and encourages their use, even in place of the alternating knots. It is worth noting here that this statement applies only to the four throwed alternating sliding knots.

Even though knot slipping was commoner in vivo than in vitro, a finding that supports Tera's observation [59], only the alternating parallel was found to be unreliable under in vivo conditions. It was also found unreliable under the in vitro conditions (previously performed).

The relevance of knot slippage in clinical surgery is well important. When a suture loop surrounding a vessel unties (due to lack of security) or breaks (due to insufficient suture strength), internal hemorrhage may be the outcome. Wound dehiscence or incisional hernia may occur when a suture in the abdominal wound is disrupted [4]. Consequently, this denotes the importance of knot security, as a crucial factor as the suture loop strength in wound closure.

From another point of view, secure knots, by definition, fail by knot breakage in more than nine out of ten cases. Insecure knots fail by slippage or breakage in a 50:50 distribution. This combination is responsible for their unreliable consistency, indicating the risk of extrapolating the findings of a laboratory study to clinical practice [58]. The S//S//S//S knot showed some good KHC values in silk 2/0 at the eleventh and twentieth days: (11-16) N and (11-20.8) N respectively. These values were comparable to the KHC values of the knots who failed by breaking, the S=S//S=S for example. However, because this knot failed by slippage in some cases, it is considered as insecure. A KHC value of 20.8 N might be clinically adequate, but a value of 11 N or 2 N as recorded with nylon 4/0 (compared with the maximum value 7.8 N) might well be a disaster. Therefore, insecure knots should be avoided in clinical practice where the mechanical reliability of the suture prevails, and not only the suture strength in tissue.

One factor affecting knot security is the knot configuration. Trimbos findings indicate that knot strength is dependent not only on the type of suture material but on the type of suture knot as well [18]. Magilligan and Deweese [60], compared the knot security of four throws knots in two configurations: i)- all squared ii)- two slip and two squared with the five throws (two slip and three squared). They used 3-0 USP suture materials. For silk, they reported best knot security with the four throws squared configuration and five throws (two slip, three squared). The two slip, two squared configuration afforded the least knot security. It has been shown, when tying slip knots, adding an additional squared throw increased knot security [60]. Whether the alternation of strands and use of different patterns in the sliding knot construction is more likely to affect knot security than adding an additional squared throw to the slip or sliding knot shall be investigated.

The knot configuration element was found to be crucial in determining the KHC and the percent decrease in KHC for the two materials of both sizes. Regarding knot security, the knot type was also a factor. The alternating sliding knots with different patterns were found to be secure in both silk and nylon (no slippage). Therefore, it seems acceptable to advocate their use and compare them not only to the sliding knots but also to the currently used ones like the square and surgeon's knots.

Many studies have been done to assess different tissues reactions to various suture materials. Recently, more attention is devoted to the knot site of the surgical loop. There is more than one reason for this. Mechanically, it represents the weakest link of a suture chain, diminishing the breaking strength of the unknotted suture thread to a certain degree. Biologically, the inflammatory reaction is more pronounced at the knot site, probably because the knot represents not only the major foreign body mass of the suture loop, but also the area containing the highest density of foreign material. In addition, the knot is considered the part of the suture loop that provokes the highest degree of mechanical trauma to the surrounding tissue [49].

In addition, the importance of the knot type in clinical surgery arises from the fact that some factors should be considered when assessing the knot strength loss under in vivo conditions (besides knot security). These factors include the change in tissue-holding power that the very presence of the suture results in and also the presence or absence of tissue strangulation that the actual stitch involves [59]. Both factors cause loss of tissue holding power. In other words, not only can the strength of the suture loop be reduced by the in vivo environment but also can the suture-bearing tissue be weakened by the suture loop.

In abdominal wounds, the suture loop has to withstand the cumulative effects of : i)- intra-abdominal pressure. ii)- lateral pull of abdominal muscles. iii)- edema, inflammatory pressure within the loop itself [9]. These tensions are applied in different directions as shown in Figure 5.19 [9], imposing pressure on the suture loop. All of these tensions cause its breaking if it is not strong enough (strength is important), or its untying or slippage if it is not secure enough (knot configuration or type is important).

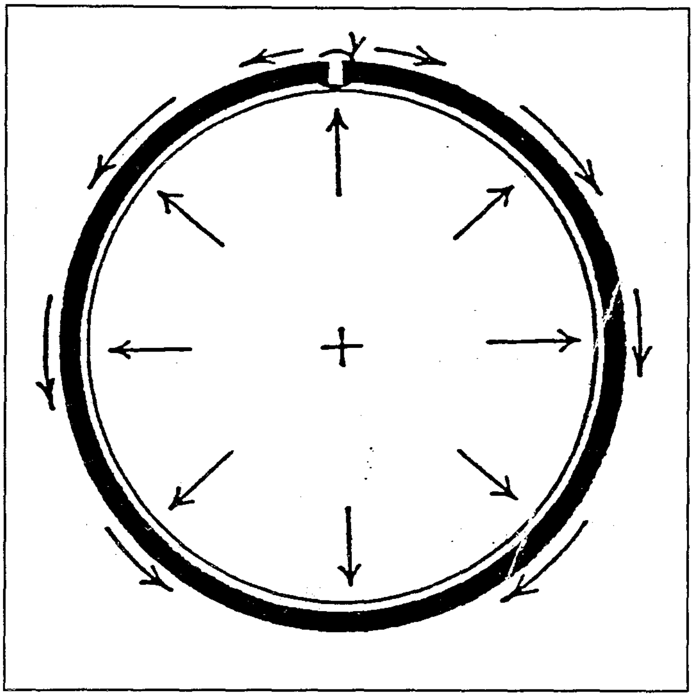


Figure 5.19 Tensions imposed on the suture loop in abdominal wall closure.

The over-all tissue reaction grades showed slight differences between the knots of 2/0 and 4/0 sizes of both materials. However, the 2/0 knots showed higher tissue reaction cylinder volume and sheath volume than the 4/0 knots. Rijssel [49], showed the suture size to be a significant independent factor in determining the score for tissue reactivity, a finding that agrees with our experimental results. The suture size is considered as an important factor since increasing the size was found to result in an increase in the knot volume by a factor of four to five. Therefore, more foreign material is introduced in the wound resulting in an increase in the sheath of the inflammatory reaction, a fact that is confirmed in this study. This conclusion is based not only on the width of the inflammatory zone but also on the height.

The factor of knot configuration in determining tissue reaction sheath volume depended well on the material used. The alternating nonidentical knot was similar in both materials, in having the highest sheath volumes for both sizes.

It was observed that the total knot body or volume for nylon was much higher than that of silk for the 2/0 size. This is because nylon is more difficult to handle than silk especially when tying a knot, with alternating axial strands. Due to its monofilament structure, this suture is relatively stiff resulting in difficulty with handling and tying [65] . Therefore, it seems more difficult to tie the nylon knot as tight as that of silk, leaving empty spaces between the alternating throws. This contributes in a significantly bigger size of the nylon knot over the silk knot (consisting of same number of throws, here four).

However, it is thought that smaller gauges of a monofilament suture have higher pliability than larger gauges because the diameter is smaller [58]. Accordingly, As the suture size decreases, the nylon becomes more supple, easier to handle, then the knot can be tightly tied in such a manner that it approximates the size of the silk knot. This was proved with the 4/0 size, where the nylon knot approximated the silk one in volume.

It could be concluded that the difference in knot volume between nylon and silk of the same size would account for the higher degree of inflammation provoked by nylon knots.

In general, the alternating knots with different patterns demonstrated better performance than the alternating ones under in vivo conditions. Obviously, depending on the intended use and ultimate function of the suture, there are some knots that will be subjected to greater tension than others. Therefore, in situations where the use of sliding knots is convenient, we recommend the alternating sliding knots with different patterns as a substitute for the alternating ones. It seems more logical to use these new configurations rather than to rely on adding additional throws to the sliding knot to enhance its security. Addition of more throws is undesirable because it introduces more foreign body to the wound, thus generating a greater amount of inflammation. Whereas the new configurations proved to be strong and secure without provoking more inflammation of the wound, since maximal wound strength requires a careful balance between maximal support and minimal foreign-body reactivity [63].

In abdominal wounds, where the risk of dehiscence exists due to the suture loop being untied [66], the knot security turns out to be as important as the knot strength. Accordingly, a careful balance should then be sought between knot-tissue strength (a very strong suture pulls out from the tissue, a weak one breaks), knot security (secure knot type or configuration that does not slip or becomes untied) and minimal tissue reaction.

Many factors have to be considered when complications occur in postoperative wounds. The most trivial wound complications comprise local hematomas, seromas, wound infection, delayed healing, incisional hernia, and extrusion of suture knots or persistent sinuses. The investigations dealing with closure of abdominal wounds demand that a suture which does not produce tissue reactions that considerably weaken the holding power of the wound, and which is able to maintain an adequate tensile strength of the thread and to make certain that the knot does not slip during the tenth to fourteenth days until the wound has regained sufficient strength. [30].

6. BACTERIAL ADHERENCE TO SURGICAL SUTURE KNOTS COMPARED TO SINGLE THREAD

6.1 Introduction

Surgical wound infections continue to be one of the most serious complications that can occur in surgical patients. Almost all postoperative wound infections are initiated along and in the vicinity of the suture lines. It has been well-demonstrated that the presence of suture material in the wound increases the susceptibility of host tissue to infection [67]. The ability of the sutured tissue to resist infection varies depending on the kind of material implanted [45, 68]. The degree of infection elicited by different sutures depends on their physical and chemical configuration [45]. The variation in the suture' capillarity and fluid absorption properties determine bacterial transport along suture filaments and correlate with the in vivo study of experimental wound infection [45].

It is well known that suture knots can hold bacteria for long periods of time and injure the surrounding tissue, thus promoting infection [47].

In abdominal wounds, the use of nonabsorbable sutures has increased, resulting in a lower incidence of dehiscence and incisional hernia but increasing the morbidity of persistent suture sinuses [46]. It is questionable here whether the knot as compared to a single thread would play a role in picking up and sheltering bacteria inside the knot thus promoting infection.

The purpose of this chapter was to examine bacterial adherence to various knot configurations compared to single strand under in vitro and in vivo conditions, and the consequent effect on tissue susceptibility to infection in rats. The importance of knots as the nidus for infection was also investigated. The bacterial adherence was examined quantitatively by a direct estimation of the number of bacterial cells upon culture under in vitro and in vivo conditions, and qualitatively by Scanning electron microscope.

6.2 Materials and Methods

Silk and nylon sutures with two sizes, were used: 2/0 and 4/0 USP. Threads of 3 cm and four different sliding knots were tested in this study. Knots were tied around two cylindrical rods attached to a board. The diameter of each rod was 1 cm and the distance between them was 8 cm to fit for implantation [17]. The knots were tied under aseptic techniques and the type of the knot was snugged down by moderate force. The knots were: the alternating sliding knots with different patterns (parallel and identical) and the alternating sliding knots (parallel and identical)

6.2.1 In Vitro Test

The knots and threads of every material and size were incubated in an overnight broth culture of S.aureus strain of $2x10^8$ /ml concentration for 30 minutes at 37° C. Then, they were soaked in a thioglycollate broth (10 ml) and shaken repeatedly for 45 minutes to wash off most of the adhered bacteria. When laid on blood agar plates, all sutures yielded only a few scattered colonies on incubation. Therefore, shaking for 45 minutes washed most of the adherent Staphylococcus from the sutures. Accordingly, 0.1 ml sample was taken from the thioglycollate broth in which the sutures were shaken, poured on blood agar plates and incubated at 37° C for 24 hours. Finally, the plates were inspected, bacterial colonies were sampled and the average bacterial count was taken for every knot and thread [35]. Comparison of the results was made using the student t-test with significance level 0.05

6.2.2 In Vivo Test

Forty Wistar rats weighing 230 g each were used for implantation. Ten rats were used for every group (2/0 silk, 2/0 nylon, 4/0 silk and 4/0 nylon) i.e. one rat for the implantation of the four knots and the single thread. Care was taken to prevent suffering so that the experiments conform to the NIH rules of animal protection. Prior to the experiment, the rats were anesthetized with an intramuscular ketamine injection (80 mg/kg). Then, the skin was undermined (midline incision) so as to create four subcutaneous pouches, two on each side. The loops were implanted into these pouches. Right at the midline and parallel to it, a thread of 3 cm was implanted. One ml of the S. aureus suspension ($2x10^8$ /ml) was injected into every knot and thread [47]. The incision was then closed with a running suture.

The rats were sacrificed four days later by ether asphyxiation. The fourth postoperative day was selected because it represents the earliest evidence of clinically apparent purulent discharge prior to the development of spontaneous drainage, suture extrusions or delayed host immune response [41]. The skin was excised to expose the suture implantation site. The local inflammatory response was assessed and classified on a scale of 0 to 2 [45]:

0- No evidence of infection.
1- Mild inflammatory response.
2- Gross purulent discharge.

After skin excision and examination of the wounds, the four knots and thread (in one rat for one material and one size) were removed. Then, they were shaken for 20 minutes in a thioglycollate broth (10 ml) from which 0.1 ml sample was taken and poured on blood agar plates which were incubated at 37° for 24 hours. Then, bacterial colonies were inspected and the average count was taken for every knot and thread of both silk and nylon and both sizes. The results were compared using the student's-test with significance level 0.05.

6.2.3 Scanning Electron Microscope Study

The suture knots and threads were incubated in an overnight broth culture of S. aureus strain of $2x10^8$ /ml concentration for 30 minutes at 37° C. They were washed and then agitated in a solution of glutaraldehyde (2.5 %) for one hour at room temperature. After washing in 0.1 M buffered sodium phosphate (pH = 7.2) and incubation in 1% osmium tetraoxide for thirty minutes, the sutures were washed again and dehydrated with increasing concentrations of ethanol and subjected to critical point drying. The sutures were then fixed to metal planchets, coated with gold and examined using a Jeol JSM-5200 Scanning electron microscope [45].

6.3 Results

Tables 6.1 and 6.2 showed the average bacterial count from silk and nylon knots and threads of 2/0 and 4/0 sizes under in vitro and in vivo conditions. From a bacterial suspension of 2x10^8 /ml, the silk suture (knot and thread) picked up about 1/10000 of the number of Staphylococci per ml of suspension. The number picked up by a nylon suture was less, about 1/100000 the number in suspension per ml. A silk suture was found to pick up 10^4 Staphylococci from a suspension of 2x10^8 per ml; a nylon suture would pick up 10^3 Staphylococci from the same suspension. A silk suture was able to pick up about 10 times as many Staphylococci as nylon.

Table 6.1
Average bacterial count (number/ml) from the silk and nylon knots and threads under in vitro conditions.

Knot Code	Silk 2/0	Silk 4/0	Nylon 2/0	Nylon 4/0
SXS#SXS	57±1	56 ±3	9.6 ±0.1	9.6±0.2
S=S//S=S	56 ±1	56 ±1	7.7 ±0.1	8.3±0.2
S#S#S#S	56±1	55±1	8.9 ±0.2	7±0.1
S//S//S//S	55±2	55±1	7.7 ±0.2	7±0.3
Thread	50±1	50±2	5.7 ±0.2	5.7±0.3

The values in the table should be multiplied by 1000.The number of bacteria is counted on blood agar plates using a standard pattern. The number of colonies is calculated per square (1 cm^2). An average of five counts is taken, then multiplied by 64 the total area in cm^2. This gives the number of bacteria per 0.1 ml. To get it per ml, it is multiplied by 10.

Table 6.2
Average bacterial count (number/ml) from the silk and nylon knots and threads after four days of implantation.

Knot Code	Silk 2/0	Silk 4/0	Nylon 2/0	Nylon 4/0
SXS#SXS	26±1	22 ±1	3.2 ±0.2	2.6±0.1
S=S//S=S	22 ±2	20 ±1	1.9 ±0.1	2.6±0.1
S#S#S#S	20±1	23±1	3.2 ±0.1	3.2±0.1
S//S//S//S	20±1	24±1	2.6 ±0.1	2.6±0.1
Thread	10±1	10±1	6400 ±10	6400±10

The values in the table should be multiplied by 1000. The number of bacteria is counted on blood agar plates using a standard pattern. The number of colonies is calculated per square (1 cm^2). An average of five counts is taken, then multiplied by 64 the total area in cm^2. This gives the number of bacteria per 0.1 ml. To get it per ml, it is multiplied by 10.

The knots of both materials picked up similar numbers of Staphylococci ; the difference being nonsignificant. It was found that the number of organisms adhered to the knot of each material exceeded that adhered to the thread by at least its half. The difference was statistically significant ($p < 0.05$). Therefore, the knot was able to pick up more Staphylococci than the thread. This was shown in Table 6.1. The knot configuration did not seem to affect bacterial adherence neither the suture size. The differences between the numbers of adhered bacteria between the knots of each material and between knots of 2/0 and 4/0 sizes were not significant. Under in vivo conditions, the number of Staphylococci that adhered to every knot and thread of both materials decreased significantly after 4 days of implantation. The inflammatory response and the infectivity scores obtained were summarized in Table 6.3. In silk, the signs of infection were redness of the skin, a marked exudate over the suture knot or thread and a pus formation after 4 days of implantation. The abscess became obvious at the fourth day. It is at this day that the rats were killed. The local inflammatory response was assessed and the pus was looked for along the suture (knot or thread). The pus was sometimes hard to find even with quite a large swelling and might be plentiful when the superficial swelling was slight. As shown in Table 6.3, in presence of silk (knots or threads), infection occurred with pus formation around suture. It means that about 10^4 Staphylococci adherent to the knot or thread of the silk suture could initiate infection with pus formation. From Table 6.3, we could notice that the number adherent to the single thread was lesser (the difference is statistically significant) than that adherent to the knots but still cause infection. The knots of both materials picked up the same number of Staphylococci in the two sizes (2/0 and 4/0). Statistical comparison of these numbers revealed no significant difference.

Table 6.3
Average infectivity scores of silk and nylon suture knots and threads.

Suture material and size	Degree of inflammatory response (Number of mice)			Infectivity score
knot code	0	1	2	
Silk 2/0				
SXS#SXS	___	4/10	6/10	16
S=S//S=S	___	4/10	6/10	16
S#S#S#S	___	4/10	6/10	16
S//S//S//S	___	5/10	5/10	15
Thread	___	6/10	4/10	12
Silk 4/0				
SXS#SXS	___	4/10	6/10	16
S=S//S=S	___	4/10	6/10	16
S#S#S#S	___	3/10	7/10	17
S//S//S//S	___	4/10	6/10	16
Thread	___	5/10	5/10	15
Nylon 2/0				
SXS#SXS	3/10	7/10	___	7
S=S//S=S	3/10	7/10	___	7
S#S#S#S	3/10	7/10	___	7
S//S//S//S	4/10	6/10	___	6
Thread	6/10	4/10	___	4
Nylon 4/0				
SXS#SXS	3/10	7/10	___	7
S=S//S=S	4/10	6/10	___	6
S#S#S#S	3/10	7/10	___	7
S//S//S//S	4/10	6/10	___	6
Thread	6/10	4/10	___	4

The nylon suture, whether in knot or thread form, failed to cause infection and pus formation. After 4 days of implantation, the local inflammatory response was restricted to a mild inflammatory reaction around the suture knot or thread. The infectivity score for nylon knots and threads was much lower than that for silk knots and threads as shown in Table 6.3.

It seemed that bacterial adherence to suture materials affects the degree of infection induced. In addition, the use of suture knot instead of thread has well increased the number of the adherent bacteria to the suture. Therefore, the degree of infection was accentuated. As shown in Table 6.3, the silk knots recorded higher infectivity scores than the thread, the nylon knots alike.

The scanning electron micrographs demonstrated the bacterial adherence to the suture threads, as shown in Figures 6.2, 6.3, in comparison to knots, Figures 6.4-6.6, corresponding to silk and nylon sutures.

Being a braided suture regarding its filament composition, as shown in Figure 6.2, the silk suture attracted more bacteria to shelter in the interlacing filaments. This increased the bacterial adherence to this suture. The knot provided more space than the thread for the bacteria to hide against the tissue phagocytic factors such as polymorphonuclears and macrophages (10 μ in diameter). This is because a knot with many throws on it retained bacteria in its interstices, safe from the humoral and cellular defenses of the body. This is shown in Figures (6.4-6.6) where Staphylococci were sheltered in between the throws of the knot. No significant difference in bacterial adherence was found between the different knot configurations. The suture size did not seem to affect the bacterial adherence to the knots and threads of both sutures.

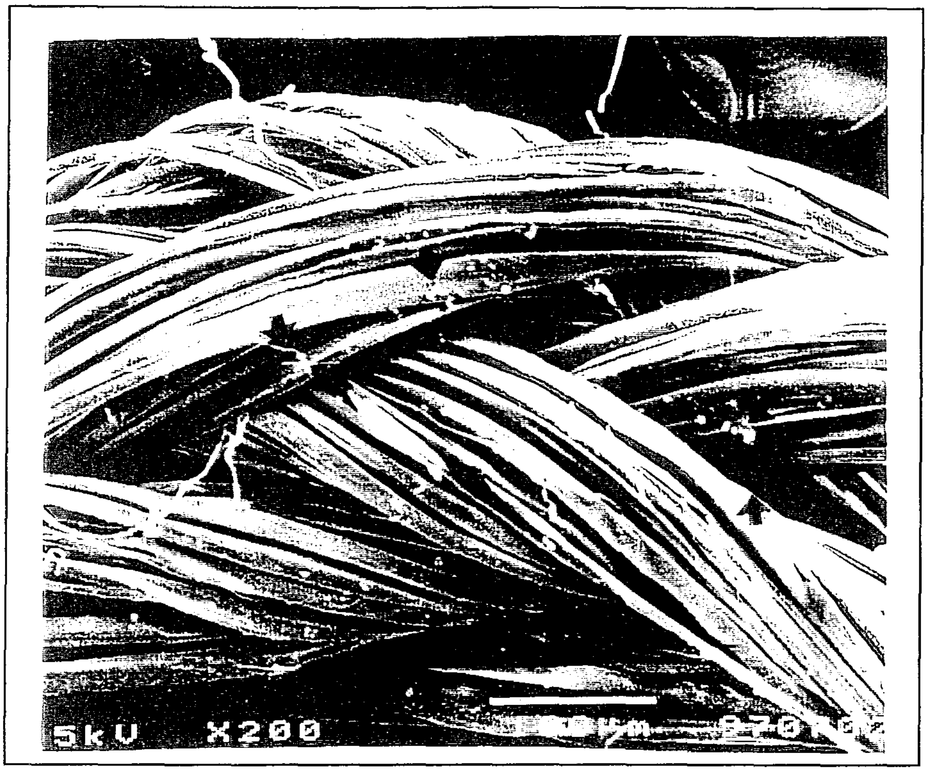


Figure 6.1 Scanning electron micrograph of a single thread of silk suture showing adherent Staphylococci.

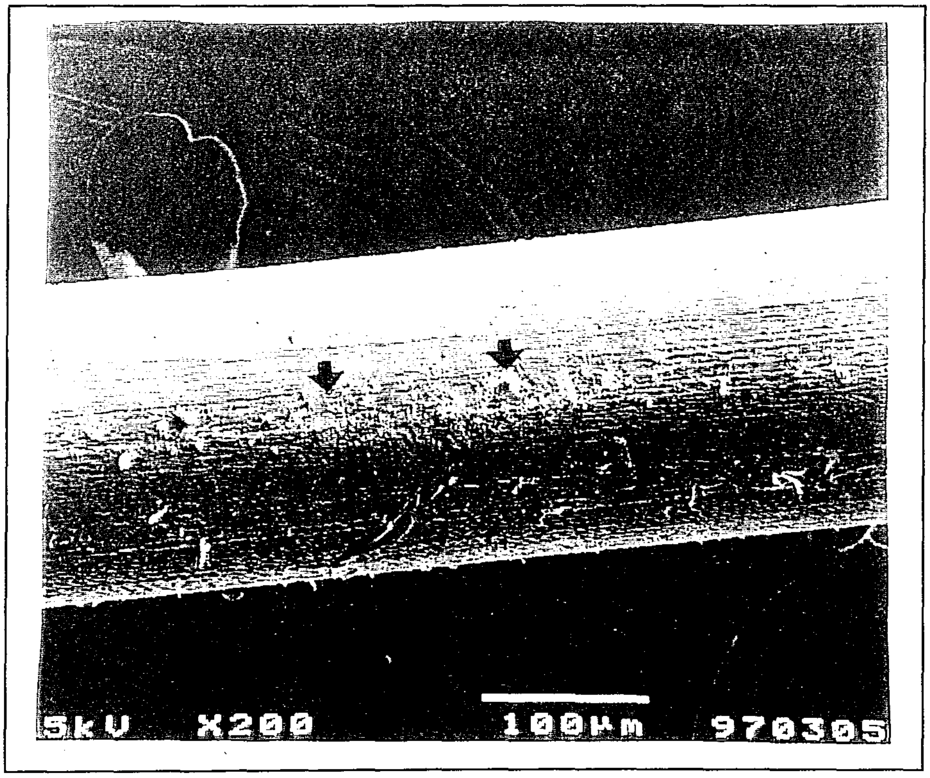


Figure 6.2 Scanning electron micrograph of a single thread of nylon with adherent staphylococci.

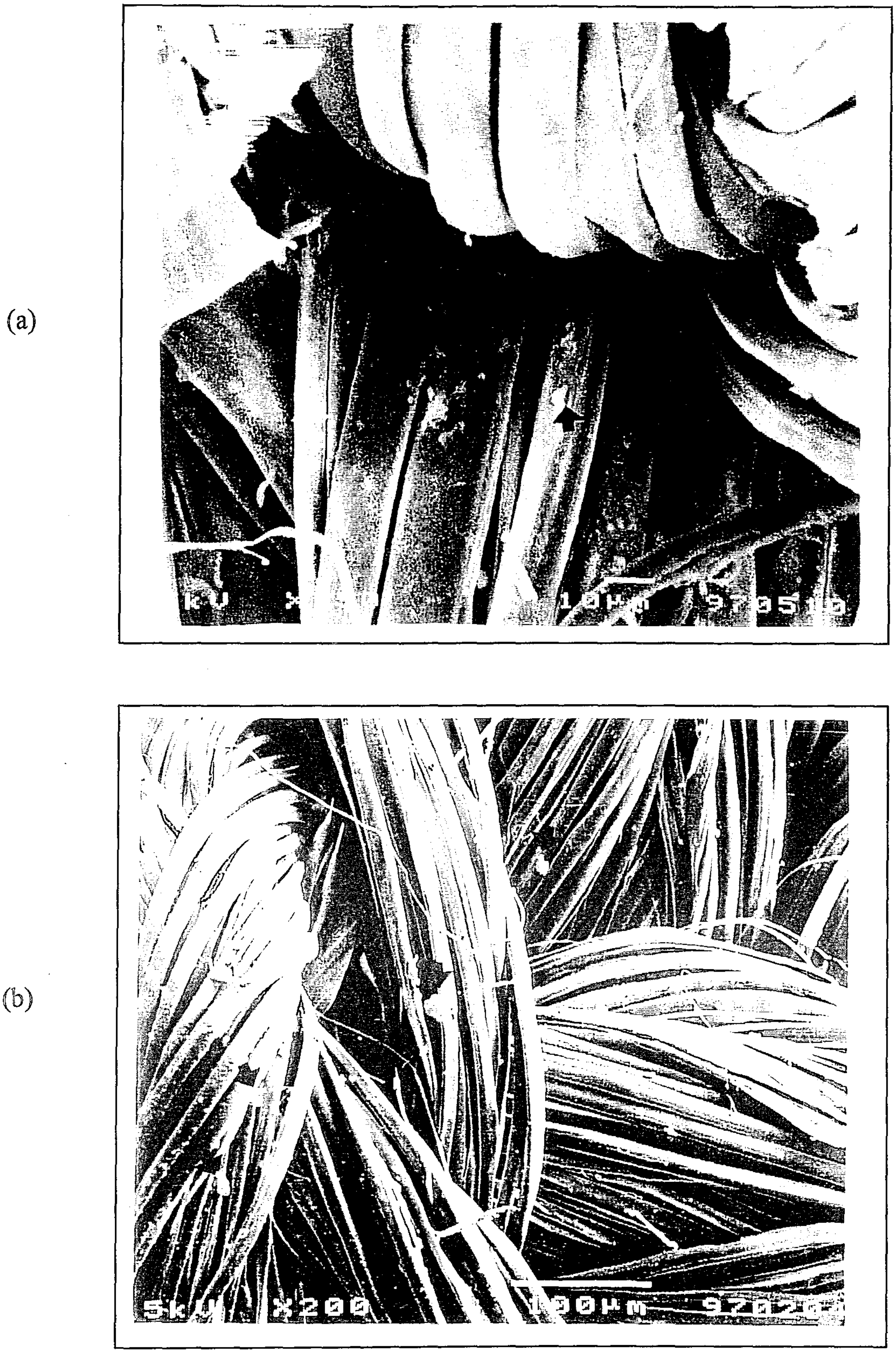


Figure 6.3 a-b Scanning electron micrographs of the silk suture knot showing adherent staph. in the interstices of throws.

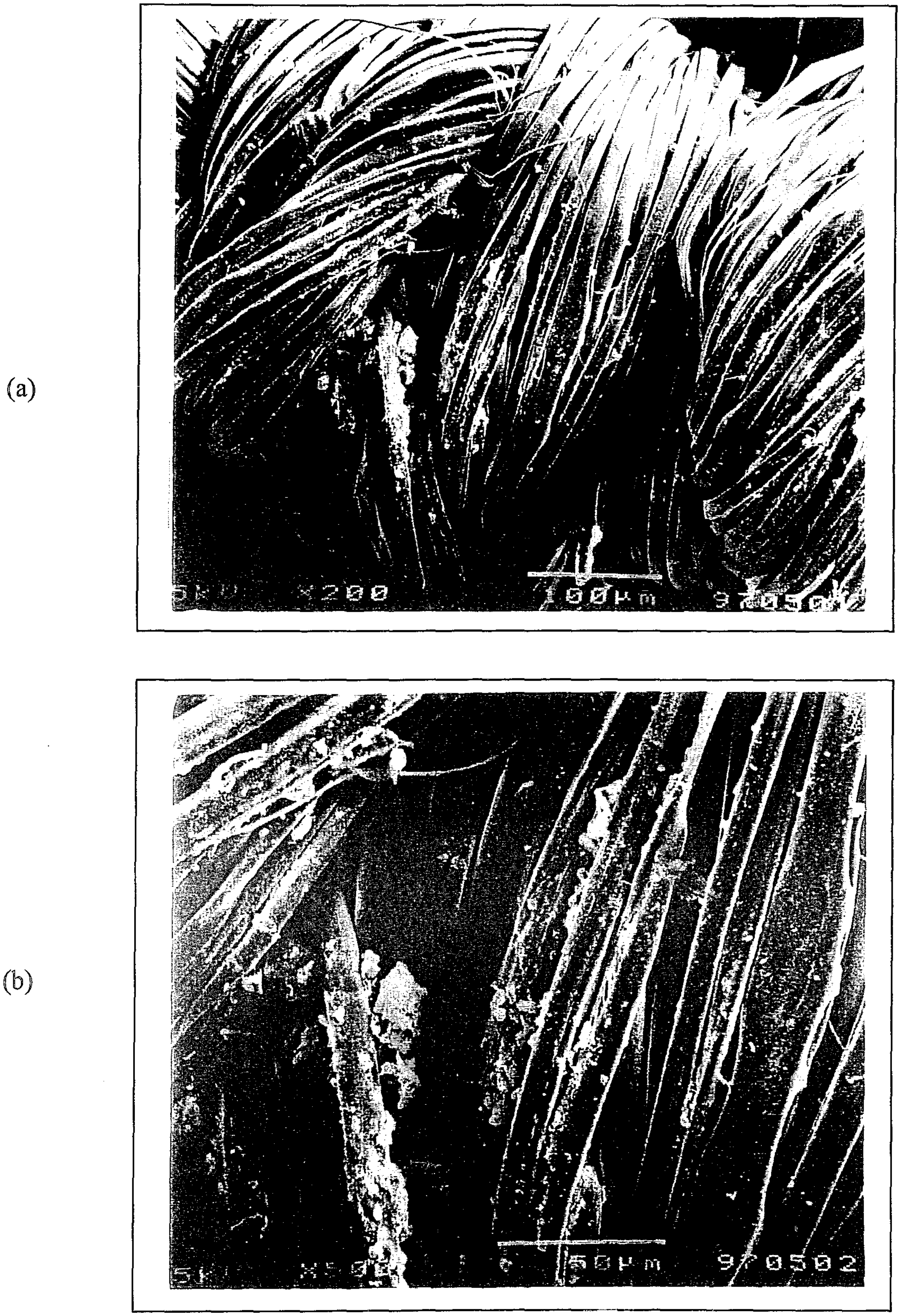


Figure 6.4 a-b More demonstrative (SEM) micrographs of the silk knot.

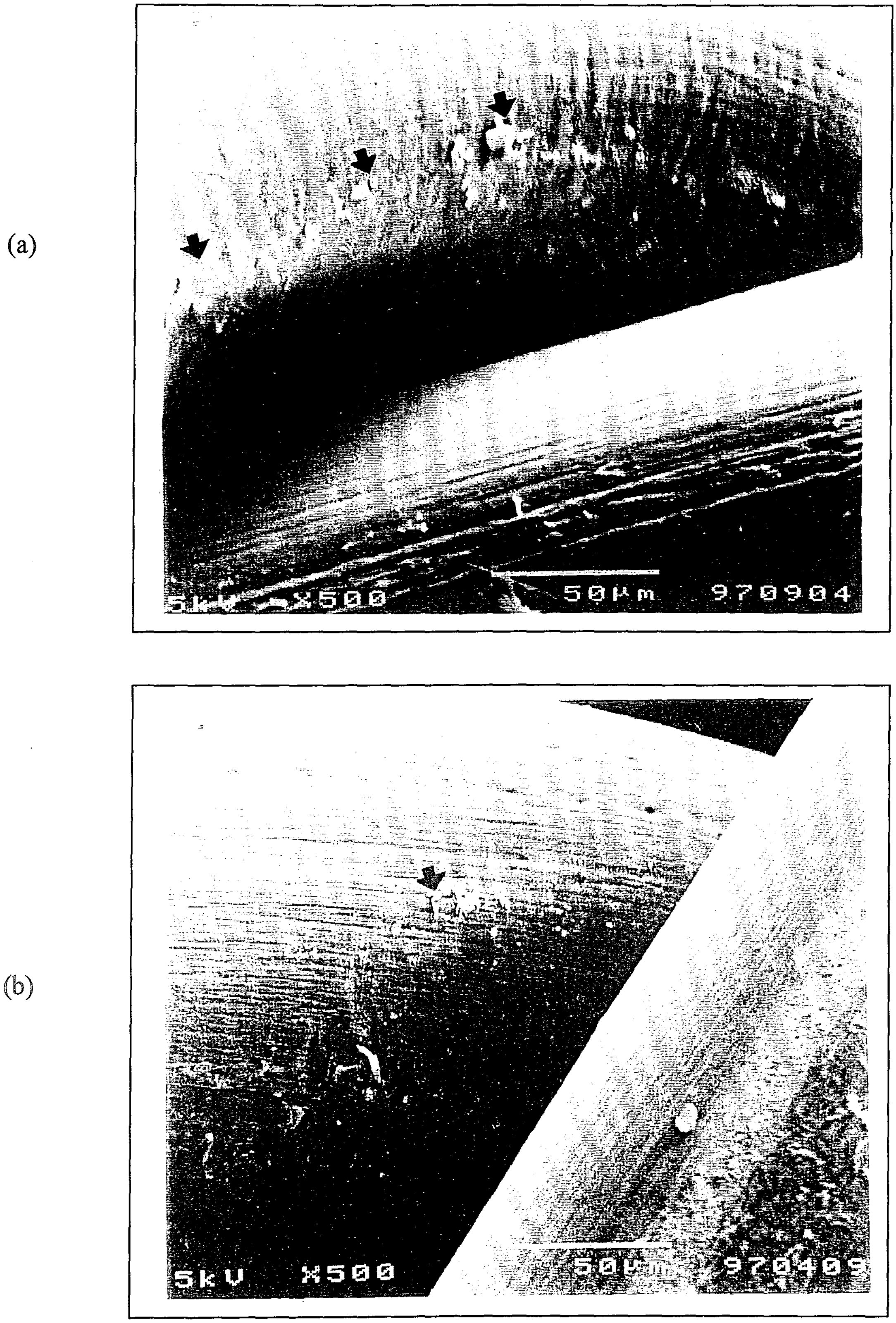


Figure 6.5 a-b Scanning electron micrographs of the nylon knot with adherent bacteria in the interstices of throws.

6.4 Discussion

It is already known that the presence of suture materials in the wound enhances the susceptibility of surrounding tissues to infection [67]. The fact that suture materials can predispose to wound infection can be explained by the formula developed by Altemeier and Culberston [69] :

$$\text{Risk of Wound Infection} = \frac{\text{Dose of Bacterial Contamination} \times \text{Virulence}}{\text{Host Resistance}}$$

The formula suggests that the inoculation of a certain number of bacteria into a wound does not necessarily result in the development of a wound infection. The key is the extent of contamination. Elek and Conen [70] demonstrated that $7.5X\ 10^6$ viable Staphylococci were normally required to induce an infection intradermally whereas as few as 300 bacteria were needed to elicit a similar infection in the presence of silk suture. We showed that as much as 10^4 Staphylococci adherent to the silk suture (thread or knot) after the fourth day of implantation, could initiate infection. In nylon, only 10^3 Staphylococci adhered to the nylon suture under the same conditions. This amount failed to elicit infection.

Bacterial adherence to suture materials has significant implications in the process of wound healing and infection. In a recent laboratory study, Katz et al. [45] noted bacterial adherence to suture materials and correlated the bacterial affinity with the degree of induced infection. They postulated that increased adherence probably lead to an increased rate of infection. In this study, our results agreed with those of Katz in that bacteria adhered to the silk suture more readily than to the nylon suture and therefore, caused more infection.

Nylon picked up less bacteria than silk under in vitro and in vivo conditions. It was not considered as an effective potentiator of infection unlike silk. Ruth [35], reported that with the more effective potentiator suture materials such as silk, introduction of approximately 10^3 coagulase positive Staphylococci caused abscess formation in almost all mice. This was also demonstrated in our study, where around 10^4 of bacteria per ml remained on the silk suture (thread or knot) after four days and this amount was enough to cause pus formation in almost all the mice. Ruth [35], stated that if the suture is retained, abscess formation takes place in a large proportion of animals given an inoculum of 10-100 Staphylococci.

Katz [45], noted a fastest bacterial removal rate with the nylon suture and a slow rate with the silk suture. Our results agreed with his, since the remaining number of bacteria, after four days, compared to that adherent to the suture (knot or thread) under in vitro, was less for nylon than for silk suture. Actually, Katz reported that almost all bacteria were removed from the nylon after implantation. We detected only very few after four days.

In this study, the main concern was to compare the bacterial adherence of the suture knots to the single threads of silk and nylon. It was found that the suture knot in both materials is more likely to retain bacteria in its interstices than the thread. This was illustrated in the scanning electron micrographs. Under in vitro conditions, the suture knot of silk and nylon picked up more bacteria than the thread. The same was true under in vivo conditions. Also, this could be due to the larger volume of the knot (consisting of four throws) compared to the thread. It is well questionable whether the bacteria is less readily removed from the knot because of its sheltering inside the interstices between the throws, away from the tissue phagocytic factors.

The importance of knots in development of infection was investigated previously. Greany [46], underwent a retrospective clinical study of suture sinuses in abdominal wounds. Twenty-six of 31 suture sinuses reviewed were associated with monofilament materials and a knot of this type of material was removed from 15 sinuses. Accordingly, he investigated the importance of the knot in the genesis of suture sinuses in an experimental study. Actually, for suture sinuses to form in abdominal wounds, bacteria must gain entry to the wound and remain locked away from the patient defenses. It is possible that the large knot, often produced provides the capillary spaces in which bacteria are safe. Therefore, it provides the nidus of infection from which wound sinus may develop.

However, his studies failed to demonstrate this hypothesis. A possibility that he mentioned in his paper but has not been investigated yet, was that perhaps, a small number of bacteria would survive in the interstices of knots. It is this possibility that we investigated in this study and showed demonstrative micrographs to support it.

It is worth noting here that the study dealt only with the early infection that a knot cause compared to the single thread. It was the aim to cast more light on the suture knot rather than the thread. Whether the knot can retain more bacteria than the thread over a considerable period causing delayed infection, should be as well investigated.

In this study, we showed that a knot is more effective than a thread in retaining adherent bacteria inside its interstices, therefore eliciting more infection in contaminated wounds in the early days of implantation.

7. ELASTICITY AND STRESS-RELAXATION OF SUTURE LOOPS IN COMPARISON TO SINGLE THREADS

7.1 Introduction

Surgeons consider the handling characteristics of a suture to be one of the most important parameters in their selection of sutures. Therefore, the mechanical performance of sutures has been the subject of numerous comprehensive studies [71]. Most of the studies have been concerned about the mechanical properties of the single thread of suture. However, in the clinical setting, the suture exists in the loop configuration. Earlier studies have been restricted to the tensile strength of knotted sutured loops. There was little concern about mechanical parameters of loops such as elasticity (expressed as the percent of recovered load upon loading and unloading) and stress relaxation (the decay of load with time expressed as the residual load fraction) as compared to the single thread of suture.

In this chapter, these two parameters were investigated in silk and nylon sutures in knotted loop configuration and compared to single thread. Standardized, reproducible tests have been developed to assess each of these parameters, providing insight into the mechanical performance of the loops of these sutures.

7.2 Materials and Methods

Silk and nylon sutures were used with 2/0 USP size. Single threads and knotted loops of four identical sliding throws are tested. Loops of the suture material were tied around two cylindrical rods attached to a board. The knots were tied under closed observation and the type of the knot was snugged tightly. The diameter of the rod was 1 cm and the distance between them was 16 cm [176]. The knots were tied by same person to minimize variability and snugged down by moderate force. The loops, were then mounted on the Zwick 1446 universal testing machine, using special clamps to hold the loop at both sides so that the knot will be placed halfway between them. The loop method was followed as it is more convenient [17].

7.2.1 Experiment 1: Elastic Properties

An estimate of the elastic properties of the knotted loop and single thread of silk and nylon sutures was determined using the American Society Testing Materials Standard Test method D1774. This method covers measurement of the elastic behavior of fibers, here suture fibers, by assessing their ability to recover strain energy, and to recover their original dimensions following a known extension.

The specimens were mounted on zwick 1446 Universal Testing Machine, they are extended to 2%, 5% and 10% of their initial gauge length (taken 50 mm for both knot and thread), respectively at a rate of 5 mm/min [65]. Then extension was stopped, and knot was left extended for a period of time: one minute. The crosshead was moved reversibly, and specimen was unloaded at same rate as the initial extension. After 3 minutes recovery period, the suture knot or thread was extended again at the same rate to 2% of initial gauge length (1 mm). An estimate of elasticity was determined by comparing the work recovered on the reverse movement with the initial work involved in the forward extension. The same procedure was repeated for the extensions to 5% an 10% corresponding to 2.5 and 5 mm respectively.

7.2.2 Experiment 2: Stress relaxation

The suture thread or knot was clamped between the jaws of the machine and the gauge length is set at 25 mm. It was loaded until a predetermined load Po was reached (taken as one third of the breaking load) [72]. Then the load was fixed and the decay was recorded over one, five and forty five minutes. Since in the surgical practice, the rate at which the knot is created is quite variable, depending on the anatomic location of the tissue being sutured, nature of the tissue , purpose of the tissue (hemostasis, wound closure etc..) and skill and experience of the surgeon, a range of strain rates was used which might be anticipated in practice. The rates selected for use in this chapter were: 6, 30, 62, 100 and 125 mm/min.

7.3 Results

7.3.1 Experiment 1: Elastic Properties

Figure 7.1 presents the elasticity, as measured by percentage work recovery, of silk and nylon sutures in thread and knot configuration respectively. The silk thread showed higher elasticity than the nylon one at 2% and 5% extension levels (62 vs 44 at 2%, 54 vs 42.8 at 5%) but lower elasticity at 10% (41 vs 52.4). As the magnitude of the extension of silk increased, its elasticity decreased as shown in Figure 7.1. This applied on both the thread and knotted loop of silk. In contrast, nylon thread and loop showed increased elasticity at higher extensions. The loops of both nylon and silk had higher elasticity than the single thread of the same material, shown in Figure 7.1. Comparing the knotted loops of both materials, the silk one had higher elasticity than nylon knot at 2% and 5% extension levels (69.6 vs 53.2 at 2%, 62.3 vs 52.7 at 5%). At 10% extension level, the silk knot showed lower elasticity than the nylon knot (43.4 vs 59.3). This is illustrated in Figure 7.1.

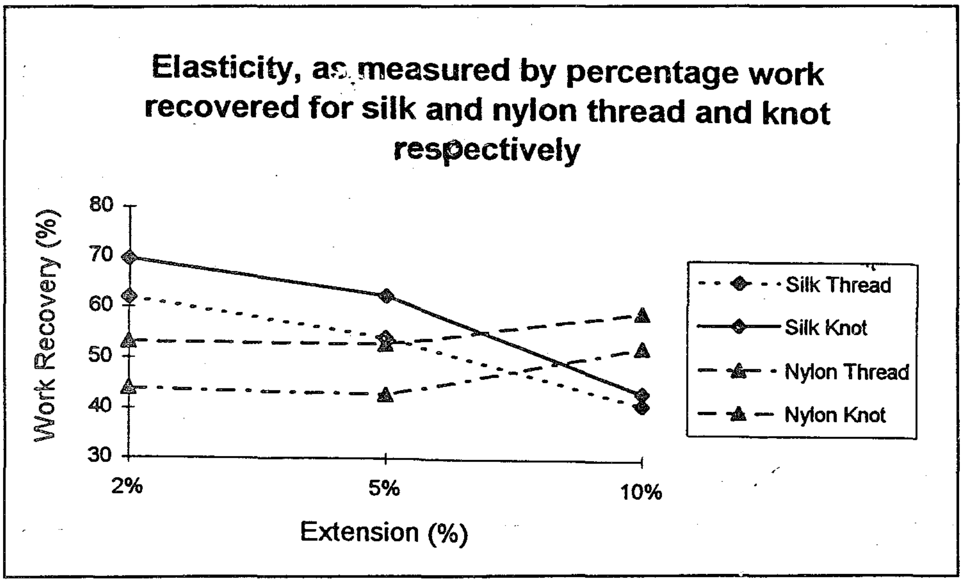


Figure 7.1 Elasticity as measured by percentage work recovered.

7.3.2 Experiment 2: Stress relaxation

The stress relaxation curve is a typical time-dependent load curve. Once the extension required to develop the target tensile load was reached (11 N for silk thread, 14 N for nylon thread and 7 N for both knots) and the jaws separation was fixed, the measured load decreased in a characteristic manner. There was initially a rapid decrement in load, followed by a more gradual decline until a steady state was reached. For purposes of comparison, the residual load fraction was defined as the ratio of residual load at time t, P(t) to the maximum or test load (Po). This parameter was found to be dependent on the rate of extension [17].

The residual load fraction dependency on extension rate for silk thread, nylon thread, silk knot and nylon knot is given in Figures (7.2-7.5) respectively. Depicted were the values at one, five and forty five minutes. At forty five minutes, an eventually steady state was reached, shown as a plateau in the load- time curve. For the silk thread it was 72%, silk knot : 75%, nylon thread: 76% and nylon knot: 78% as shown in the corresponding Figures (8.2 through 8.5).

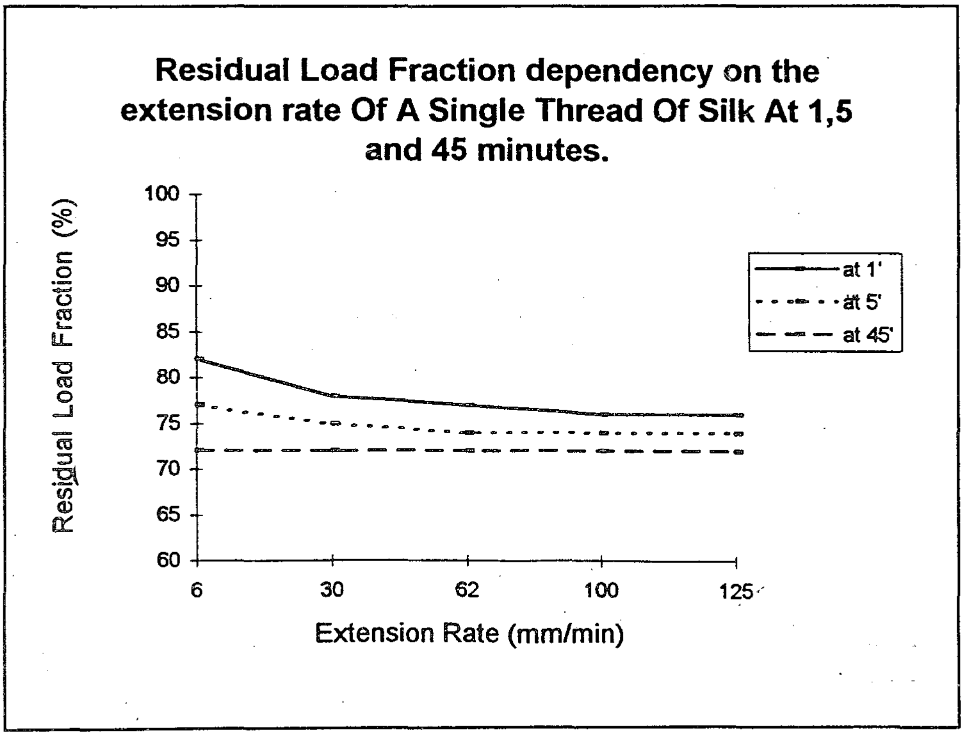


Figure 7.2 Residual load fraction as a function of extension rate for a single strand of silk.

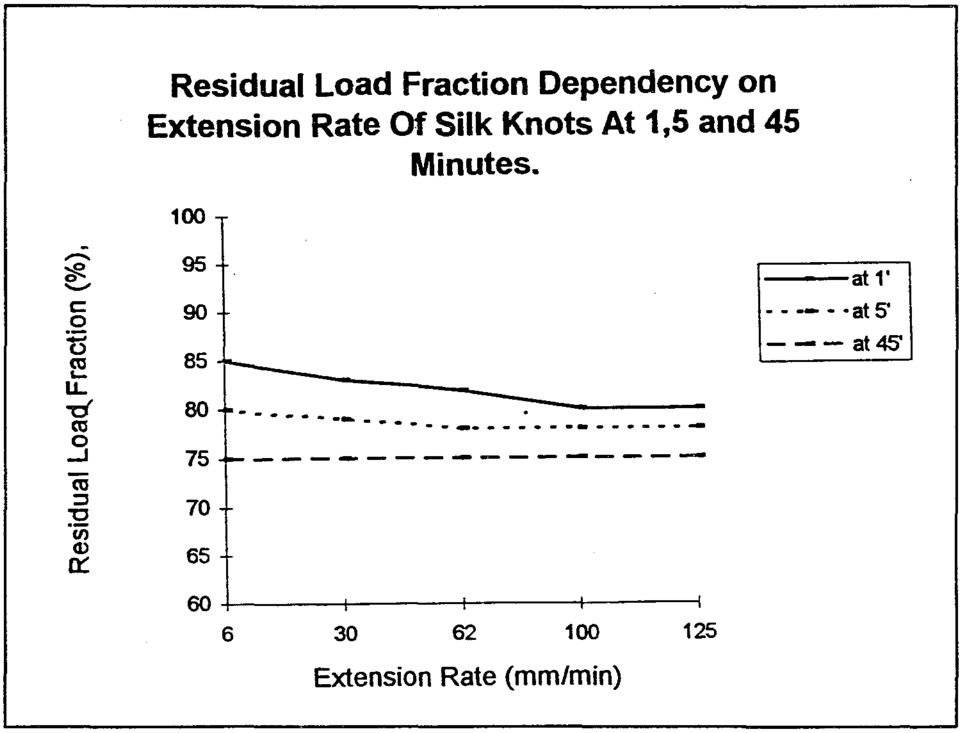


Figure 7.3 Residual load fraction as a function of extension rate for a knot of silk.

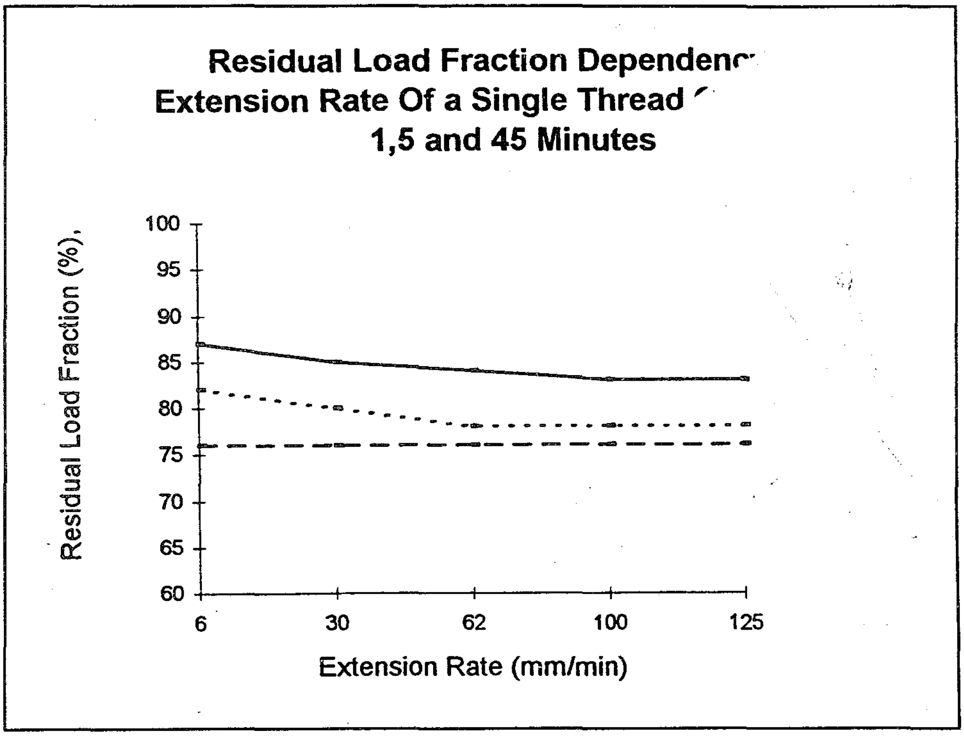


Figure 7.4 Residual load fraction as a function of extension rate for a single strand of nylon.

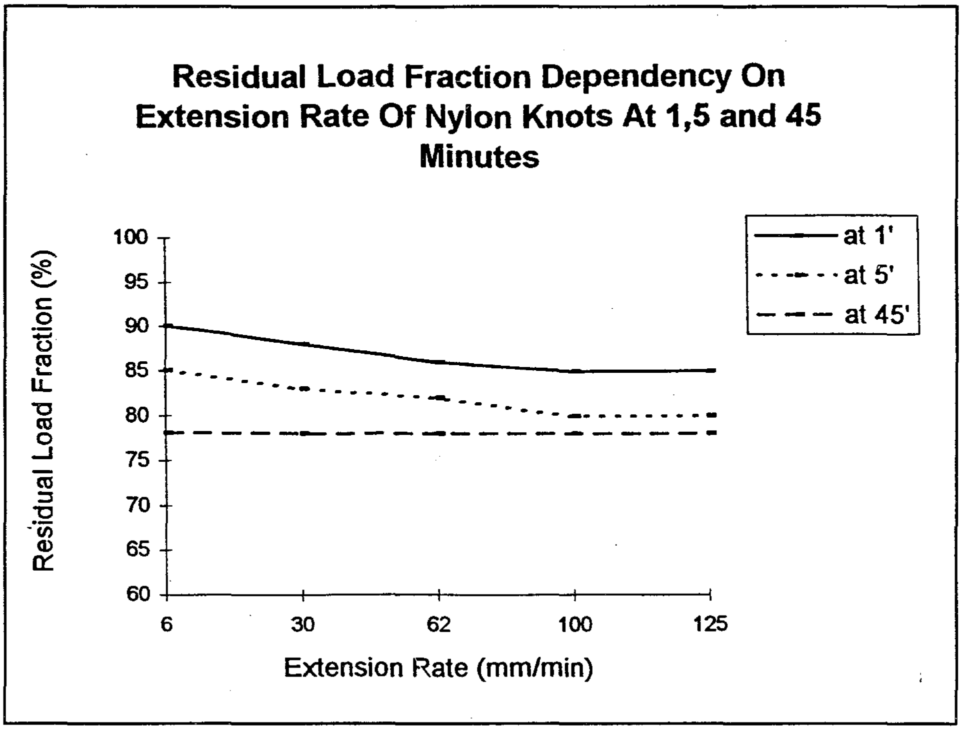


Figure 7.5 Residual load fraction as a function of extension rate for a knot of nylon.

7.4 Discussion

In abdominal wounds, the suture knot is subjected to various loads imposed at different rates. When the patient coughs, a load is suddenly imposed on the suture knot at high rate. When he breaths, slow loads are imposed at low rates. When he moves, different rates of loads are imposed according to the speed of movements.

In a vessel or artery sealing, the suture knot is subjected to the tension of blood flow inside the vessel. Emotional disorders such as fear, excitement etc., cause changes in the blood flow rate (higher rate) which can impose a higher tension on the suture knot.

These changes in tension at different rate levels may well distort the suture. Consequently, this may result in wound dehiscence in abdominal wounds and hemorrage in case of artery.

Moreover, in a clinical setting when edema occurs, it imposes tension on the suture at a slow elongation rate. Accordingly, the suture knot is extended. Upon resolution of the swelling, the suture knot is expected to return to its original length. If the suture knot is not elastic enough, it is plastically deformed and cannot resolve to the original length [65]. This leaves the wound edges in separation, and later the wound opens.

All of this reveals and emphasizes the importance of suture knot elasticity as a crucial mechanical parameter in suture material selection in every clinical application. In this chapter, we wanted to know the effect of the configuration (thread and knotted loop i.e. knot) and the extension level on the elasticity of nylon and silk sutures. It turns out that the knot configuration had higher elasticity than the single thread for both materials. As for the effect of extension level , the silk thread and knot showed decreased elasticity as the degree of extension increased, denoting permanent deformation of the suture after a certain extension level (5% of the initial length). Nylon showed increased elasticity with more extension for both the thread and the knot configuration, denoting that the deformation is elastic, and the suture loop or thread was more able to elongate at higher extension levels (10% of the initial length).

Rodeheaver [65], reported a constant elasticity of nylon at all levels of extension (2%,5% and 10%). Whereas Van Meter [71], showed increased elasticity of nylon going from the extension level 5% to 10%, he reported percentage values of 41.3 at 5% and 53.1 at 10% extension. We obtained similar results: 42.8 at 5% and 52.4 at 10% extension for nylon single thread, while the knot percentages values were higher: 52.7 at 5% and 59.3 at 10%.

The silk suture was found to have higher elasticity than nylon at low extension levels. Therefore, it should be used in wounds where small degrees of extension are required. However, when at high extension levels, the use of an elastic suture is beneficial to the wound, nylon is preferred.

Abdominal wounds tend to tear across the suture knot i.e. the tissue tear apart with the suture knot remaining untied. It is because sutures are so tightly tied that tissue edges are too stretched to come into apposition. Being under much tension, the wound edges tear apart resulting in wound opening: Dehiscence.

When a suture knot is overtightly tied across a vessel obstruction of the lumen of the vessel occurs as a result, blood flow is blocked and the vessel is strangulated. Also, over-tight approximation of skin and fascia is known to affect wound healing adversely [72]. In a wounded tissue, necrosis occurs as result of decrease in blood flow to the capillaries and arteries in that area, leading to the death of cells.

In this situation, the relaxation of stress in suture knots may be advantageous. Stress relaxation has been known in suture materials as the suture is stretched and then held at the new length, the tension gradually falls [12]. The decay of stress with time relaxes the wound tissue edges, relieving them from the excess tension, thus preventing the wound opening. In the artery, the stress decay releases the blood flow; in wounded tissues, it helps blood to reach the wounded area to nourish it. Consequently, it enhances the healing process.

In this chapter, we investigated the stress relaxation in knot and thread configurations for the two sutures. We also, demonstrated the dependency of this phenomenon on the extension rate. The extension rate influenced the tension in the suture knot during tying and while being tied. We were concerned here in the postoperative tied tension because postoperatively, suture knots are exposed to a variety of elongation rates (described previously). It was found that the residual load fraction for the two configurations used (thread and knotted loop) of the two materials was dependent on the extension rate at one and five minutes respectively. However, at forty five minutes, this parameter was nearly constant at all rates used. It has been shown that it is a material characteristic [72]. We found that it is dependent on the configuration tested since it was higher for the knot than for the thread of both materials.

In this chapter, we wanted to focus on the behavior of the knotted loop of sutures rather than the thread. Actually, it is the suture loop that is used in clinical situation. Accordingly, it is the resistance of the suture loop that is applied against imposed tension rather than the thread resistance. In abdominal wounds, for instance, the suture loop has to withstand the cumulative effects of : i) - Intra-abdominal pressure. ii) - Lateral pull of abdominal muscles. iii) - Edema, inflammatory pressure within the loop itself [9]. This is shown in Figure 7.6. These tensions are applied in different directions. However, in the single thread of suture, the load is applied unidirectionally and parallel to the fiber axis.

Moreover, the tension in the loop is proportional to the area which it encloses. The wider is the area, the lower is the bursting strength of the loop [9]. In the abdominal wound, the tensions imposed on the suture loop may well cause a considerable enlargement of the area it encloses, thus leading to its premature rupture.

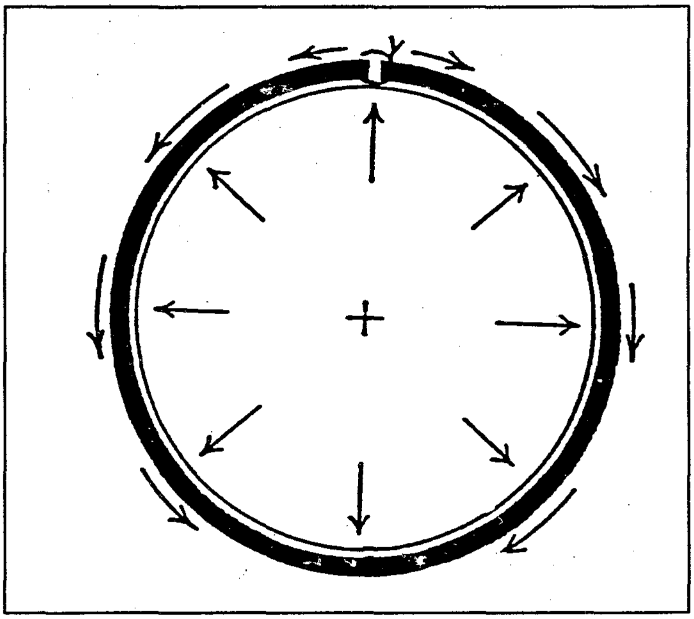


Figure 7.6 The tensions imposed on the suture loop in the abdominal wound [59].

It is concluded that the loop form should be considered instead of the thread since it is the loop that is applied in surgery. Although it is very difficult to reproduce the in vivo conditions experimentally in the laboratory, to rely on the suture loop's mechanical behavior seems closer to the real situation where many factors are considered.

8. FACTORS AFFECTING SUTURE PULLOUT FORCE IN THE RAT ABDOMINAL WALL

8.1 Introduction

Despite advances in suture technology, the perfect suture is not yet been developed and wound dehiscence continues to be an important complication of abdominal surgery. Campbell reported the incidence of dehiscence to be about 2 % of major laparatomies with an associated mortality rate of 18 % [66]. Poole reported a range from 1% to 3% incidence rates with a mortality rate ranging from 9.4 % to 43.8% [73].

As Campbell stated [66], three possible causes of dehiscence exist. The suture knot may slip, the suture may break or may tear through tissue and pullout. Generally, a surgeon can avoid knot slippage by adding throws to the knot; breaking by using a suture that must be at least as strong as the tissue. It remains the suture pullout force through the tissue which is considered as major cause of dehiscence.

Most frequently, in wounds that have separated, the sutures are found to have cut through the fascia. It is because, wound dehiscence occurs when an unexpected stress is placed on the wound edges, causing the suture to cut though the tissue. The triad of abdominal distension, coughing and vomiting (imposed at different rates) appear to be most significant causative factors of this increased abdominal pressure that leads to wound dehiscence [65].

Many other factors that affect wound dehiscence must be considered. Some of them are beyond the surgeon's control. However, the surgeon can control mechanical factors in wound closure such as suture technique, wound orientation, selection of the suture material and gauge size. This would provide a greater margin of safety against wound dehiscence.

In this chapter, the rat abdominal wall is tested for suture pullout force. Tensile testing is performed on nonsutured tissue in advance to determine the intact tissue strength. In the sutured samples, many factors are studied that would influence the suture pullout force through the tissue. Consequently, they may play a role in the problem of dehiscence. Accordingly, the questions asked are the following :

- Does the direction of the incision affect the suture pullout force from the tissue?
- Does the knot configuration and suture material play a significant role in tissue tearing through the suture knot ?
- Does the smaller diameter suture knot cut more readily than larger one?
- Is the suture more likely to cut through tissue at higher strain rate ?
- What is the effect of tissue healing strength after 7, 14 and 21 days postoperatively on the suture knot pullout force?

In an attempt to answer these questions and to cast further light on the possible factors that lead to the suture cut through the tissue, the present study was undergone.

8.2 Materials and Methods

Wistar rats and two suture materials (silk and nylon) of two USP sizes (2/0 and 4/0) were used. Prior to the experiment, the rats were killed by an overdose of ketamin HCL injection administered intramuscularly. Care was taken to prevent suffering so that the experiments conform to the rules of animal protection. The skin was excised at midline to expose the abdominal wall. The entire abdominal wall (full thickness) was included in the test specimens. The specimens were chosen according to the standard test method for tensile properties of plastics by use of microtensile specimens corresponding to ASTM D 1708-84. Vertical sections (parallel to the linea alba :1 cm from it) and transverse sections (perpendicular to the linea alba) were tested for the nonsutured specimens. This is illustrated in Figures 8.1 and 8.2. For the sutured specimens, a vertical incision through the linea alba (1 cm) is made. It is sutured with three interrupted knots of either silk or nylon using one of the two sizes:2/0, and 4/0. Suture knots were placed 3 mm apart and 5 mm from the edge of the incision (here linea alba).The corresponding specimen is cut transversely according to the standard method so that the sutured linea alba is at the middle. Another incision is made transversely, (through all abdominal layers), at 1 cm from the linea alba and perpendicular to it. It is sutured as described above, and shown in Figure 8.3. The correspondent specimen is taken longitudinally, parallel to the linea alba where the transverse incision is at the middle. To study the effect of knot configuration, three types of four throwed knots were tested: square, parallel alternating sliding and parallel alternating sliding with different patterns. They are illustrated in Figure 8.4 [28].

Figure 8.1 The specimen is cut according to testing method ASTM D 1708-84, longitudinally.

Figure 8.2 Specimen taken transversely, according to testing ASTM D 1708-84.

Figure 8.3 Example of specimens: one sutured at middle, the other is nonsutured.

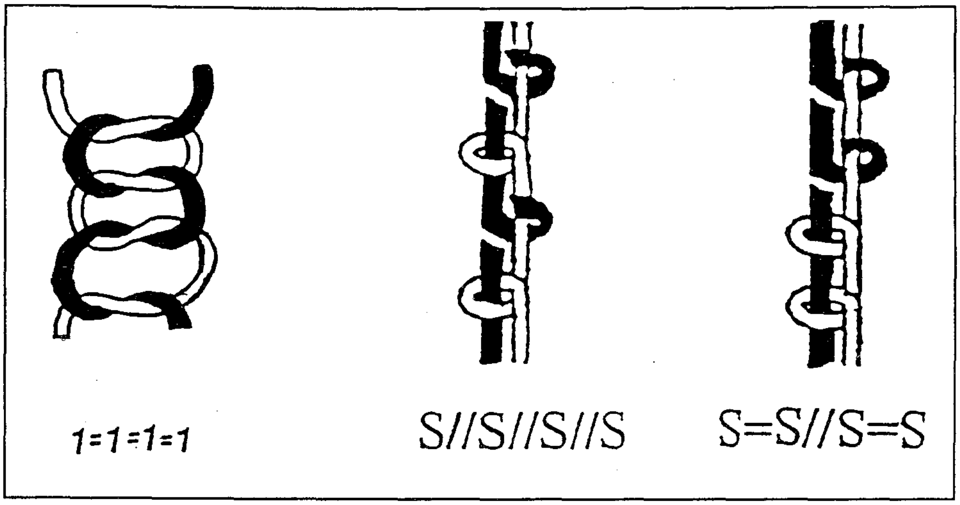


Figure 8.4 The codes and configurations of tested knots [28].

The specimens are mounted on Zwick 1446 Universal tensile machine and the load is applied gradually at 100 mm/min till rupture in the nonsutured specimens and till the tissue tears through the suture knot in the sutured specimens. Figure 8.5 demonstrates a representative suture pullout test. "Pullout force" is defined as the maximum tissue resistance beyond which tissue tearing occurs [66].

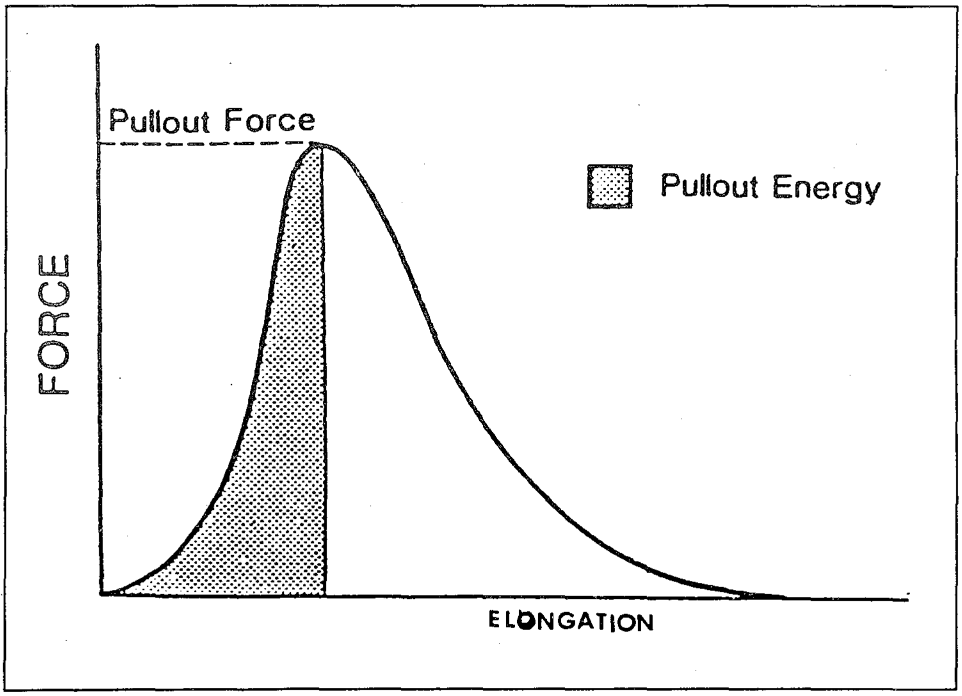


Figure 8.5 A representative suture pullout test [66].

To study the effect of strain rate on the suture pullout force, the following strain rates were used: 50, 100, 150, 200 mm/min. Finally, transverse and vertical insicions were sutured as described earlier and left till the seventh, fourteenth and twenty first days postoperatively. At the designed day, the rats were killed with an overdose of ketamin HCL injection. The specimens were taken to perform a suture pullout test on each of them as described previously. The evaluation of the results was determined using multi-way ANOVA test with significance level 0.05.

8.3 Results

Table 8.1 gives the average suture pullout force for sutured and nonsutured specimens in vertical and transverse sections. Vertical sections were stronger than the transverse ones. In the sutured specimens, the suture pullout force through the transverse incisions (vertical sections) was weaker than that of the vertical incisions.

Table 8.1
Average suture pullout force for sutured versus nonsutured specimens in vertical and transverse sections.

	Suture Pullout Force (N)	
nonsutured specimens sutured specimens	Transverse Sections (vertical incisions)	Vertical Sections (transverse incisions)
Nonsutured specimens	17.2±0.4	17.9±0.2
Sutured specimens	14.9 ±0.3	14.0±0.3

An average of 10 specimens were taken for every sutured and nonsutured section. A total of 20 specimens were tested. The results are expressed as average values with standard deviations.

As for the knot configuration element, Table 8.2 shows the results of the suture pullout force for three different knot configurations cut through longitudinal and transverse sections for silk and nylon sutures of two sizes.

Table 8.2
Average suture pullout force using different knot configurations, suture materials and sizes, in transverse and vertical incisions.

	Suture Pullout Force (N)					
	Vertical incisions			Transverse incisions		
Knot:	S//S//S//S	S=S//S=S	1=1=1=1	S//S//S//S	S=S//S=S	1=1=1=1
Suture Material and Size						
Silk 2/0	13.6±0.4	14.7 ±0.5	14 ±0.5	13.25±0.6	13.8±0.1	13.9±0.1
Silk 4/0	13.2±0.2	14.0 ±0.1	14.2 ±0.2	13.0 ±0.2	14.0 ±0.1	14.7±0.2
Nylon 2/0	13.3±0.2	14.7±0.4	14.5±0.2	13.5 ±0.3	14.3±0.1	14.0±0.2
Nylon 4/0	13.2±0.2	14.6 ±0.3	14.6±0.1	13.0 ±0.4	13.9±0.2	14.8±0.1

An average of 7 specimens was taken for every combination of knot configuration, suture material and suture size in vertical and transverse incisions respectively. A total of 168 specimens were tested. The results are expressed as average values with standard deviations.

Table 8.3 presents the results of the statistical analysis. It shows that the knot configuration, the direction of the incision are important factors in determining the suture pullout force through the rat abdominal wall. In addition, Table 8.3 shows that the suture gauge size did not affect the suture pullout force. Of critical importance, was the element of suture material (p = 0.052). It is thought this is due to the relatively small sample size (n=7). Probably, increasing the experiment number may well alter this result.

Table 8.3
Multivariate analysis of incisional direction, knot configuration, suture material and size as possible factors determining the suture pullout force in silk and nylon.

Analysis of variance	MS	DF	F-Ratio	P
Direction of incision	1.14	1	5.62	0.020
Knot configurations	13.95	2	68.80	0.000
Suture material	0.78	1	3.87	0.052
Suture size	0.02	1	0.12	0.731
Incision x knot	0.68	2	3.35	0.039
Incision x material	0.01	1	0.07	0.793
Incision x size	0.60	1	2.97	0.088
Knot x material	0.28	2	1.41	0.249
Knot x size	2.03	2	9.99	0.000
Material x size	0.01	1	0.03	0.856
Knot x incision x material	0.23	2	1.13	0.327
knot x incision x size	0.38	2	1.88	0.159
Incision x material x size	0.57	1	2.83	0.096
Knot x material x size	0.01	2	0.04	0.963
Incision x knot x material x size	0.28	2	1.40	0.252

MS = Mean -square; DF = degree of freedom, P = significance. N.B. =The corresponding data sheet is given in Appendix C.

Table 8.4 presents the suture pullout force of silk and nylon sutures, in two gauge sizes (2/0 and 4/0) at different postoperative days: 7, 14, and 21. The suture pullout force differed between the postoperative days, for the two materials and sizes. It increased significantly from the seventh to the fourteenth till the twenty first day.

A similar increase was observed for both the vertical and transverse incisions. This increase in suture pullout force means an increase in the wound strength throughout the healing period reaching its maximum at the twenty first day: around 80 % of the intact tissue tensile strength. Table 8.5 gives the results of the statistical analysis. The postoperative days element is well a factor affecting the suture pullout force, a fact that agrees with our hypothesis. The suture material and size as well as the incision direction were not important factors.

Table 8.4

Average suture pullout force of transverse and vertical incisions sutured with silk and nylon of 2/0 and 4/0 sizes at different postoperative days

	Suture Pullout Force (N)					
	Vertical incisions			Transverse incisions		
Days	7	14	21	7	14	21
suture material and size						
silk 2/0	11.0 ±0.2	13.0±0.2	14.1±0.1	10.7±0.1	13.0±0.1	14.3±0.1
silk 4/0	10.8±0.1	12.5±0.2	14.2 ±0.4	11.0±0.2	13.4±0.1	15.0±0.2
nylon 2/0	10.9±0.4	12.7±0.4	14.4±0.2	11.0±0.3	13.5±0.2	15.5±0.1
nylon 4/0	10.8 ±0.1	12.8±0.1	14.0±0.1	10.8±0.1	12.1±0.1	14.0 ±0.3

Seven specimens were tested for every suture material and size at each of the postoperative days. A total of 168 specimens were tested. Average values with standard deviations are presented.

Table 8.5

Multivariate analysis of incisional direction, postoperative day, suture material and size as possible factors determining the suture pullout force in silk and nylon.

Analysis of variance	MS	DF	F-Ratio	P
Postoperative days	143.2	2	40.52	0.000
Direction of Incision	1.58	1	0.44	0.505
Suture material	0.67	1	0.19	0.663
Suture size	0.001	1	0.00	0.988
Days x Incision	0.52	2	0.15	0.863
Days x material	4.31	2	1.22	0.301
Days x size	5.42	2	1.53	0.222
Incision x material	0.04	1	0.01	0.911
Incision x size	0.09	1	0.03	0.870
Material x size	7.96	1	2.25	0.137
Days x incision x material	0.21	2	0.06	0.943
Days x incision x size	0.14	2	0.04	0.962
Days x material x size	2.93	2	0.83	0.440
Incision x material x size	3.98	1	1.13	0.292
Days x incision x material x size	0.44	2	0.12	0.883

MS = Mean -square; DF = degree of freedom, P = significance. N.B.= the data sheet is given in Appendix D.

The effect of strain rate on the suture pullout force is shown in Figure 8.6. The statistical analysis is given in Table 8.6. The strain rate unlike the suture material and size was a contributing factor in determining the suture pullout force, as illustrated in Table 8.6. As the strain rate increased, the suture pullout force increased significantly, for both the transverse and vertical incisions. At 50 mm/min, the suture pullout force was nearly 10.5 N, at 100 mm/min: 14 N, 200 mm/min: 15.5 N and at 300 mm/min, it was about 17 N as shown in Figure 8.6.

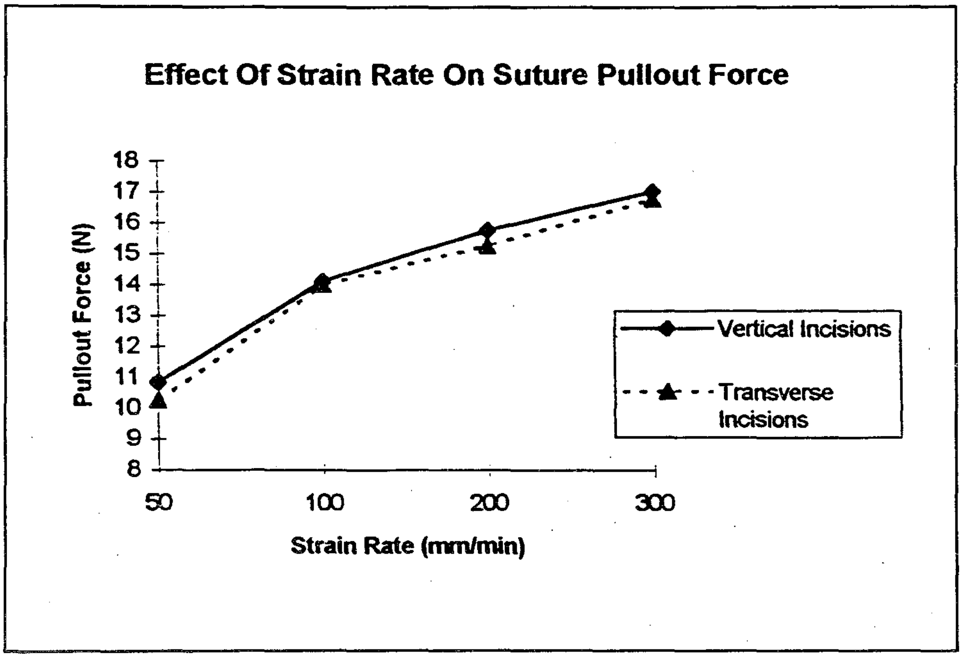


Figure 8.6 The effect of strain rate on Suture Pullout force.

Table 8.6
Multivariate analysis of strain rate, suture material and size as possible factors determining the suture pullout force in silk and nylon.

Analysis of variance	MS	DF	F-Ratio	P
Suture material	0.37	1	0.21	0.650
Suture size	0.013	1	0.01	0.933
Strain rate	138.1	3	76.9	0.000
Material x size	0.013	1	0.01	0.933
Material x Strain rate	0.69	3	0.39	0.763
Size x Strain rate	0.31	3	0.17	0.916
Material x size x strain	0.36	3	0.20	0.894

MS = Mean -square; DF = degree of freedom, P = significance. The data sheet is given in Appendix E.

8.4 Discussion

The most common reason for wound rupture is the sutures cutting through the tissue while the knots remain intact [64]. This has been noted in as many as 88% of dehisced wounds. In his study, Sanders, [64], showed that it was the cause in 81% of 11 clinical dehiscences. When the suture tear through the tissue, either the tissue is too weak to hold the tissue, or the technique of suturing is inadequate. However, weak tissues are rarely the cause. Therefore, the problem must lie in the method of closure [64].

Several other factors lead to dehiscence and are beyond the surgeon's control. In this chapter, we are interested in the mechanical factors that are the responsibility of the surgeon, like the incision direction, knot configuration, suture material and gauge size. In addition, the tissue healing strength and strain rate effects are studied. The suture pullout model is a powerful tool to supplement clinical data because the event of dehiscence can be simulated a large number of times under controlled conditions [66].

Transverse abdominal incisions were thought, at one time, to be more resistant to dehiscence than vertical ones, because they are under less tension [74]. However, some experimental studies have failed to support this view. They demonstrated no significance in wound strength between vertical and transverse incisions [64, 75]. In this chapter, we found that vertical incisions were stronger. The fact is that the techniques of suturing vary considerably, so that one cannot conclude that the choice of incision is the cause of dehiscence. Actually, when the techniques of closure were standardized experimentally, transverse incisions showed no superiority over vertical ones [76,77]. Even though, there are still some studies that advocate the use of transverse incisions over the vertical ones. Greenall [78], assumed that the relative strength of the transverse incisions is explained by the greater suture holding capacity of the musculo-aponeurotic layer when incised transversely. Tera and Aberg [79], showed similar results to Greenall.

Sutures whose knots slip are always a potential hazard, particularly with monofilament sutures of nylon. Slip or sliding knots with monofilament materials are more apt to untie than with silk [64]. However, properly tied square knots, with each throw of suture snugged into the throw below it, will hold as well with monofilaments as with silk

[64]. In this chapter, we used three different knot configurations: simple alternating sliding, alternating sliding with different patterns both belonging to the category of sliding or slip knots and a square knot. No one knot became untied in nylon and silk as well, denoting equal security of the sliding and square knots. However, not all the knots teared through the tissue at similar pullout force values. This led us to conclude that the knot configuration is a possible factor in suture pullout force, therefore in dehiscence. However, Rodeheaver [65], reported that knot slippage or breakage was never a cause of wound dehiscence.

The sliding knots performed well as the square knot and did not untie under increasing tensions. This applied to the nylon and the silk suture of two USP sizes. It is worth noting here, that all the knots were four-throwed indicating that they are secure. It appears that these knots have close holding capacities but the alternating sliding knot with different patterns had better performance than the simple sliding one and similar to the square knot in terms of knot holding capacity.

However, tying the knot could lead to dehiscence when too tight tying would cause strangulation of tissue with ischemic necrosis [73]. Also, the tying technique is an important factor but is not investigated in this study.

Moreover, the use of silk as a multifilament and nylon as a monofilament suture did not seem to affect the suture pullout force. The influence of the suture material on the pullout force seems to be related to the elasticity of the suture material. Rodeheaver [65], stated that the greater the ability of the suture to expand with the abdomen, the smaller the amount of stress that is transmitted to the tissue. In the suture pullout test, the silk and nylon sutures had good elasticity to allow them to expand while the tissue elongates to reach its yield point. Beyond that point, the tissue teared through the suture knot. The element of suture material was a significant factor in the comparison of the knots, but was not significant when comparing the suture pullout force throughout the postoperative days, also when comparing the strain rates. Whether it is really a contributing factor should be further investigated.

The gauge size (2/0 and 4/0), was not a factor in determining the suture pullout force. Campbell [66], has made the same conclusion using a range of gauges from 00 to 2. However, Dudley [80], proposed that fine suture would cut tissue easily, likening it like a knife edge with high pressure per unit area of contact. His hypothesis was not supported in this study.

Under in vivo conditions, the suture knot in the abdominal wound, is subjected to various loads. These loads are imposed at different rates. When the patient coughs or vomits, a load is suddenly imposed at a high rate. When he breaths, a low load is imposed at a low rate. Since in surgical practice, the rate to which the knot is subjected is quite variable, a range of strain rates was used to simulate the in vivo conditions and study its effect on the suture pullout force. As a viscoelastic material, the rat abdominal tissue cuts through the suture at a higher load when the strain rate is increased. This is the behavior of any viscoelastic material subjected to increased strain rates. It is ductile at low strain rates but becomes brittle at high strain rate [81]. This results in a decrease in the amount of energy absorbed to the maximal point, here the maximum tissue resistance. It is beyond this maximum value that the tissue tears through the suture knot. Therefore, the pullout force is influenced by the rate at which the tissue is pulled.

To simulate the in vivo conditions, the sutured wounds were tested for suture pullout force at different specific postoperative days. The healing properties of the abdominal tissue were considered as an important factor. It is during the period of four to six days that the strength of the wound must be reinforced by the use of sutures. In this chapter, the strength of the abdominal wounds at the seventh day were found to be 60% of that of the unwounded wound for both vertical and transverse incisions. During this period, the suture pullout force was low. As the fibroplasia phase began, the strength of the wound increased slowly to reach around 75% of the tensile strength of the unwounded tissue at the fourteenth day. The maximal value was found at the twenty first day (80%). It is at the period of fourteen days that the use of sutures is no more needed. This conclusion agrees with that of Adamson and Enquist [82], who stated that in healing abdominal wall wounds in rabbits, the useful role of suture material in contributing to the tensile strength of the wound is well completed by fourteen days. However, Lichtenstein [83], disagreed with this conclusion.

As a conclusion, the suture pullout test may be used to quantify the resistance of the abdominal tissue against the pulling force of the sutures. Therefore, it could be a measure of the security of the sutural closure technique.

In this chapter, the influence of some factors on the suture pullout force was evaluated. The relevance to the problem of abdominal dehiscence is discussed. However, it is worth saying that still other factors which role is important as well such as suture bite size, fascial thickness, method of closure, needle type etc..

9. CONCLUSIONS AND RECOMMENDATIONS

Although surgeons warn against the sliding knots, the use of these knots in clinical practice is well prevalent due to their convenience [17], [58]. Recently, there is the type of alternating sliding knots with different patterns. This type has not been studied yet [28]. It ought to be introduced to surgeons in the clinical practice. Therefore, testing the properties of these knots would seem encouraging to compare them to the currently used sliding knots (simple and alternating).

A knot of a suture is the weakest point where the suture is most likely to fail by rupture or slippage. It is also, the site where most of the foreign material is present, therefore, the site of the most pronounced tissue reaction. Besides, it represents the shelter where bacteria hide away from the tissue phagocytic factors. Accordingly, to have an ideal suture knot performance, the surgeon must compromise between maximum support (suture knot strength or knot holding capacity), maximum knot security, minimum tissue reaction, less or absence of infection and good handling properties (extensibility, elasticity, stress relaxation etc.) that are beneficial to the desired medical application, and not affected by the in vivo environment of the wound.

It is on this basis that the sliding knots with different patterns were tested. It turned out that they have a good knot holding capacity even if it was comparable to other sliding knots but better efficiency, and less KHC decrease throughout twenty days of implantation in rats. The knot configuration was a factor in determining the percent decrease in KHC and knot security of silk and nylon sutures of 2/0 and 4/0 sizes. They did not evoke more tissue reaction than the sliding alternating knots. However, being a knot, bacteria was able to shelter in the interstices of the throws. They did not differ in bacterial adherence from the other knots.

As the problem of dehiscence persists in abdominal surgery, most studies point out to the suture pullout through the tissue as the main cause [66]. Accordingly, this study aimed to test the effect of the knot configuration (using these new knots) in priority along with other elements' effects on the suture pullout force. Therefore, a model was created using the rat abdominal wall. It was concluded that the knot configuration was one among other factors affecting the suture pullout force. The sliding alternating knot with different patterns was less likely to pull out from the rat abdominal wall than the alternating knot but similar to the square knot.

The study attempted to investigate some knotting properties that are considered important in the clinical theater. The elasticity and stress relaxation are one of the properties that are beneficial to the wound situation. Knotting was found to enhance these properties in comparison to the single suture threads.

As a conclusion from these experiments, in situations where the use of sliding knots is convenient the use of the alternating sliding knots with different patterns to substitute for the alternating and simple sliding knots is well recommended. As the sliding knot performance can achieve that of a square knot by adding one more throw, it seems more advantageous to use the new knots configurations instead of adding a throw to a sliding knot and risk the wound inflammatory response. Therefore, the performance of these knots should also be compared to other knots categories such as the square, and surgeon's knots.

APPENDIX A
KNOT HOLDING CAPACITY

A typical data sheet of the knot holding capacity of silk and nylon knots of size **2/0** over postoperative days as designed for statistical analysis by multivariate ANOVA test using SYSTAT (the possible factors were the knot configuration, postoperative day and suture material).

KHC (N)	Knots	Days	Material
17.5	1	1	1
21.0	1	1	1
20.0	1	1	1
18.0	1	1	1
18.6	1	1	1
17.2	1	1	1
18.0	1	1	1
17.0	1	1	2
16.5	1	1	2
16.75	1	1	2
15.5	1	1	2
16.7	1	1	2
16.0	1	1	2
19.2	1	2	1
19.4	1	2	1
20.0	1	2	1
18.3	1	2	1
20.3	1	2	1
16.0	1	2	1
16.0	1	2	1
16.0	1	2	2
17.5	1	2	2
17.0	1	2	2
16.5	1	2	2
15.0	1	2	2
15.0	1	2	2
20.0	1	3	1
13.2	1	3	1
15.6	1	3	1
20.5	1	3	1
19.0	1	3	1
12.5	1	3	1
12.0	1	3	1
13.7	1	3	2
15.0	1	3	2
15.5	1	3	2
15.0	1	3	2
14.0	1	3	2
14.9	1	3	2
18.6	1	4	1
15.3	1	4	1
14.5	1	4	1

APPENDIX B
PERCENTAGE DECREASE IN KNOT HOLDING CAPACITY

A typical data sheet of percentage decrease in knot holding capacity of silk and nylon knot over postoperative days as designed for statistical analysis by multivariate ANOVA test using SYSTAT (the possible factors were the knot configuration, suture material and size).

% decrease in KHC	Knot	Material	Size
02.10	1	1	1
19.47	1	1	1
23.68	1	1	1
18.15	1	1	1
27.63	1	1	1
32.89	1	1	1
01.84	1	1	1
17.64	1	2	1
17.65	1	2	1
20.00	1	2	1
20.58	1	2	1
18.23	1	2	1
11.76	1	2	1
25.26	1	1	2
26.31	1	1	2
36.84	1	1	2
21.05	1	1	2
44.21	1	1	2
47.36	1	1	2
28.57	1	2	2
28.57	1	2	2
38.57	1	2	2
35.71	1	2	2
41.42	1	2	2
42.85	1	2	2
57.14	1	2	2
31.81	2	1	1
48.63	2	1	1
30.45	2	1	1
43.18	2	1	1
45.45	2	1	1
26.81	2	1	1
40.00	2	1	1
16.66	2	2	1

APPENDIX C
SUTURE PULLOUT FORCE (SPF)

A typical data sheet of the suture pullout force values as designed for statistical analysis by multivariate ANOVA test using SYSTAT (the possible factors were incisional direction, knot configuration, suture material and suture size).

SPF (N)	Incision	Knot	Material	Size
13.5	1	1	1	1
13.2	1	1	1	1
13.0	1	1	1	1
15.0	1	1	1	1
13.4	1	1	1	1
14.9	1	2	1	1
15.0	1	2	1	1
14.5	1	2	1	1
14.2	1	2	1	1
14.8	1	2	1	1
13.5	1	3	1	1
13.9	1	3	1	1
14.5	1	3	1	1
14.0	1	3	1	1
14.0	1	3	1	1
13.5	2	1	1	1
13.5	2	1	1	1
13.6	2	1	1	1
12.7	2	1	1	1
13.0	2	1	1	1
13.5	2	2	1	1
14.0	2	2	1	1
14.0	2	2	1	1
13.7	2	2	1	1
13.8	2	2	1	1
13.5	2	3	1	1
14.0	2	3	1	1
14.5	2	3	1	1
13.8	2	3	1	1
13.5	2	3	1	1
13.2	1	1	1	2
13.0	1	1	1	2
13.0	1	1	1	2
13.5	1	1	1	2
13.4	1	1	1	2
13.5	1	2	1	2

APPENDIX D
POSTOPERATIVE SUTURE PULLOUT FORCE (SPF)

A typical data sheet of suture pullout force as designed for statistical analysis by multivariate ANOVA test using SYSTAT (the possible factors were incisional direction, postoperative days, suture material and size).

SPF (N)	Incision	Material	Size	Days
07.0	1	1	1	1
11.0	1	1	1	1
10.5	1	1	1	1
12.0	1	1	1	1
10.5	1	1	1	1
04.0	1	1	1	2
12.0	1	1	1	2
12.5	1	1	1	2
13.4	1	1	1	2
14.0	1	1	1	2
21.0	1	1	1	3
14.5	1	1	1	3
14.5	1	1	1	3
14.0	1	1	1	3
13.5	1	1	1	3
07.0	2	1	1	1
10.5	2	1	1	1
10.2	2	1	1	1
10.0	2	1	1	1
12.0	2	1	1	1
04.0	2	1	1	2
12.0	2	1	1	2
12.5	2	1	1	2
13.5	2	1	1	2
14.0	2	1	1	2
21.0	2	1	1	3
15.0	2	1	1	3
14.5	2	1	1	3
13.6	2	1	1	3
14.0	2	1	1	3
11.0	1	1	2	1
11.5	1	1	2	1
10.7	1	1	2	1
10.0	1	1	2	1
12.0	1	1	2	2
12.5	1	1	2	2
14.5	1	1	2	2

APPENDIX E
EFFECT OF STRAIN ON SUTURE PULLOUT FORCE (SPF)

A typical data sheet of suture pullout force as designed for statistical analysis by multivariate ANOVA test using SYSTAT (the possible factors were strain rate, suture material and size).

SPF (N)	Material	Size	Strain
10.8	1	1	1
08.8	1	1	1
12.6	1	1	1
11.2	1	1	1
14.0	1	1	2
13.0	1	1	2
14.0	1	1	2
15.5	1	1	2
16.0	1	1	3
16.0	1	1	3
15.0	1	1	3
16.0	1	1	3
17.0	1	1	4
18.0	1	1	4
17.0	1	1	4
16.0	1	1	4
10.5	1	2	1
09.0	1	2	1
12.0	1	2	1
11.0	1	2	1
15.0	1	2	2
13.0	1	2	2
12.0	1	2	2
15.0	1	2	2
16.0	1	2	3
17.0	1	2	3
17.0	1	2	3
15.5	1	2	3
17.0	1	2	4
19.0	1	2	4
16.0	1	2	4
15.0	1	2	4
11.0	2	1	1
12.0	2	1	1
10.0	2	1	1
8.0	2	1	1
16.0	2	1	2
14.0	2	1	2
13.0	2	1	2
12.0	2	1	2
18.0	2	1	3
16.0	2	1	3

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The work presented in this dissertation began in September 1993 and was completed in March 1998.It was supported by Boğaziçi University Research Fund. Two papers out of this work are accepted for publication in the *Archives of Surgery* and the *Journal of the American College of Surgeons.*