2005 IUGA GRAFTS ROUNDTABLE

International Urogynecological Association: The Usage of Grafts in Pelvic Reconstructive Surgery Symposium 2005
July 8–10, 2005, Lago Mar Resort, Fort Lauderdale, FL, USA

Published online: 16 May 2006
© International Urogynecology Journal 2006

Roundtable participants
Host: G. Willy Davila, M.D.

Gopal Badlani, M.D.
Thomas A. Barbolt, Ph.D.
Mauro Cervigni, M.D.
Raymond Connolly, Ph.D.
Jan Deprest, MD Ph.D.
Harold P. Drutz, M.D.
Peter Dwyer, M.B.B.S., M.D.
Gamal Ghoniem, M.D.
Michael Hiles, Ph.D.
Bernard Jacquetin, M.D.
Dennis Miller, M.D.
Pamela Moalli, M.D., Ph.D.
Paulo Palma, M.D.
Paul Riss, M.D.
George T. Rodeheaver, M.D., Ph.D.

Harry Vervest, M.D.
Hans VanGeelen, M.D.
Mark Walters, M.D.
J Christian Winters, M.D.

Other guests

Daniel Biller, M.D.
Marjorie Jean-Michel, M.D.
Carolyn Langford, D.O.
Szylvia Udvari-Nagy, M.D.
Ed Koullik
Keith Modert
Rodney Bosley

Acknowledgements Special appreciation to Szylvia Udvari-Nagy for her tireless efforts in assisting with the preparation of the manuscripts in conjunction with the participant authors. Thanks also to Kristin Dunn for her assistance with the planning and organization of a successful meeting.

This meeting was made possible through educational grants from the following companies: AMS, Mentor, Cook, Gynecare, USSurgical/Tyco
Table of Contents

G. Willy Davila, M.D.

Introduction

G. Willy Davila, MD, FACOG is Chairman of the Department of Gynecology and Head of Urogynecology and Reconstructive Pelvic Surgery at Cleveland Clinic Florida in Weston, Florida. He also serves as Director of the Fellowship training program in Urogynecology and Reconstructive Pelvic Surgery.

Harold P. Drutz, M.D.

Pelvic Organ Prolapse: Demographics and future growth prospects

Dr. Harold Drutz is Professor of Obstetrics and Gynecology, and Head of the Division of Urogynecology and Reconstructive Surgery (URPS) in the Department of Obstetrics and Gynecology at the University of Toronto. He is Chief of the Urogynecology and Urodynamic Investigative Units at Mount Sinai Hospital and Baycrest Geriatric Centre, and is the Director of the University of Toronto Postgraduate Fellowship Training Program in Urogynecology and Reconstructive Pelvic Surgery (URPS).

Peter Dwyer, M.B.B.S., M.D.

Evolution of biological and synthetic grafts in reconstructive pelvic surgery

Peter Dwyer MBBS, FRCOG, FRANZCOG, CU, is Director of the Urogynaecology Department at the Mercy Hospital for Women, Melbourne, Australia and is Associate Professor of the Department of Obstetrics and Gynaecology Mercy Hospital and University of Melbourne. He was President of the International Urogynaecological Association (2002 to 2004) and is a past President of the Continence Foundation of Australia.

Jan Deprest, M.D., Ph.D.

The biology behind fascial defects and the use of implants in Pelvic Organ Prolapse repair

Born 1960, MD 1985, Dr Deprest has an MD (1985), a Special Licence in Obstet Gynecol (1991), PhD (1999), and is a Professor in Obstetrics and Gynaecology at the KU Leuven, Belgium. He is consultant in University Hospitals Leuven, with subspecialties in Fetal Medicine and Urogynaecology. He is Director of the Centre for Surgical Technologies, Research and Training Center in Surgical Techniques and is the General Project Manager of Eurofoetus and EuroTwin2Twin projects of the European Commission.

Thomas A. Barbolt, Ph.D.

Biology of Polypropylene/Polyglactin 910 Grafts

Thomas (Tom) Barbolt has a Ph.D. in Experimental Pathology and Toxicology from Albany Medical College, NY. Tom is a Senior Research Fellow in the Corporate Product Characterization group at ETHICON. Tom was instrumental in demonstrating the safety of PRONOVA suture, a then new non-absorbable material, and the safety of VICRYL Plus Antibacterial Suture, the first regulatory clearance for the internal use of triclosan, the active agent. He was active in the due diligence assessments of Oxiplex AP anti-adhesion gel and OmNex Vascular Sealant, two important growth devices that have recently joined the ETHICON portfolio of products.

George T. Rodeheaver, M.D., Ph.D.

Surgipro* Mesh: Not all Multifilaments are the same

Dr. Rodeheaver has been the Director of Plastic Surgery Research at the University of Virginia Medical Center since 1972. He is presently the Richard F. Edlich Professor of Biomedical Research. His primary research focus is the management of injured tissue. Using standardized animal models, he evaluates wound management techniques such as debridement, wound cleansing, bacterial control, dressings, and closure techniques.

J. Christian Winters, M.D.

InteXen™ tissue processing and laboratory study

A native of New Orleans, Dr. Winters is a Clinical Associate Professor in Urology at LSU and is the Residency Program Director for the LSU/Ochsner urology residency training program. He also holds the position of Associate Chairman, Department of Urology and Director of Urodynamics & Female Urology at the Ochsner Clinic Foundation in New Orleans, Louisiana.

Michael Hiles, Ph.D.

Tissue Engineering a Clinically Useful Extracellular Matrix Biomaterial

Michael Hiles is the Vice President for Research and Clinical Affairs at Cook Biotech Incorporated—a medical device firm specializing in the development of extracellular
matrix (ECM) technologies for implantable and topical medical devices. He holds an appointment as Adjunct Professor of Veterinary Clinical Sciences at the Purdue University School of Veterinary Medicine. Mike received his BS and MS degrees in Electrical Engineering from Purdue and his Ph.D. in Veterinary Physiology and Pharmacology from the Veterinary Medical School at Purdue.

Raymond Connolly, Ph.D.

**Evaluation of a Unique Bovine Collagen Matrix for Soft Tissue Repair and Reinforcement**

Dr. Connolly received his PhD from the University of Pennsylvania in 1969. After a postdoctoral fellowship at Brookhaven National Laboratory, he joined the Scientific and Special Staff at Tufts-New England Medical Center in Boston. He is currently an Associate Professor of Surgery and is the Director of the Surgical Research Laboratory at T-NEMC. Among his research interests is the testing and evaluation of materials for medical applications.

Pamela Moalli, M.D., Ph.D.

**Cadaveric Fascia Lata**

Dr. Moalli is an Assistant Professor at the Magee-Womens Hospital/Magee-Womens Research Institute, University of Pittsburgh. She also works as Director of Female Athletic Medicine for the University of Pittsburgh and University of Pittsburgh, Department of Athletics, and is the Director of Basic Science Laboratory Training for female Pelvic Medicine Fellowship.

G. Willy Davila, M.D., Harold Drutz, MD, and Jan Deprest, MD, PhD

**Clinical Implications of the Biology of Grafts: Conclusions of the 2005 IUGA Grafts Roundtable**
Implanted grafts are being used with increasing frequency by pelvic reconstructive surgeons. In addition, the variety of grafts available for use has expanded quite rapidly. In fact, there has been an exceedingly rapid progression from the concept of using the graft for recurrent prolapse cases to that of using grafts in primary therapy procedures. A cursory review of abstracts recently presented at professional meetings reveals a growing number of case series reporting on use of grafts in reconstructive pelvic surgery. More importantly, only a minute number report on a comparative trial between a grafted vs a non-grafted technique. It is, thus, assumed that reconstructive surgeons are implanting grafts in their reconstructive procedures with increasing frequency, despite the lack of controlled trials documenting the value of a graft in reconstructive procedures. In fact, a review of the published scientific literature to date reveals only a small percentage of papers that meet scientific criteria for a randomized prospective trial.

Manufacturers of surgical implants have been prompt to develop and modify grafts for use in reconstructive surgery. The rate of development and marketing of new grafts has certainly exceeded the rate of scientific analysis and scrutiny of presently used grafts. Structural and composition changes in grafts have been based on theoretical benefits rather than scientifically demonstrated improved outcomes. However, industry is not solely to blame for the presumption that they are “driving the cart.” Many modifications to existing graft materials are based on surgeons’ requests. It is, thus, increasingly clear that the “ideal graft” has not yet been identified.

Except in very specific situations, the benefits of implanting a graft have not been proven. The implantation of a graft in reconstructive pelvic surgery is not risk-free. Experienced surgeons have witnessed complications associated with the use of grafts. These have included problems with infection, rejection, and erosion of various graft materials. As a rule, these materials have been released for marketing before sufficient data are collected regarding the risks and benefits of the particular graft’s utilization. Any experienced surgeon can enumerate a list of materials used in the recent past that were eventually noted to be associated with well-described and recognized adverse outcomes.

There is currently little science behind the choice of a material for reconstructive procedures. This is true from a clinically-evidenced outcomes basis as well as from a basic science research basis before initial clinical use. In an ideal scenario, a graft would be developed in the laboratory, tested on animals, be subjected to monitored clinical trials, be compared to non-grafted procedures in randomized trials, and be subsequently compared to other materials in randomized prospective trials. Unfortunately, this process, although highly desirable, is likely impractical due to time and cost constraints.

The International Urogynecological Association (IUGA) is uniquely positioned to be able to help expand knowledge regarding particular aspects of urogynecologic surgery. At the 2004 annual meeting, a decision was made to pursue a series of mid year symposia to discuss aspects of particular interest to the membership. The symposia would then result in a supplement to the International Urogynecology Journal such that the presentation, discussions, and conclusions could be communicated to the IUGA membership and pelvic surgeons in general. It was decided that this initial symposium would focus on the basic science of currently available graft materials. The overall format of the symposium was conceptualized such that basic science researchers would present pre-clinical data on currently available graft materials to a group of experienced reconstructive pelvic surgeons. This basic science data could then be translated into clinically useful information, which could then be used to help design clinical trials. In its simplest form, the concept was to gather basic science researchers and pelvic surgeons in one setting to enhance...
communication and create an open forum for sharing of information regarding pre-clinical testing as well as early clinical experience.

The goals for this graft symposium were:

1. To understand the current status and role of grafts in reconstructive pelvic surgery
2. To review current knowledge regarding the biology of currently marketed synthetic and biologic grafts for pelvic reconstructive surgery
3. To discuss currently available literature on grafts’ usage for pelvic surgery
4. To develop proposals for future clinical and laboratory studies to further improve our knowledge regarding graft selection for reconstructive pelvic surgery

The basic science presenters were identified via a literature review as well as input from reconstructive surgeons and manufacturers of graft materials. Thus, some were employees of industry, some were academic consultants to industry, and others were surgeons who also ran a basic science lab within their academic institution.

Not all currently available graft materials were represented at the symposium. An attempt was made to include a basic scientist knowledgeable in the development of each available biologic and synthetic graft material. Presenters were identified and invited to present data on the most commonly utilized synthetic and biologic materials. Not all were able to attend.

The costs of this symposium were underwritten by unrestricted educational grants to IUGA from various companies manufacturing and marketing graft materials. IUGA was responsible for all the expenses including those of industry employees who presented at this symposium.

It is the hope of IUGA that those who read the following manuscripts will find the included information useful and that they will be able to apply it to their clinical practice. In addition, it is hoped that the use of graft materials in reconstructive pelvic surgery will be approached with a greater degree of scientific scrutiny and that more randomized trials will be undertaken to attain a better understanding of the potential benefits and drawbacks of the usage of a graft in reconstructive pelvic surgery.
Abstract Pelvic Organ Prolapse (POP) is the hidden epidemic. Demographic studies have shown that women over the age of eighty are the fastest growing population segment in the United States and Canada. Over the next thirty years the rate of women who will seek treatment for POP will double. Risks for the development of POP have been categorized into factors that predispose, incite, promote, and decompensate. Connective tissue disorders may play a role in the pathogenesis which may involve a reduction in total collagen content secondary to increased collagenolytic activity. Eventually clinicians may be able to identify women who may be genetically predetermined to develop POP. The role of adjuvant materials in performing reconstructive pelvic surgery may improve success rates, but evidence based medicine and randomized controlled trials are currently lacking.

Keywords Pelvic organ prolapse · Prevalence · Risk factors · Materials

Pelvic organ prolapse is the protrusion of pelvic organs into or out of the vaginal canal. The first surgical treatment for this problem was introduced by Soranus of Ephesus (AD 98–138), who was considered the foremost gynecologic authority of antiquity. He proposed vaginal hysterectomy for uterine prolapse in 120 AD [1]. Since then, pelvic surgeons have been dealing with this common, often debilitating condition. Demographic studies have shown that women over the age of 80 are the fastest growing population segment in Canada and the United States. This elderly female population segment will continue to grow at this pace until approximately 2050 (Figs. 1 and 2).

Pelvic organ prolapse (POP) affects almost half of all women over 50 years of age, with a lifetime prevalence of 30–50% [2]. A 1997 study found that women with normal life expectancy by the age of 79 years have an 11–12% chance of undergoing at least one operation for prolapse or incontinence, with a reoperation rate of 29.2% [3] (Fig. 3). As the current generation of women maintains a more active lifestyle into an older age, it is likely that an increasing number of women will seek treatment for prolapse. It has been projected that over the next 30 years, the rate of women seeking care for pelvic floor disorders will double [4]. Each year, approximately 300,000 women require surgery for pelvic organ prolapse (POP) and stress urinary incontinence [5, 6]. The direct cost of prolapse surgery is greater than $1 billion per year [2]. Moreover, surgically managed patients represent only a subset of symptomatic patients. Many patients may opt to treat their problem conservatively. They may fail to seek medical advise, thinking this is part of a normal aging process, or they may feel shy to discuss their problem with a health care provider. Pelvic organ prolapse is also commonly found upon physical examination in asymptomatic women [7, 8], in which case no treatment is indicated.

Patients’ symptoms of vaginal apical prolapse can vary from being asymptomatic to complaints of vaginal pressure, feeling something “coming down,” coital difficulties, urinary symptoms (urgency, frequency, incontinence, or voiding dysfunction), or bowel emptying difficulties, due
to concomitant anterior and posterior vaginal wall support defects. Burrows et al. [9] looked at the correlation of symptoms with the severity of pelvic organ prolapse and concluded that the more advanced the pelvic prolapse, the less likely the women will have stress urinary incontinence and the more likely they will manually reduce prolapse to void. However, prolapse severity was not associated with sexual or bowel symptoms. Another study correlating symptoms in women with or without enterocele showed that women with an enterocele were more likely to be older and postmenopausal. They were also more likely to have previous hysterectomy or vaginal prolapse repairs, with more advanced apical and posterior vaginal prolapse than women without an enterocele; they did not differ from them in regard to bowel function [10].

Weber et al. [11] described risk factors for the development of POP (e.g., race, gender, genetic factors, pregnancy and delivery, hysterectomy for prolapse, myopathy, neuropa-thy, obesity, smoking, pulmonary disease, constipation, occupational activities, aging, menopause, and medication) and categorized them into factors that predispose, incite, promote, and decompensate. Depending on the combination of risk factors that women are exposed to, prolapse may or may not develop during their lifetime. Looking at 150,000 patients, Olsen [3] found that women with prolapse were older, postmenopausal, parous, and overweight. Sze et al. [12] have shown that caesarean section (CS) during labor and SVD had similar effects on pelvic support, considering the role of pregnancy, labor, delivery, race, and POP. Black women were as susceptible to prolapse during childbirth as white patients, and elective CS provided only a partial protection against prolapse. O’Boyle et al. [13] found that in nulliparous women, pregnancy alone is associated with increased pelvic organ prolapse compared with nonpregnant control subjects. Strohbehn et al. [14] found that young women under the age of 35 undergoing surgery for genital prolapse were more likely to have potentially predisposing medical conditions (congenital anomalies or neurological or connective tissue diseases) compared to elderly women. Lang et al. [15] looked at the correlation between estrogen level and the occurrence of POP, and found that serum estrogen and estrogen receptor (ER) leads were significantly lower in the uterine ligaments of premenopausal women with POP. There was no significant difference in the ER values in the postmenopausal group.

Etiology may also play a role in connective tissue disorders as a factor in the pathogenesis of POP. Women with Marfan or Ehlers–Danlos syndrome have been shown to have high rates of urinary incontinence and pelvic organ prolapse [16]. Collagen is a fibrous protein providing much
of the tensile strength for skin, tendons, and bone. Nineteen types have been identified, with types I and III being the main structural component of epithelial tissue. Jackson et al. [17], in a cross-sectional study, analyzed vaginal epithelial tissue from premenopausal women with genitourinary prolapse and compared them with controls; they found a reduction in total collagen content secondary to increased collagenolytic activity.

Other morphological studies have confirmed abnormal histological findings in association with prolapse. Wong et al. [18] have shown a decrease in collagen content in nonsupport tissue (cervix) in women with prolapse regardless of age, parity, BMI, or smoking. Chen et al. [19] have shown an increase in collagen breakdown in stress urinary incontinence (SUI) and POP groups compared to controls, and they have shown a decrease in alpha-1 antitrypsin mRNA in the vaginal wall of premenopausal women with POP compared to controls [20]. This altered ellastin metabolism may contribute to the connective tissue alterations observed in pelvic floor dysfunctions. Smooth muscle fraction of the round ligament in women with POP is reduced compared to controls [21]. Keane et al. [22] showed that the collagen composition of the vaginal epithelium closely resembles that of the endopelvic fascia; hence, information about the pelvic floor integrity may be obtained by sampling the vaginal fascia.

The same group looked at premenopausal nulliparous women with and without SUI and found significantly less collagen and a lower ratio of collagen I to III in the SUI group [23]. Collagenolytic activity is increased in SUI patients, as measured by urinary helical peptide alpha in urine [24]. Despite the difference in collagen breakdown between women with and without SUI, there was no difference in collagen synthesis between the two groups when Chen et al. [25] examined endopelvic fascia and skin.

In conclusion, pregnancy and delivery are considered major risk factors in the development of POP and SUI; however, different risk factors have been noted in young women with POP. The fact that collagen content is reduced in women with POP/SUI is most likely due to increased collagen destruction, as production does not appear to be altered. We, as clinicians and researchers, should ask the question: What is it that triggers collagen breakdown in POP and SUI patients, and can that be inhibited or reversed? More research is clearly needed in this field to look at the genetic background in women with POP and SUI in aiming to identify women who may be genetically predetermined to develop pelvic support disorders. This information would be enormously helpful in the counseling of such women to possibly avoid events such as vaginal birth and vaginal route pelvic organ prolapse surgery that is associated with a higher recurrence rate in these women. Also, it would be prudent to counsel these women to avoid activities and other risk factors that play a role in the development of this debilitating and costly health problem.

The role of adjuvant materials in reconstructive pelvic surgery is an important area where evidence-based medicine and randomized controlled trials are lacking. Issues such as the need for concomitant repair of asymptomatic hernial vaginal defects, performance of concomitant vaginal hysterectomy, and implantation of mesh supports will require greater clinical evaluation and scientific scrutiny.

References

Evolution of biological and synthetic grafts in reconstructive pelvic surgery

Abstract  Surgery is an evolving science in the attempt to make surgical procedures more effective, safer, and less invasive. Recurrence and subsequent re-operation for stress incontinence and prolapse has been reported to be necessary in one of three patients, so there is a need for improvement [1]. In reconstructive pelvic surgery (RPS), the use of biological and synthetic grafts for the transabdominal and transvaginal treatment of pelvic organ prolapse (POP) or stress urinary incontinence (SI) has improved long-term support and function after surgery. However, the potential benefits of using grafts need to be carefully balanced against the risks of using materials foreign to the patient’s body. Pelvic organ prolapse develops secondary to defective endopelvic fascial and muscular support. The levator ani provides resting tonic muscular support for all three pelvic compartments. Once neuromuscular damage occurs, extra strain is placed on the connective tissue supports, which may also subsequently fail. To date, there is no surgery that adequately addresses the issue of neuromuscular damage of the pelvic floor musculature. In conventional POP surgery, defective support is repaired by suturing of the patient’s own connective tissue, fascia, or ligaments. The rationale for the use of grafts is to reinforce and strengthen pelvic organ repairs similar to the use of grafts to strengthen abdominal hernia repair.

Keywords  Pelvic organ prolapse · Stress incontinence · Surgery · Biological graft · Synthetic graft

Biological and synthetic grafts

Biological grafts may be derived from the patient’s own body (autologous graft), from post-mortem tissue banks (allograft), or from animals (xenograph). Donor sites for allografts are dura mater, rectus sheath, and fascia lata. Xenografts used in pelvic reconstructive surgery include modified porcine dermis (Pelvicol, Bard, Intexen, AMS), porcine small intestine (SIS, Cook), and bovine pericardium (Veritas, Synovis). Xenografts and allografts are carefully selected from a screened donor population and are appropriately sterilized and processed. However, there remains a small risk of prion or viral infection estimated to be approximately 1 in 2 million [2]. The other important issue with some biological grafts is their ability to provide long-term support to the grafted repair. Xenografts have been found to degrade after implantation with loss of graft integrity and strength, both in animal [3] and human studies [4]. Claerhout et al. [3], in a study using rabbits, found that the explanted porcine grafts of pelvicol and SIS had significantly reduced tensile strength compared to polypropylene mesh over a 12-month period.

Synthetic mesh may be non-absorbable, absorbable, or a mixture of the two. Amid [5] in 1997 published a classification for synthetic grafts used in abdominal hernia surgery based on the pore size and fiber type of the synthetic mesh. The mesh may be mono- or multi-filament, and the tensile strength of the mesh will vary with fiber type, the weight-to-area ratio, and the weave. The pore size and interstices distance (Fig. 1) are important mesh characteristics that determine whether host inflammatory cell and fibrocollagenous tissue can penetrate the mesh construct. The diameter of the open spaces is the pore size, greater than 75 μm is defined as macroporous, and the interstitial distance is measured between the fibers. Polypropylene mesh is usually a type I macroporous and monofilament graft and is the most frequently used synthetic non-absorbable mesh in reconstructive pelvic surgery. Meshes can be of different weaves and weights, with Prolene (85 g/m²) and Marlex (95 g/m²) as examples of a heavier mesh and Gynemesh PS (43 g/m²) as a lighter
mesh recently developed for reconstructive POP surgery (Fig. 2). Other lighter meshes are Vipro I and II; polypropylene is mixed with Polyglactin 910 fibres, which were primarily developed for abdominal hernia surgery.

Type I monofilament macroporous mesh has been reported to have a lower incidence of infection and vaginal erosion. The larger pore size and monofilament nature of the mesh is thought to lessen the risk of infection by allowing easier penetration of inflammatory cells such as leukocytes (9–15 μm) and macrophages (16–20 μm) into the graft to phagocytose bacteria (<1 μm). However, this explanation may be overly simplistic as macrophages, despite their size, have also been shown to penetrate microporous multifilament grafts. Another possible factor may be the greater surface area of multifilament grafts relative to monofilament grafts. Monofilament macroporous grafts allow connective tissue ingrowth into the mesh with collagen deposition around the polypropylene fiber (Fig. 2). Bobyn et al. found that the best mechanical anchorage occurs when pore size was between 50 and 200 μm with an average of 90 μm. Polypropylene can also be woven into a multifilament graft (type III) as in the intravaginal slingplasty (IVS) tapes (Tyco) used for stress incontinence and prolapse surgery. The tensile strength of synthetic meshes used in RPS varies considerably; the tension-free vaginal tape (TVT) polypropylene sling is very distensible with low tensile strength while the IVS polypropylene multifilament tape is densely woven and non-distensible (Fig. 3). After implantation, the TVT mesh becomes fully incorporated with dense mature connective tissue while there is encapsulation of the IVS tape (Fig. 4). In microporous multifilament grafts such as Gore-Tex, there is encapsulation of the entire graft by collagen but no ingrowth (Fig. 5).

The different physical properties of synthetic grafts will impact on the clinical outcome once implanted into humans. This may have clinical disadvantages (erosion, infection) and benefits (easier tension adjustment of the graft after surgery) of the different mesh characteristics in vivo. Pifarotti et al. [6] in a prospective randomized study of TVT (macroporous monofilament) vs IVS (microporous multifilament) slings found they had similar effectiveness for the treatment of stress incontinence. The IVS tape had a lower incidence of postoperative voiding difficulty but had a 9% incidence of tape removal for vaginal erosion and infection/rejection. Bafghi et al. [7] found that 11 of 149 woman (7.5%) with the IVS tape for stress incontinence developed tape erosion, and ten required surgical exploration and removal (four vaginal and six abdominal approach). Eight of the ten women developed urinary incontinence postoperatively with five of them having symptoms of stress incontinence and three with urge incontinence.

Good clinical studies are necessary to provide us with a clear understanding of the risks and benefits of grafts before widespread introduction.

**Grafts for stress incontinence**

The controversy of the use of grafts vs the body’s native tissue is as old as surgery itself. In reconstructive pelvic surgery, at various times, procedures become popular and are seen as the “gold standard” only to be challenged when
new procedures are found to offer a more effective or safer option.

The suburethral sling using rectus sheath fascia described by Aldridge in 1942 [8] and again popularized by McGuire in the 1980s continues to be a popular procedure for both primary and recurrent stress incontinence. This procedure requires an abdominal and vaginal incision for graft harvesting and placement of the sling under the bladder neck. However, the abdominal incision does increase postoperative morbidity and pain, as well as dissatisfaction with this procedure.

Suprapubic urethral suspension procedures described by Marshall, Marchetti, and Krantz (MMK) in the 1950s and by Burch in the 1960s [9] were certainly seen as gold standard procedures by many gynecologists for the latter part of the 20th century. Suture placement in the symphyseal periosteum caused osteitis pubis in a small number of patients having an MMK so the iliopectineal ligament was used for the suspension site in the Burch colposuspension. Both the fascial sling and the Burch colposuspension have high rates of cure, with the Burch colposuspension having a lower postoperative incidence of voiding dysfunction.

The use of synthetic slings is not new although there have been many changes in recent years. Moir [10] and Morgan [11] were using polypropylene mesh slings in the 1960s, which they placed under the bladder neck through an abdominal and vaginal incision. The tension-free vaginal tape and intravaginal sling were introduced by Ulmsten [12] and Petros [13]; polypropylene tapes were placed under the mid-urethra through small suprapubic and suburethral stab incisions. The TVT procedure has been found in retrospective and prospective randomized studies to be as effective as the Burch colposuspension with a lower morbidity and less postoperative hospitalization [14]. Vaginal erosion rarely occurs (<1%) and is caused by failure of healing over the suburethral incision or “button holing” of the lateral vaginal sulci not recognized at the time of surgery. Mesh penetration into the bladder and urethra can occur, usually at the time of surgery when it may go unrecognized. The transobturator sling [15] is the latest variation, which may have a lesser risk of bladder penetration and be an easier procedure to learn. The question remains whether the long-term effectiveness of transobturator slings is equal to the retropubic tension-free vaginal sling.

**Grafts in prolapse surgery**

The choice of surgical procedure for POP will be guided by the site and severity of defective pelvic support, the patient’s previous history and co-existing pelvic pathology, and the surgeon’s preference based on experience and training. Synthetic grafts have been used in the surgical treatment of post-hysterectomy vaginal vault prolapse since the early 1960s [16]. In the abdominal colposacropexy, synthetic non-absorbable mesh is sutured over the anterior and posterior vaginal walls and attached to the anterior sacrum. Synthetic mesh is placed on the anterior and posterior vaginal walls and will usually correct loss of apical as well as lower vaginal defects such as cystocele, enterocele, and rectocele. If the uterus needs to be conserved, synthetic mesh can be used to support the upper vagina and cervix to the sacrum (colpohysterosacropexy). Complications of the abdominal prolapse approach include hemorrhage, particularly from the anterior sacral vessels, and injuries to bowel and urinary tract. There are also complications from the abdominal incision, including wound infection and dehiscence. Vaginal mesh erosion has been reported to occur in approximately 2% of cases using polypropylene mesh [17, 18] and up to 11% of cases with polytetraflourethylene (Gore-Tex), a microporous multifilament mesh (type II) [19], and polyethylene tetraphalate mesh (Mersilene) (type III). Concurrent hysterectomy with abdominal mesh colpopexy is a significant risk factor [20]. The treatment of mesh erosion will also vary with mesh type. Polypropylene monofilament macro-porous mesh erosion, even in the presence of infection, can be successfully treated by transvaginal excision and repair with antibiotics; however, microporous multifilament
meshes will usually require complete removal by abdomi-
nal and vaginal exploration before infection is eradicated
[8].

Allogenic graft materials using fascia lata [4] and rectus
sheath [21] have been used for abdominal vault suspension.
This procedure has not gained popular acceptance possibly
due to reports of high short-term failure [4] but may be of
value in patients where synthetic mesh is inappropriate
(e.g., presence of infection).

The abdominal approach using synthetic mesh is now
well accepted by reconstructive pelvic surgeons for the
treatment of pelvic organ prolapse. In a recent review,
Brubaker [21] concluded: “it is clear for anatomical
restoration the abdominal colpopexy is the gold standard
but at a cost of higher short term morbidity and potential
foreign body problems.” This opinion was based on the
results of two well-conducted prospective randomized
studies comparing the abdominal colpopexy with polyprop-
ylene mesh to the transvaginal sacrospinous colpopexy
for apical and vault prolapse by Benson et al. [22] and
Maher et al. [23]. The use of synthetic mesh with the
transvaginal repair of prolapse is more controversial.
Gynecologists have been reluctant to use synthetic mesh
vaginally because of the risk of infection and erosion into
the urinary, genital, or gastrointestinal tract. Synthetic non-
absorbable mesh using polypropylene has been used in the
transvaginal repair of anterior and posterior compartment
prolapse as well as in the perineal approach for the repair
randomized study using polypropylene mesh in 24 women
with recurrent cystoceles. After 2 years, four women in the
control group and none in the mesh group had recurrent
anterior wall prolapse. Three of the 12 women (25%) with
mesh repairs had erosions. Synthetic absorbable mesh has
also been used. Sand et al. [25] reported a prospective study
of Poligleactin 910 mesh (Vicryl mesh, Ethicon) placed on
the anterior endopelvic fascia for cystocele. At 12 months,
there was a significantly lower recurrence rate in the mesh
group compared to the controls (25 vs 43%). Weber et al.
[26] found no significant difference in recurrence of
cystocele in a prospective randomized study using Vicryl
mesh.

Mesh erosion has been a common postoperative problem
particularly after transvaginal mesh repair occurring in 9
[27] to 13% [28] (Fig. 6a,b). This usually occurs secondary
to failure of healing of the vaginal incision. Other risk
factors for erosion in the transvaginal mesh repairs found
by Achtari et al. [28] were the surgeon’s experience and
previous vaginal surgery; the weight of the mesh was not a
significant factor. Small mesh exposures may re-epithe-
lialize spontaneously, often aided by the use of intravaginal
estrogen. Transvaginal excision of the exposed polyprop-
ylene mesh and vaginal closure can be easily performed if
the mesh exposure persists or is symptomatic (bleeding or
pain).

Milani et al. [29] reported a high rate of dyspareunia
after vaginal repair with mesh particularly in the posterior
compartment. In our experience [27], the incidence of
dyspareunia decreased after transvaginal repair with mesh
from 20% preoperatively to 7% at 24 months, similar to our
results with abdominal mesh colpopexy [24]. The place of
synthetic grafts in the transvaginal repair of prolapse
remains to be clarified by good prospective randomized
studies assessing the long-term anatomical and functional

Fig. 6 Photograph of erosion of Atrium mesh after a transvaginal repair and b histopathology

Fig. 7 Transvaginal mesh placement for anterior and posterior wall prolapse (Dwyer et al., Br J Obstet Gynaecol 2005)
outcomes. At the present time, there is no agreement between investigators on who should have synthetic mesh or how and where the mesh should be placed and secured. The transvaginal approach will need to be standardized by investigators before prospective studies can occur. In our study [28], the mesh was placed over a midline fascial repair as an overlay graft after wide lateral and superior dissection in both the anterior and posterior compartments (Fig. 7). We have found that removal of vaginal epithelium is usually unnecessary with implanted polypropylene mesh.

**Conclusion**

The use of grafts to repair damaged fascia and defective support is only one aspect of achieving a successful outcome in POP or SI surgery. Of equal importance is the careful preoperative evaluation of the patient’s symptoms and sites of defective support, the surgical procedure chosen (abdominal vs vaginal approach), the experience and the skill of the surgeon, and the patient’s own characteristics (e.g., smoker, obesity, and previous surgery).

Synthetic grafts have clearly established an important role in the surgical treatment of stress incontinence and the abdominal repair of vaginal prolapse. The role of synthetic or biological grafts in the transvaginal repair is controversial and awaits clarification by further prospective randomized studies. Surgical mesh reinforcement should not be used to replace good surgical technique. At the very least, good animal studies and retrospective clinical trials are needed before the widespread introduction of grafts in the transvaginal repair of uterovaginal prolapse.

Newer synthetic and biological grafts hold considerable future promise but we should proceed cautiously unless we do more harm than good. Shull and Karram [30] in an editorial in the *International Urogynecology Journal* advised caution in too hastily taking up new procedures and products proposed by industry without critical evaluation for efficacy and safety. They emphasized the need for a good knowledge of surgical anatomy and basic surgical principles of colporrhaphy and vaginal reconstruction for all pelvic surgeons.

**References**


Abstract  Implant materials are increasingly being used in an effort to reduce recurrence after prolapse repair with native tissues. Surgeons should be aware of the biology behind both the disease as well as the host response to various implants. We will discuss insights into the biology behind hernia and abdominal fascial defects. Those lessons from “herniology” will, wherever possible, be applied to pelvic organ prolapse (POP) problems. Then we will deal with available animal models, for both the underlying disease and surgical repair. Then we will go over the features of implants and describe how the host responds to implantation. Methodology of such experiments will be briefly explained for the clinician not involved in experimentation. As we discuss the different materials available on the market, we will summarize some results of recent experiments by our group.

Keywords  Graft biology · Vaginal prolapse · Biologic implant · Synthetic implant · Surgical repair

Parallels to herniology

Although the patient presents with a single and precisely located symptom (protrusion of organs through a locus minoris resistentiae), this is usually a sign of a more systemic change in the host’s connective tissues. This concept is now widely accepted by general surgeons dealing with inguinal hernia. Hernia patients have a different fibroblast phenotype, who qualitatively and quantitatively synthesizes abnormal collagen [1]. Collagen is normally nearly completely made up of type I and type III collagen, in a ratio of about 4:1. Hernia patients now have an altered collagen I/III ratio. Friedman described over-expression of collagen III by fibroblasts from hernia patients. This leads to an excess of non-polymeric, extractable collagen, by inhibiting cross-linking of, and with, predominant type I [2]. The resulting fibers are inherently thinner and weaker, and more susceptible to lysis. This phenomenon affects different tissue types, including fascias, but also those at distance from the defect [3]. Collagen normally protects fibroblasts from apoptosis; conversely abnormal collagen does the opposite [4]. Read also described enhanced elastolytic activity and reduced anti-protease capacity [5]. Those proteases are increasingly released by leukocytes primed by smoke, linking this to another recognized co-factor in herniation. Matrix metalloproteinases (MMP) that can break down extracellular matrix proteins are over-expressed in hernia patients as well, although this was first shown by Jackson et al. in vaginal tissue of women with prolapse [6, 7]. A subpopulation of hernia patients may be affected by other signs of connective tissue abnormalities, such as joint hypermobility or aneurysmata [8, 9], and their lesions show the same molecular alterations as described above. Obviously, some genetic conditions, among which is Marfan’s or Ehlers–Danlos syndrome, directly affect collagen metabolism, hence, predispose to hernia.

In summary, hernia patients have a systemic diminished collagen synthesis, a protease–antiprotease imbalance, an increased MMP activity, and reduced collagen I/III ratio. Unfortunately, this problem persists also in the postoper-
ative phase, and it has been shown that after surgical correction they, expectedly, display failing wound healing [10]. However, in certain life events, acquired or co-morbidity factors may be the decisive factor rendering patients with the above changes symptomatic. The mechanisms through which this happens in hernia patients are well-identified. Smoking was one of the first factors to be recognized. Its effects on collagen metabolism are systemic as decreased oxygen levels preclude normal cross-linking activity all over. Nicotine has on itself an inhibitory effect on fibroblast proliferation. Women who smoke are at higher risk than men [11]. Later, it was even shown that heavily smoking women increase the risk for congenital hernia in their offspring; however, this might be an indirect effect by an increased risk for preterm birth [12].

Aging contributes in different ways to the development of inguinal herniation: there is muscle wasting, the composition of connective as well as fat tissue changes, and age may directly affect collagen metabolism, e.g., by increased or prolonged MMP activity. Even some drugs, like the antihypertensive angiotensin-converting enzyme (ACE) inhibitors also directly interfere with collagen metabolism. Much of these “herniology” insights applies to patients with pelvic floor disorders as well. Those patients also have a certain predisposition, with increasingly better defined molecular and biochemical alterations. The latter were recently excellently summarized in the proceedings of a meeting on pelvic floor disorders sponsored by the National Institutes of Health, and this is beyond the scope of the talk [13]. Anyway, to the underlying changes additional co-factors may add. All those earlier described in hernia patients may apply. One additional typical life event is usually named to be causative for POP and incontinence, i.e., pregnancy and childbirth injury. Claimed mechanisms include direct injury to pelvic floor muscle and/or their attachments, nerve injury due to stretch or compression, but this is beyond the scope of this talk. Available epidemiologic data as to a causal relationship are at present controversial, as the effect of birth on pelvic floor problems disappears in the elder population, as shown in the EPINCONT study [14].

Animal models in the study of fascial defects

Tissue remodeling processes can be studied in culture or in vitro conditions. In vivo evaluation is, however, an additional important research tool. Animal models are convenient as they allow for complex experimental design or discounting an abundance of interfering co-factors as in the clinical situation. Moreover, their use reduces risks to patients, and saves time and money in understanding the problem. There are limitations to their applications in the study of POP, like anatomical ones: nearly all animals are quadrupeds, with a different pelvic floor musculature including a functional tail, and they have a different birth process. It is difficult, if not impossible, to evaluate functional problems correctly, and it is complex to mimic known co-factors, such as smoking, obesity, chronic lung disease, etc.

Rodents are cheap and widely available. (Immuno)histologic as well as molecular techniques are generally applicable, and transgenic mice, which become increasingly important, will eventually show up in this field as well. Lower urinary tract problems can already be reproduced in rodents. They have been used as models for pudendal neuropathy induced by trauma, toxic substances, or diabetes [15]. Sophisticated experiments were conducted to mimic hypoestrogenism, the effect from birth using prolonged vaginal distention and inducing pudendal nerve damage [16, 17]. More recently, the structural properties of the rat vagina and supportive structures were documented [18]. Other models include dogs because certain breeds spontaneously develop stress incontinence as they age [19]. An effect of body weight or birth has been shown and they were used to study therapeutic interventions. Rhesus and squirrel monkeys can be used to study prolapse, but their shorter lifespan is a limitation, next to limited access due to economical and ethical restrictions. In rhesus monkeys the direct influence of estrogens on vaginal smooth muscle density, biomechanical strength and collagen content was shown [20]. Squirrel monkeys develop age- and parity dependent spontaneous vaginal prolapse and pelvic floor innervation is very comparable to that in humans [21, 22]. This was not so in baboons, although they are anatomically closer to humans [23].

Physical properties required for implant materials

Clinicians need to become familiar with some relevant features when describing prosthetic materials. Products are first discriminated on the basis of the source they are derived from (Fig. 1). Synthetic materials have long been known and their physical properties include tensile strength and elasticity. As implants are designed to provide permanent reinforcement of the reconstructed fascia, so they should be strong enough to withstand naturally occurring forces. For prolapse surgery, the mechanical requirements for implants to meet have not been defined. Usually, insights from hernia surgery are extrapolated (for what they are worth). The tensile force implants that should withstand naturally occurring forces were modeled mathematically resulting in a calculated minimal strength of 16 N/cm for hernia and 32 N/cm for abdominal wall replacement [24]. In dry conditions, most meshes are much stronger than what is physiologically needed, and recently it was understood that wider woven (“light weight”) material could be used without compromising the physical needs. Physiological elasticity requirements, measured at 16 N/cm, are between 20 and 35%; most implants are not that elastic, which may cause symptoms [25]. Clinically intra-abdominal pressures are at their highest 100–150 mmHg. Measurements of intra-abdominal pressure during Shouldice hernia repair under local anesthesia
showed maximum pressures during coughing of around 60 mmHg [26]. Similar to the breaking strength of healthy tissues and that of different suturing techniques, these pressures correspond to a strength far below 10 N/cm, so ultimately, even lesser tensile strength might result in good clinical results [27]. Of note is that elasticity and strength can be tested in “dry” lab conditions (before implantation) but measurements in explanted meshes are clinically more relevant.

Host response to implants

Although many of the used materials are referred to as chemically and physically inert, stable and not immunogenic, none of them are truly biologically inert. Most synthetic materials (unlike biomaterials) induce a relatively vigorous foreign body response [28]. The nature of the material, structure, amount, and for synthetics, filament and pore size, determines this process, as well as further tissue in-growth. However, the course of events is initially fairly constant irrespective of the species or material. After implantation, the host immediately reacts to the injury and covers the material with a biofilm. Host proteins are adsorbed at the interface and a complex host-to-implant material interaction sets off [29]. The first step of protein adsorption takes place within seconds and does not require any cellular response. It follows a fixed and hierarchical pattern referred to as the Vroman effect, a phenomenon any cellular response. It follows a fixed and hierarchical pattern sets off [29]. The first step of protein adsorption takes place within seconds and does not require any cellular response. It follows a fixed and hierarchical pattern sets off [29].

As capillaries and inflammatory cells get less in number, fibroblasts become increasingly predominant and deposit collagen and other matrix proteins, and a more fibrous tissue becomes evident. There is progressively more zonation in the interface area, with most cells in direct contact with the foreign material. At this moment in time, the implant is also mechanically stabilized, at least if there is no remnant micromotion. The collagen deposited at the mesh interface unfortunately is of lesser quality than after suture repair, irrespective of the type of material used [35]. The remodeling process also takes place in an individual who already had a defective collagen metabolism to start with.

While the inflammatory phases are necessary for the desired fibrosis part of the process, it may be the source of some adverse effects, such as implant-shrinkage, erosion, or adhesion formation [36, 37]. The amount of foreign body reaction increases with the surface of the foreign material being exposed to the host. Reduction in material can be achieved through different variables. It would follow that multifilament meshes inherently induce more reaction, but there seems to be an absolute lower limit for filament size. When filament size is <4/0, a constant granulomatous reaction was demonstrated, irrespective of the polymer or number of filaments [38]. Pore size is also an important factor for fibroblast infiltration, flexibility, and mechanical integration. Pore sizes >50 μm allow for rapid in-growth of fibroblasts and vascular elements necessary to anchor the implant within the native tissue [39, 40]. Peak in-growth is reached at pore size around 400–500 μm. Larger pores limit the fibrosis process to the perifilament region, and the pores get filled with fat [38]. A solid product as well as one with smaller (<50 μm) pores will be encapsulated or induce an increased foreign body

From then on, a more typical course of an acute inflammatory, followed by chronic inflammatory reaction will follow, transiting ultimately into a foreign body reaction, i.e., formation of granulation tissue with fibroblasts, macrophages and eventually foreign body giant cells, with the occurrence of neovascularization as well as fibrosis. The inflammatory cell type predominantly present at the interface will vary according to the phase of the host response. Initially, neutrophils predominate, but they are gradually replaced by monocytes, which differentiate extravascular to macrophages. Macrophages are the crucial cell type in the ultimate clinical response of either biotolerance or rejection of the foreign body. In reality, they already play a role from the moment the complement cascades and blood clotting process is activated; they interact with lymphocytes, and produce mediators that induce protein synthesis and cell proliferation (such as endothelial cells and fibroblasts) [33]. They are also constantly found in explanted materials. They can morphologically mimic other cell types, like epitheloid macrophages, or fuse in typical foreign body giant cells, just like the Langhans cells in tuberculosis. Because of the activated status and production of mediators, other cell types migrate to the implant site and participate in a chronic wound-healing process that can go on for years [34].

As capillaries and inflammatory cells get less in number, fibroblasts become increasingly predominant and deposit collagen and other matrix proteins, and a more fibrous tissue becomes evident. There is progressively more zonation in the interface area, with most cells in direct contact with the foreign material. At this moment in time, the implant is also mechanically stabilized, at least if there is no remnant micromotion. The collagen deposited at the mesh interface unfortunately is of lesser quality than after suture repair, irrespective of the type of material used [35]. The remodeling process also takes place in an individual who already had a defective collagen metabolism to start with.

While the inflammatory phases are necessary for the desired fibrosis part of the process, it may be the source of some adverse effects, such as implant-shrinkage, erosion, or adhesion formation [36, 37]. The amount of foreign body reaction increases with the surface of the foreign material being exposed to the host. Reduction in material can be achieved through different variables. It would follow that multifilament meshes inherently induce more reaction, but there seems to be an absolute lower limit for filament size. When filament size is <4/0, a constant granulomatous reaction was demonstrated, irrespective of the polymer or number of filaments [38]. Pore size is also an important factor for fibroblast infiltration, flexibility, and mechanical integration. Pore sizes >50 μm allow for rapid in-growth of fibroblasts and vascular elements necessary to anchor the implant within the native tissue [39, 40]. Peak in-growth is reached at pore size around 400–500 μm. Larger pores limit the fibrosis process to the perifilament region, and the pores get filled with fat [38]. A solid product as well as one with smaller (<50 μm) pores will be encapsulated or induce an increased foreign body

Fig. 1 Schematic representation of classification of implant materials, first on their source, then by other variables
reaction, bridging from one filament to the other—filling the entire pore [41]. Based on all the above, there is now a consensus in the surgical literature that improved results will be achieved by using a low weight, large pore, monofilament mesh, with an elasticity between 20 and 35% [42].

**Animal studies in fascial repairs with implants**

To study the improvement of fascial repairs using meshes, surgical animal models are used. Again, rats often serve as an incisional hernia model or, more generically, as a model for fascial defect repair, with or without the use of implant materials [43, 44]. These are laid in or over the defect, with or without direct contact to the peritoneal cavity (Fig. 2). Direct contact with the viscera may be interesting in the study of adhesion formation. Such animal models are very important in preclinical “mechanical” evaluation of implants, as well as evaluation of their biocompatibility.

Adult rabbits weigh between 3 and 5 kg in larger breeds, so their size allows for the creation of several defects and for these to be covered by different materials within the same animal [45]. Even laparoscopy is possible. Rabbits have a larger bowel size than rats, but have a different collagen metabolism, a factor that must be taken into account when analyzing the wound-healing process. Vaginal surgery is possible, and recently the model was proposed by de Tayrac [65] for the study of the occurrence of erosions.

Understandably, most clinicians are not familiar with the experimental design of studies on implants. Basically, one or more standardized full-thickness defects are created in the abdominal wall of the selected species (Fig. 3). Defects are primarily repaired, using an implant overlay and sutures, and/or a suture repair group. Animals are allowed to survive days to months, and in rabbits even for years. Sacrifice takes place at given times, depending on the purpose of the study. At that moment, the implant, the interface and neighboring host tissue, together referred to as explant, are harvested, divided, and conserved in the appropriate way for further evaluation. We have used mainly the rat to document all phases of the remodeling process. This includes the early phases of the acute and chronic inflammatory reaction, when standard morphometric techniques are used to identify and count the inflammatory cells (polymorphonuclear, mononuclear/macrophages, leukocyte-type, and fibroblast neovascularization) at the host-implant interface. Occasionally, immunohistochemistry or flow cytometry will be used [47]. Fibrosis is semi-quantitatively assessed by Movat or Sirius Red stain. More quantitative and qualitative methods are available but logistically more complex and expensive. Immunohistochemistry for different subtypes of collagen can be done, but it needs to be species-specific. Molecular techniques can be used to quantify cytokines or other proteins of interest. Biomechanics are tested by tensiometry of explants; for long-term (2 years or longer) experiments, rabbits may be more suitable. Immunologic reactions can be studied in euthymic mice or mouse strains without functional T or/and B lymphocytes (nude or SCID mice) [48]. Rats and mice are very resistant to infections, rendering them suboptimal models for the study of perioperative wound infections.

**Synthetic materials**

Synthetic implants can be made from knitted single-fiber filaments (monofilament materials) or they can be braided with monofilament yarns, further woven as multifilament fibers in different ways and pore sizes (Figs. 1 and 4). Knitted fabrics have a more open structure than woven [49]. Such materials are usually further classified according to their pore size (Amid classification; Table 1) [50], the nature of their composing fibers (mono- or multifilament), and their resistance to degradation (absorbable, non-absorbable, or composite or mixed materials).

The nature and extent of the inflammatory reaction these materials generate is regulated by the chemical and physical structure of the implant, the amount of material, and surface of the contact-area with the host [51]. Flexibility and strength are other physical properties. The flexibility of an implant is determined by the individual stiffness of its yarns, the knitting procedure, the pore size, or, in general, the amount of material per unit of surface. In that respect, the density of the product is an interesting variable. Implants with large pores are more flexible than implants with smaller pores. Implants that are more looped have smaller pores and a higher degree of stiffness; multifilament meshes are more flexible or supple [41, 49].

Polypropylene (Fig. 4) is the most widely used material to fabricate cheap, inert, and easily tailorable implants. It preserves its chemical and mechanical integrity for years.

Fig. 2 Different implantation sites for meshes in experimental models. First the skin is incised (yellow or top bar) and a defect is created involving the muscle and fascia of the abdominal wall (red or middle bar). An implant (green or lower bar) can be positioned as an “inlay” (a) or “overlay (b), and even retroperitoneal (c). Theoretically, the fascial defect is not relevant when only adhesion formation is the purpose of the study (d). Top of the section is the skin and subcutis, bottom is the peritoneum (blue)
and exhibits high burst and tensile strength in dry laboratory conditions and when incorporated in the native tissue [52]. The majority are monofilament, woven over the years with increasingly large pore sizes, resulting in more elasticity without compromising the physical strength and less local side effects. When applied within the peritoneal cavity, adhesions may develop, which bears relevance to procedures such as sacropexy or abdominal wall hernia repair. Polypropylene is used to weave different type of fabrics, sometimes under the same name. This might be confusing. For instance, Surgipro (Tyco Healthcare, Mechelen, Belgium) is a trademark of a range of polypropylene products, including a typical monofilament as well as two multifilament mesh types (Table 1; Fig. 5). Multifilament implants have *interstices* between threads, which are much smaller than the actual pores, and increase the contact surface with the host. Again, this results in better integration but also a stronger inflammatory reaction. Pore sizes or interstices of less than 10 μm *theoretically* allow passage of bacteria (usually 2 μm or less) but not leucocytes (9–15 μm) and macrophages (16–20 μm). Both cell populations are important in the fibrosis process and in eventual clearance of infection.

Interestingly, these three different products are marketed without clear directions towards their exact clinical applications. The multifilament materials are typically used for hernia repair, where increased malleability, resistance to wrinkling, and softness are important to prevent undesired local side effects [53]. In urology, Surgipro SPM is used to manufacture the Intra Vaginal Sling (IVS) tape. SPMW is used by some surgeons to augment vaginal prolapse repairs (von Theobald, personal communication). We recently compared these different Surgipro products and suture repair experimentally in the rat fascial defect model [54]. Multifilament materials

![Fig. 3](image1.png) **Typical design of a rat experiment.** a A standardized defect is induced using a grid, involving the entire thickness of the abdominal wall. b A mesh is laid over the defect, with its border exceeding the initial incision, and the mesh is sutured to the fascia. Experiments are done sterile and under general anesthesia.

![Fig. 4](image2.png) **Terminology used to classify synthetic implants.** Magnified view of a part of a polypropylene tape as used for TVT procedure (Gynaecare, Johnson and Johnson) with identification of filament, interstitium, and pore.

---

**S20**
induced a shorter lasting acute inflammatory response, transiting in a more pronounced chronic inflammatory reaction, as compared to monofilament implants. Foreign body giant cells were localized mostly around individual filaments. Macrophages could, in contrast to what is usually assumed, be found in interstices as small as 7.5 by 12.5 μm. No difference in collagen deposition and neovascularisation was observed between the three constructs. Multifilament materials were equally resistant to tensiometry, except at the last (90 days) time point, when tighter woven multifilament SPM explants were significantly weaker than polypropylene sutured or SPMM controls. Overall shrinkage was 10% over 3 months, and comparable for all groups.

Other polymers have been used as well, such as polyester. This leads to more foreign body reaction as well as more wound-healing complications, at least in hernia patients [55].

### Biologic implants

Biologic materials conceptually would be an alternative for synthetics as to alter the foreign body reaction, hence avoiding local complications. Biologic implant materials can be divided into autologous implants, where the patient serves as its own donor, heterologous implants, where the material usually comes from the same species but another individual (usually cadaveric material), and xenogenic implants, i.e., material derived from other species. At present, most xenogenic materials are from porcine source, as bovine material became less acceptable.

### Table 1 Classification of synthetic implant materials

<table>
<thead>
<tr>
<th>Component</th>
<th>Trade name</th>
<th>Fibre type</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type I: Totally macroporous</td>
<td>Polypropylene</td>
<td>Monofilament</td>
</tr>
<tr>
<td></td>
<td>Prolene, Gynemesh, Gynemesh PS (Ethicon)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Marlex, Pelvitex&lt;sup&gt;a&lt;/sup&gt; (Bard)</td>
<td>Monofilament</td>
</tr>
<tr>
<td></td>
<td>Surgipro&lt;sup&gt;b&lt;/sup&gt; SPMM (Tyco)</td>
<td>Mono-multifilament</td>
</tr>
<tr>
<td></td>
<td>Vypro (Ethicon)</td>
<td>Monofilament</td>
</tr>
<tr>
<td></td>
<td>Vicryl (Ethicon)</td>
<td>Multifilament</td>
</tr>
<tr>
<td></td>
<td>Polypropylene/Polyglactin 910</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Expanded polytetrafluoroethylene</td>
<td>Multifilament</td>
</tr>
<tr>
<td></td>
<td>Gore-Tex (Gore)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Polyethylene</td>
<td>Multifilament</td>
</tr>
<tr>
<td></td>
<td>Mersilene (Ethicon)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Polytetrafluoroethylene</td>
<td>Multifilament</td>
</tr>
<tr>
<td></td>
<td>Teflon (Gore)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Braided polypropylene</td>
<td>Multifilament</td>
</tr>
<tr>
<td></td>
<td>Surgipro&lt;sup&gt;b&lt;/sup&gt; SPM (Tyco)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Braided polypropylene-open weave</td>
<td>Multifilament</td>
</tr>
<tr>
<td></td>
<td>Surgipro&lt;sup&gt;b&lt;/sup&gt; SPMW (Tyco)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Perforated Expanded Polytetrafluoroethylene</td>
<td>Multifilament</td>
</tr>
<tr>
<td></td>
<td>Mycro-mesh (Gore)</td>
<td></td>
</tr>
<tr>
<td>Type II: Totally microporous</td>
<td>Polypropylene/Polyglactin 910</td>
<td></td>
</tr>
<tr>
<td>Type III: Micro or macro-micro</td>
<td>Polyethylene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Mersilene (Ethicon)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Polyethylene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Mersilene (Ethicon)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Polyethylene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Mersilene (Ethicon)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Polyethylene</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Mersilene (Ethicon)</td>
<td></td>
</tr>
<tr>
<td>Type IV: Submicronic pore size</td>
<td>Polypropylene sheet</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Cellgard</td>
<td>Monofilament</td>
</tr>
</tbody>
</table>

Macroporous is defined as pore size >75 μm, microporous ≤75 μm

<sup>a</sup>Recently collagen coated macroporous polypropylene materials like Pelvitex (Bard) came on the market; their place, if any, needs to be defined

<sup>b</sup>Several kinds of Surgipro (Tyco) materials are marketed under the same name and have different constructs

![Fig. 5](https://example.com/fig5.png)

A selection of different polypropylene implants on the market, showing monofilament (upper three) vs multifilament texture (lower two) and variability of knitting and pore size in between. Variables of interest are pore size and density of material. Materials may not have been photographed at the same magnification (lower three at ×38)
Autologous grafts usually induce very limited foreign body reaction, are well-incorporated into native tissue and they can, in theory, be used in an infected environment. Disadvantages, however, are the surgical morbidity at the prelevation site and the unpredictable durability of the repair, because after absorption they are replaced by host connective tissue that is inherently weak.

Allografts can overcome the problem of surgical morbidity associated with the prelevation of autografts, but not the unpredictable resorption and integration process. Fitzgerald reported autolysis in 20% of the implanted fascia lata grafts [56]. Histological analysis of the grafts showed areas of linear organization of the collagen fibers similar to native fascia altered with areas of poor linear organization and total tissue breakdown. Donor fascia lata is obtained from post mortem tissue banks. Although fastidious steps are taken in the preparation, concerns about the potential risk of viral particles, but more particularly prion transmission remain. The risk of HIV transmission is estimated to be one in eight million [57].

Xenografts are acellular collagen-based scaffolds harvested in certain animals raised for that purpose. Production is strictly controlled by Food and Drug Administration (FDA) guidelines, which include knowledge of the animal herd, vaccination status, feed source, abattoir approval and bovine spongiform encephalopathy clearance. The most commonly used xenografts are porcine derived; bovine material is on the market as well (Table 2). Collagen based implants can either be cross-linked or not. Cross-linking protects the implant against degradation by collagenases, so that they remain intact very long, if not ever. Surgisis is manufactured from porcine small intestinal submucosa in such a way that all cells responsible for an eventual immune response are removed, but the complex extracellular matrix and natural growth factors are left intact. It contains collagen types I, III, and V and growth factors TGF-beta and FGF-2. It exists at present in two-, four-, and eight-layer implants. Small intestinal submucosa (SIS) is degraded in 4 to 12 weeks by a “constructive” remodeling process that replaces the graft gradually by host connective tissue [58, 59]. Tensiometric strength initially decreases down to 45% 10 days after implantation, but by 1 month it is identical to that of the native material [60]. Two years after implantation it exceeds the strength of native tissue. We compared SIS with Marlex (Bard), an elder, rather dense type I polypropylene mesh. The strength of Marlex gradually increased over the experiment, but it induced a more pronounced inflammatory and foreign body reaction. Fibrosis occurs faster with more intense collagen deposition. A number of rats implanted with SIS developed seromas, with fluid accumulation between layers, and even some low-grade local infections. SIS

<table>
<thead>
<tr>
<th>Component</th>
<th>Trade name (non-limiting list)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Autologous grafts</td>
<td>Rectus fascia</td>
</tr>
<tr>
<td>Fascia lata</td>
<td>Vaginal mucosa</td>
</tr>
<tr>
<td>Allografts</td>
<td>Fascia lata</td>
</tr>
<tr>
<td>Dura mater</td>
<td>Lyodura</td>
</tr>
<tr>
<td>Xenografts</td>
<td>Porcine non-cross-linked small intestine submucosal collagen</td>
</tr>
<tr>
<td>Porcine non cross-linked dermal collagen</td>
<td>InteXen (AMS)</td>
</tr>
<tr>
<td>Porcine dermal cross-linked collagen</td>
<td>Pelvicol, Pelvisoft, Pelvilace (Bard)</td>
</tr>
<tr>
<td>Fetal bovine skin derived collagen scaffold</td>
<td>Xenform (Boston Scientific)</td>
</tr>
<tr>
<td>Bovine non-cross-linked pericardium</td>
<td>Veritas (Synovis)</td>
</tr>
</tbody>
</table>

Fig. 6 Cross-section through implant areas in rats, either Prolene (left) or Pelvicol (right). The material can be easily recognized at smaller magnification (a, c). At larger magnification, the intensity of a more pronounced inflammatory reaction becomes more clear in Prolene (b) than Pelvicol (d). From Zheng et al. (2004) [47]
induced less adhesions, an initial slower deposition of less mature collagen, but ultimately became more organized by the end of the study period. Tensile strength was initially lower than for Marlex; however, it reached comparable levels after 3 months [61].

Pelvicol is a porcine dermal collagen implant consisting of a sterile off-white tough but flexible flat sheet of fibrous, acellular collagen and its constituent elastin fibers cross-linked with hexamethylene-di-isocyanate (HMDI). In a rat model, we documented a lesser inflammatory response as compared with Prolene (Johnson & Johnson): there were lesser granulocytes and macrophages, and cells expressed less surface activation markers ICAM-1 and CD11b. Pelvicol induced a slower, but more orderly collagen deposition paralleling the surface of the implant. However, the product is encapsulated rather than a formal in-growth process occurs (Fig. 6). Encapsulation may challenge mechanical strength and has other side effects, such as seroma formation. Tensile strength was initially lower in Pelvicol, slowly increasing to comparable levels by day 90 [47]. An improved strength early on could be obtained by modifying the product with pores of 2.0 mm in diameter. This allows for better tissue ingrowth and more neovascularization [62]. In rabbit experiments, we demonstrated that 1 year after implantation half of the Pelvicol implants were still intact. However, half of them showed progressive signs of degradation, its cause and clinical relevance remaining undetermined. On tensiometry, tensile strength of Pelvicol was initially as strong as Prolene explants but started to decrease 3 months after implantation. In that study also four-layered SIS was used. Tensiometric strength of SIS explants progressively decreased, and some animals displayed bulging through the defect [45] (Fig. 7).

The nature and course of the inflammatory process is different for synthetic and bio-grafts as studied in immunologic models [48, 63, 64]. Synthetics make leukocytes produce a cytokine profile dominated by TNF-α, IFN-γ, and IL-1, which are usually referred to as “T helper 1 type” (Th1) cytokines. They are “pro-inflammatory” in nature: they activate macrophages and are typical for rejection processes. SIS and also Pelvicol, as we demonstrated, will induce a “T helper 2 type” (Th2) cytokine profile, wherein IL-10 and TGF-β are predominant. These do not activate macrophages and affect humoral immunity through production of non-complement-fixing antibody isotypes. After organ transplantation, activation of the Th2 pathway coincides with graft acceptance. Seemingly xenogenic implants are more biocompatible, which may reduce local complications. This remains to be clinically demonstrated. Actually, the mechanisms causing local complications in urogynaecology are still poorly understood. Prospective data collection on patient presenting with pain, infections, wound dehiscence, erosions or fistula (for as much as the latter can be discriminated), as well as recurrence rates, are very important objectives in any clinical study on the use of meshes for prolapse repair.

Conclusion

A growing number of aging women are presenting as candidates for surgical repair of genital prolapse. The scientific background of tissue changes and wound healing, both in hernia as well as prolapse patients; as well as the fate of implant materials used to reinforce repairs, is, therefore, a clinical need. The perfect implant material certainly is not available yet, but in general surgery there is now a consensus, funded on experimental data, that low-weight, large pore, monofilament materials are preferable. However, local complications may still occur and seem related to an increased foreign body reaction. Xenogenic implants were introduced in an effort to reduce these local complications, hopefully without compromising surgical results. Animal experiments show that the inflammatory response to these materials definitely is different. In tensiometric experiments, some products show comparable strength, but the longevity of these products, as well as functional results, remain to be demonstrated.

Acknowledgements Our own animal research has been supported by unconditional grants from Tyco Healthcare, Bard, and Cook. The companies may also have donated standard commercially available implants for experimental implantation. None had input into the surgical protocols, randomization tables, or control on data analysis nor its reporting. The authors have no financial interests in these companies.

References

Thomas A. Barbolt

Biology of polypropylene/polyglactin 910 grafts

Abstract The biological evaluation of polypropylene (PP)/polyglactin 910 grafts was reviewed including regulatory considerations, biocompatibility assessment, tissue reaction and integration, and infection potentiation of these synthetic materials used in urogynecological surgical procedures. The physical characteristics of the grafts including base composition, monofilament vs multifilament, and non-absorbable vs absorbable materials were compared. Grafts were implanted in rats to evaluate the tissue reaction and integration characteristics of the materials over time. Grafts were also implanted in mice and inoculated with Staphylococcus aureus to assess the potential for bacterial attachment and growth. The tissue reaction to PP/polyglactin 910 grafts was characterized by minimal to mild inflammation with some qualitative differences related to the physical construction of the different grafts. The tissue reaction to polyglactin 910 mesh was also mild but resolved after the material was absorbed 70 days post-implantation. The integration of PP/polyglactin 910 grafts by fibrosis with surrounding tissue was initially mild for all materials but decreased over time for the lightweight and multifilament PP-based grafts, including a graft with an absorbable polyglactin 910 component. Residual fibrosis was not observed for the graft constructed from polyglactin 910 alone. The biological performance of PP/polyglactin 910 grafts is dependent on multiple factors including the composition and physical construction of the base materials, the overall biocompatibility of the materials, particularly tissue reaction and integration of the grafts, and the resistance of the grafts to bacterial attachment and growth.

Keywords Polypropylene grafts · Polyglactin 910 · Animal models · Tissue reaction · Infection potentiation

Introduction

The focus of this presentation was on polypropylene (PP)-based meshes (polypropylene grafts), alone or in combination with absorbable elements (composites), and polyglactin 910 mesh alone. Regulatory considerations applicable to the development of these materials were reviewed, and physical characteristics and biocompatibility results were described.

Regulatory considerations

The International Standards Organization has set forth guidelines for the biological evaluation of medical devices in ISO 10993, part 1. The USA FDA has issued further guidance on the application of this international standard in the G95-1 Memorandum. In addition, the US FDA has more specific guidance for industry on the development of surgical meshes for 510(k) submission. The main emphasis of these guidance documents is to ensure patient safety. Materials characterization is a critical first step in the evaluation of the safety of a biomaterial. The manufacturing processes, including the use of extrusion aids, antioxidants, and/or stabilizers, are important because additives may leach out from the mesh once it is implanted in the body resulting in local irritation or systemic toxicity. It is possible that these extractables contribute to the different tissue reactions observed with various base materials. The degradation products released to the body are also important for polypropylene-based composite materials that are manufactured with absorbable components.
Biocompatibility testing is more than simply implanting the material and evaluating the local tissue reaction. For example, the potential for allergic reactions must be addressed. Moreover, systemic toxicity, cytotoxicity, and genotoxicity are important considerations with implantable meshes and must be evaluated to minimize risks to patients.

**Design requirements**

A key factor contributing to the success of a PP-based graft is the specific design of the device. In general, the “voice of the customer” is solicited from surgeons with routine clinical experience. Similar information but from a different perspective is obtained from clinical thought leaders at the forefront of their science and with extensive clinical experience. Both views are important for a product to be accepted, and the materials and accompanying procedures need to be useful to a broad spectrum of surgeons, not just to a few surgeons with unique surgical skills.

Animal models can be useful for evaluating tissue response after implantation and host integration of different materials, but assessment of functionality is limited because of obvious anatomical differences. However, cadaver models are anatomically proportioned and can be useful to help predict the success of a product in a clinical environment.

The ideal pelvic floor repair material needs to provide long-term support to result in a durable repair. Experience has demonstrated that materials that are totally absorbable will not work. The literature for hernia repair supports this view. There is now an interest to leave a minimal amount of “foreign” material behind after the surgical procedure for a more natural repair. This is one of the reasons that heavyweight meshes are falling out of favor. In addition, the material needs to be conformable. The pelvic floor is not the same as abdominal fascia so that less may be more. The amount of porosity, after a certain point, is less problematic. Almost any degree of porosity is acceptable, but some degree of porosity is necessary for sufficient integration. Elasticity in the range of 20–35% has been reported to match the compliance of surrounding tissues to avoid both extrusion of the material and patient discomfort. Lastly, materials should be “neutral” to the presence of intra-operative bacterial contamination; that is, infection is not exacerbated by the presence of the material. These characteristics are dependent on the nature of the mesh construction, e.g., multi-filament or monofilament, and base material characteristics, e.g., synthetic or naturally derived materials.

**Polypropylene-based grafts**

Commonly used meshes consisting only of polypropylene are Marlex mesh and Prolene polypropylene mesh. The mesh characteristics and the construction of the materials are different in terms of filament diameter and textile construction. An example of a new generation of lightweight, large-pore material is Gynecare Gynemesh PS Nonabsorbable Prolene Soft Mesh. The overall weight is approximately half that of Prolene mesh. Surgipro mesh is a relatively heavyweight mesh, but it is constructed with multifilament polypropylene yarn. The PP/polyglactin 910 polymer composites have much larger pore sizes and utilize absorbable polyglactin 910 yarn commonly used in absorbable sutures. The polyglactin 910 yarn is part of the knot construction and results in improved intraoperative handling characteristics. Otherwise, the mesh would be too flimsy to handle and position in the patient. Vypro II mesh is stronger than Vypro I as it has twice the amount of polyglactin 910. Vypro II also has an additional reinforcing monofilament to give a different handling characteristic and a greater overall strength once all of the polyglactin 910 yarn has been absorbed. There should still be sufficient strength left behind from the polypropylene component to give a durable repair after approximately 70 days when the absorbable components are gone from the surgical site.

**Polyglactin 910 grafts**

An example of a surgical mesh constructed with only absorbable polyglactin 910 yarn is Vicryl (polyglactin 910) knitted mesh. Experience in animal models has demonstrated that tissue support is temporary, and any connective tissue resulting after implantation will remodel over time. The reaction will eventually resolve with no remaining connective tissue.

**Points to consider**

The physical characteristics of materials are critical when matching a graft to a particular surgical procedure. The base composition is important, even for materials manufactured with polypropylene alone, because not all polypropylene is the same. Mesh constructed with monofilaments or multi-filament yarns will also have different characteristics that will result in different functionality. The use and extent of non-absorbable vs absorbable elements will relate to durability of the repair after absorption is complete. Flexural rigidity or elasticity is a characteristic that determines the potential for mesh extrusion and migration to adjacent structures after implantation. For example, a heavyweight stiff material would be non-compliant with surrounding anatomy while a lightweight, more flexible material would have a different result. The expected functionality of a material, coupled with a specific surgical approach, is what will ultimately drive design characteristics.

**Tissue reaction and integration**

Four different PP-based graft materials were implanted in dorsal subcutaneous pockets in female Long–Evans rats (5 rats/material/explant period) [1]. Marlex mesh was con-
sidered an example of a monofilament heavyweight material while Surgipro mesh was considered an example of a multifilament heavyweight material. Gynecare Gyne-mesh PS represented a monofilament lightweight mesh and Vypro I represented a composite mesh of monofilaments of PP and absorbable polyglactin 910 yarn. The tissue reaction and extent of integration with surrounding connective tissue was assessed macroscopically and by histomorphologic evaluation at 7, 28, 63, and 91 days post-implantation.

Similar results were observed macroscopically between the different graft materials. The formation of a “scar plate”, the bridging of fibrous connective tissue between elements of the mesh, was not observed even with the heavyweight materials. However, two of five sites implanted with Surgipro mesh had abscess and/or extrusion of the material. Clarification of this result awaits further investigation.

The tissue reaction to monofilament PP materials was characterized histomorphologically by chronic inflammation consisting of an accumulation of macrophages and some fibroblasts around the periphery of the monofilament. In some cases, scattered foreign body giant cells were observed. On the other hand, the tissue reaction to multifilament materials was characterized by a greater number of foreign body giant cells and fewer macrophages. This response is still considered a kind of chronic inflammation but a special category of chronic inflammation attributed to the small diameter of the filaments and the ability of foreign body giant cells to surround them. The tissue reaction and degree of fibrosis was rated on a scale of 1–4 depending on the severity and extent of the observation where 1 was minimal, 2 was mild, 3 was moderate, and 4 was marked. The intensity of fibrosis was considered a measure of integration of the mesh. It should be noted that a difference of a whole integer in the mean score would be necessary for a difference to be considered biologically meaningful. If consistent, differences of a half integer could be considered a biological trend. Overall, the tissue reaction to most of the materials was comparable and was considered to be stable at 91 days post-implantation. A mild degree of chronic inflammation or foreign body reaction was typically observed depending on the nature of the material (Table 1).

The first explant period evaluated was 7 days post-implantation when the trauma from the surgical procedure has subsided and the tissue reaction specific to the implanted material can be evaluated. For Marlex mesh, the tissue reaction was mild with increased density around the elements of the mesh (Fig. 1). At 91 days post-implantation (Fig. 2), the amount of cellularity was decreased in the connective tissue in the “pores” of the mesh and was confined to the PP elements of the mesh. However, there was some fibrotic “bridging” across the pores of the mesh from one PP element to another. A mild degree of fibrosis was observed at 7 days post-implantation, which was sustained over the 91-day study. This response was typical for a monofilament heavyweight PP mesh. The intensity of the tissue response to Surgipro mesh was similar at 7 days post-implantation, although qualitatively different because there were more foreign body giant cells around the small diameter filaments of the PP yarn. At 91 days post-implantation, foreign body giant cells were still present at the periphery of the bundles of PP filaments, but there was very little penetration of inflammatory cells to the interior of the knitted construction. In addition, decreased fibrosis was observed beginning at 28 days post-implantation, which was sustained for the duration of the study.

The tissue reaction to Gynecare Gynemesh PS was characterized by mild chronic inflammation at 7 days post-implantation.
implantation (Fig. 3). Although the overall tissue reaction was comparable at 91 days post-implantation, there was a reduction in cellularity in the surrounding connective tissue and a decrease in the amount of fibrosis in the mesh pores (Fig. 4). For Vypro I mesh, the tissue reaction was also mild at 7 days post-implantation and a reduction in the amount of fibrosis was also observed at 91 days post-implantation. For these lightweight materials, a trend of decreasing fibrosis was observed beginning at 28 and 63 days post-implantation, respectively, and was attributed to the overall decreased mass of these materials. For Vypro I, a contribution to the decreased fibrosis over time was attributed to the absorption of the absorbable component, leaving behind only the nonabsorbable elements of the mesh.

Gynecare Gynemesh PS was similarly implanted intramuscularly in the paravertebral muscle in rabbits to evaluate tissue reaction over time. Some myofiber necrosis resulting from trauma from the implantation procedure was observed but, overall, the tissue response was mild, and the reaction decreased over the 2-week study. Lastly, a multifilament absorbable polyglactin 910 mesh (Vicryl knitted mesh, style 7) was implanted between fascial planes overlying the gluteal muscle in rats, and the tissue reaction and absorption profile was evaluated histomorphologically. At 7 days post-implantation, the tissue reaction was characterized by mild foreign body reaction. This reaction decreased at 28 days post-implantation and was resolved as the material was absorbed at 70 days post-implantation. The connective tissue that was part of the early tissue response was remodeled and no longer present.

**Infection potentiation**

A standard mouse model was used to determine whether there were any differences in the ability of bacteria to attach and grow on synthetic and naturally derived implant materials [2]. It is well known that, in the presence of a foreign implant, it takes fewer bacteria to cause infection at the surgical site than without a foreign material present. Any bacteria present can also migrate along the device to contaminate other regions of the wound. Once a biofilm has formed on an implanted device, bacteria are more resistant to the natural defense system of the host as well as to the systemic administration of antibiotics.

Gynecare Gynemesh PS, a lightweight mesh, and Marlex mesh, a heavyweight mesh, were chosen to represent PP-based synthetic materials. SURGISIS, derived from porcine small intestinal submucosa, and DermMatrix, derived from porcine dermis, were chosen as examples of naturally derived collagen-based materials. Bilateral subcutaneous pockets were created in four mice for each material. Each material was implanted in sterile condition directly from the package using standard aseptic surgical technique. Soaking with antibiotics was not performed. Each of the eight implants was inoculated with 10⁴ colony-forming units of *Staphylococcus aureus*.
The implants were excised 4 days after inoculation, as this was determined from past experience to be the optimum time for bacterial growth. The implants were then ground and the bacteria enumerated using standard plate counting techniques. Both Gynecare Gynemesh PS and Marlex materials were “neutral” to the presence of bacteria (Fig. 5); that is, the number of bacteria recovered was generally similar to the initial inoculum. In contrast, the number of bacteria recovered from both naturally derived materials was generally three to four logs higher than the initial inoculum.

Conclusions

An understanding of the biological behavior of PP-based materials is a necessary prerequisite for the intelligent selection of biomaterials for use in pelvic floor repair. The tissue reaction to PP/polyglactin 910 grafts was generally mild with some qualitative differences related to mesh construction. For grafts constructed of polyglactin 910 alone, the tissue reaction, including fibrosis, resolved after the material was absorbed. The fibrotic response to the grafts was initially mild and, in general, decreased over time for the lightweight constructions and for the multifilament material. The number of bacteria recovered from PP-based materials was comparable to the initial inoculum while the number of bacteria recovered from naturally derived materials increased by three to four logs. The design of PP-based grafts, including the base materials, the textile construction, and physical characteristics will determine the overall biocompatibility and usefulness of the material.

References

Surgipro mesh: not all multifilaments are the same

Many surgeons believe that the use of multifilament implants should be avoided because they potentiate the development of infection. This is primarily based on historical reports involving very reactive materials. Standardized studies in a mouse model documented that multifilament sutures composed of silk, catgut, and Dacron were very reactive and greatly potentiated the development of infection [1]. Multifilament sutures of the synthetic polymers of nylon and polyglycolic acid were very weak potentiators of infection. In fact, when monofilament nylon sutures were compared to multifilament nylon sutures, there was no significant difference in their susceptibility to infection. Polypropylene sutures, like nylon, were documented to have a low level of infectivity. These results suggested that the chemical composition of the implant was more important than its physical structure in potentiating the development of acute infections; studies from other labs have confirmed these results [2, 3]. Other studies conducted over longer periods have indicated that multifilament sutures have a higher risk of potentiating infection than monofilament sutures [4–8]. In all of these studies, one of the sutures with the lowest risk of potentiating infection was monofilament polypropylene. No multifilament polypropylene suture is available to evaluate its infection-potentiating effect.

The clinical inertness of monofilament polypropylene was one of the primary reasons it was used for the construction of surgical mesh for the repair of abdominal wall defects and hernias. The polypropylene mesh with the longest history and most extensive use is Marlex (C.R. Bard). Marlex is composed of monofilament strands woven into a macroporous material. Because of the monofilament fibers, Marlex is relatively stiff. When Ethicon developed their polypropylene mesh, they used two filaments of small diameters to weave Prolene Mesh. US Surgical extended this concept and used multiple smaller fibers to weave their Surgipro Mesh.

In a controlled study in rats, the performance of the multifilament Surgipro Mesh was compared directly to that of the monofilament Marlex (Rodeheaver, unpublished). The animal model involved sewing a mesh patch into a 2×3-cm, full-thickness abdominal wall defect; for uncontaminated rats, healing strength, adhesion formation, and cellular reactivity were studied. For contaminated rats, after the mesh had been sewn in, the peritoneum was contaminated with known inocula of E. coli suspended in a sterile suspension of rat cecal contents. The results documented that there was no difference in the infectivity of the Surgipro Mesh. In addition, both mesh materials promoted moderate to extensive adhesion formation but were both very biocompatible with acceptable inflammatory responses. Marlex mesh had a faster rate of healing strength, but by 8 weeks, the Surgipro Mesh became significantly stronger.

These favorable results in animals have been confirmed by clinical results in humans [9]. In a review of 1,700 herniorrhaphies using Surgipro Mesh, the recurrence rate was only 0.29%. No mesh infection or rejection was observed. The mean follow-up time for these results was 5.3 years.

As the title of this article suggests, all multifilaments are not the same. As the previous animal and human results indicate, multifilament polypropylene mesh gives results that are similar to monofilament polypropylene mesh. However, multifilament polyester mesh (Mersilene) has been associated with significantly higher complication rates, including postoperative infection (16% vs 0–6% for other meshes) [10]. Leber et al. concluded, “polyester mesh should no longer be used for incisional hernia repair.”

Unfortunately, the clinical performance of Surgipro Mesh has been tarnished by the mesh classification system of Amid [11]. In that four-category classification system, Prolene and Marlex are classified as Type I—totally macroporous. Surgipro is grouped with Mersilene
as a Type III mesh—macroporous prosthesis with multifilamentous or microporous components. Thus, when Amid concludes “the risk of infection can be avoided by utilization of Type III and particularly Type I prostheses,” he is most likely giving the nod to Type I prostheses because of Mersiline’s higher infection rate, not because of any reports of infection with Surgipro.

Surgical meshes are also used by urologists and urogynecologists for surgical repairs in the pelvic area. Although these procedures often involve mucosal surfaces that are difficult to sterilize, the results, in general, seem to be as positive and uncomplicated as those of the general surgeons. Of particular interest is a modification of the urethral sling procedure for stress urinary incontinence created by Petros and Ulmsten [12]. In this procedure, a thin tape of surgical mesh is positioned to support the midurethra. The most common synthetic materials used in this procedure are the Surgipro Mesh tape used in the intravaginal slingplasty (IVS) approach or the Prolene Mesh tape used in the tension-free vaginal tape (TVT) procedure.

Although the risk of infection is a concern because of the difficulty of decontaminating the vaginal vault, it does not appear to be a major problem. In a series of 1,455 cases that involved the use of TVT in Finland, there was only a 0.8% incidence of wound infection [13]. In Austria, review of results on 2,795 cases that involved the use of TVT identified only 0.1% of cases where the TVT had to be removed for various reasons [14]. In a prospective study comparing TVT and IVS in 100 patients, no postoperative infections were reported [15]. In another prospective study, TVT, IVS, and another product were compared in 195 patients [16]. Again, no infection was reported for any of the products. These results would suggest that postoperative infection is not a problem with either the monofilament Prolene mesh (TVT) or the multifilament Surgipro Mesh (IVS).

In contrast to these reports, Bafghi, et al. have recently reported on their results with IVS in 149 women [17]. In 11 (7.5%) of these patients, retroperitoneal infection occurred between 4 and 17 months following surgery. In ten of these patients, the tape had to be removed despite antibiotic therapy. These results are concerning but need to be compared to the results obtained by others.

The use of a multifilament yarn to weave Surgipro Mesh provides greater flexibility and less stretch for better control. Even though Surgipro is more dense than other polypropylene meshes, it is still macroporous, with triangular pore dimensions of $244 \times 686 \mu$. This pore size allows for effective tissue integration and the migration of tissue defense cells such as neutrophils and macrophages. As mentioned earlier, when Surgipro was compared to Marlex, the healing strength of Surgipro at 8 weeks was greater than that of Marlex and the histology showed few differences in cellular reactivity (Rodeheaver, unpublished).

The issue of cellular reactivity between multifilament mesh (Surgipro) and monofilament mesh (Prolene) was evaluated in more detail in a pig model by Beets et al. [18]. They implanted each mesh type bilaterally in the preperitoneal inguinal areas of six pigs and harvested the samples at 3, 6, and 12 weeks. At each time period, the numbers of foreign body giant cells were greater at the Surgipro interface than at the Prolene interface. The numbers at the Surgipro interface were 3.5–5.4 times greater than those at the Prolene interface, and probably reflect the difference in surface area exposed by the multifilament product. The clinical relevance of this observation is not known.

Another histological study was prompted by a unique clinical case study where a woman had tapes of both types removed 2 years following surgery [19]. In this analysis, the tissue reaction associated with the Surgipro Mesh compared to Prolene contained foreign body giant cells of smaller size, fewer foci of inflammatory leukocytic exudates, and thicker and more compact collagen I fibers around the yarns. Again, these results are due to differences in physical structure, and the clinical relevance is unknown.

In conclusion, the existing evidence suggests that both types of polypropylene mesh, IVS and TVT, are very effective and very safe synthetic materials for use in pelvic surgery. The inferences that the multifilament IVS material may be more susceptible to infection are based on other multifilament materials, such as Mersiline, and do not relate to the performance of IVS, because not all multifilament materials are the same.

References


Abstract The goal of this manuscript is to discuss the utilization of InteXen graft material, which is a natural, biocompatible matrix. There is unfortunately little data concerning this material; so, many of the concepts introduced in this manuscript are theoretical. We will discuss the rationale behind using InteXen as the biologic material of choice.

Keywords Sling · Stress urinary incontinence · Vagina · Bladder · Urethra · Graft

InteXen biocompatibility

Before its introduction in female pelvic surgery, porcine dermis met the industry standards for biocompatibility testing, and it has been safely utilized in wound care as well in facilitating wound healing in burn injuries. InteXen (American Medical Systems) is aseptically processed and terminally sterilized. It is an acellular material that should provide minimal risk of rejection or inflammatory response, and it should not prolong the healing process. In addition, InteXen is a non-cross-linked, structurally intact collagen matrix, which theoretically should facilitate host tissue integration, both enhancing and strengthening graft performance.

Preparation of porcine dermis

After the fat and epidermis is removed from the harvested hide in experimental pig animals, a sequential process of soaking in various detergent solutions is completed. First, povidone–iodine and hydrogen chloride solutions are used to deactivate bacteria and viruses by rupturing the cell wall. Then, hydroxide solutions are used to remove the cellular debris and inactivate prions and viruses. The processed tissue is then further sterilized via gamma irradiation.

Cross-linking

InteXen is not cross-linked as compared to Pelvicol (CR Bard) which, due to the crosslink, will stabilize the implant to prevent collagenase destruction and render the collagen nonresorbable. Pelvicol is dicyclohexylmethane-4,4-diisocyanate (HMDI) cross-linked, three amino acids in triple helix cross-linked with hexamethylene di-isocyanate. The HMDI cross-linking process has fewer cytotoxic effects than the more commonly utilized stabilization techniques with glutaraldehyde [utilized in Contigen (CR Bard)]; therefore, it may have less “leaching” into the host. Cross-linking of collagen fibrils is depicted in Fig. 1.

A potential advantage of cross-linking is the stabilization of the collagen implant to prevent tissue destruction by inflammatory cells. In addition, there should be less of an immune response utilizing the HMDI cross-linking process as compared to other commonly utilized cross-linking procedures. A potential disadvantage is that this process may not completely eliminate the cytotoxic response. Thus, when cross-linking this material, a cytotoxic response by the host may occur. Gandhi et al. [1] noted a variable host response to cross-linked porcine dermis and, in some instances, noted significant graft degradation. A cross-linking process may actually ultimately prevent cellular infiltration by the host. This lack of cellular infiltration would retard remodeling and ultimately lead to graft breakdown. Thus, the major concern about cross-linking this material is that it could inhibit cellular in-growth. Some of the HMDI material could elicit a host response, which might increase the risk of host reaction to this material. To date, there is no control data that examines the issue of cross-linking vs non-cross-linking in a similar model. Figure 2 illustrates the difference in the appearance of the collagen matrix in InteXen and Pelvicol.
InteXen LP is a new graft, which is processed exclusively by American Medical Systems. It will be fully distributed by the end of 2005. The tissue processing procedure differs from InteXen in that InteXen LP is not irradiated; it is terminally sterilized by ethylene oxide then lyophilized. The rationale is that these alterations in processing are going to ultimately promote host tissue infiltration. InteXen LP macroscopically looks similar to InteXen, but the processing technique is quite different.

**Background: animal study**

The differences of these materials have been examined in a rat model, where tensile strength, graft integrity, and host infiltration were examined.

**Study methods**

Three porcine dermis reconstructive materials were implanted in a controlled manner in male Sprague–Dawley rats. The three porcine dermis grafts studied were InteXen LP, InteXen, and Pelvisoft (CR Bard). A 2-cm-diameter suture loop of polypropylene was tied into the end of each sample before implantation. This was for the purpose of suture retention testing, which was carried out after explantation. Two different graft materials were implanted into the ventral subcutaneous space just lateral to the midline in each rat. The rats were killed and materials were explanted at the controlled endpoints: 16, 49, and 84 days. Gross necropsy photos were taken, and the specimens were biomechanically tested for suture retention strength. An extensive and systematic histological examination of each specimen was then carried out.

Tensile strength testing was performed with an Instron machine (Fig. 3). The graft material (and muscle wall where the material was not separated from the graft) was gripped in lower jaw. The suture loop (excised from the pannus layer of tissue) was gripped in upper jaw, and it was pulled at a control rate with a crosshead speed of 2 in./min. Failure was defined as the force at which the suture loop was tearing from the material. Seven to eight samples for each graft material at each timeframe were utilized.

The suture retention data shows that, at time zero, the suture pull strength of the AMS InteXen graft was significantly higher than the Bard Pelvisoft graft. Over time, the tensile strength of the Bard Pelvisoft was constant and actually increased by the end of the study period. After each explant time and at the endpoint of the study, the tensile strength was similar in all groups. There was no statistically significant difference, as compared by ANOVA analysis (p<0.05) (Fig. 4).

![Fig. 1 Cross-linking of collagen fibrils](image1)

![Fig. 2 a InteXen (×10) and b Pelvicol (×10)](image2)

![Fig. 3 Tensile test with Instron machine](image3)
Capillary count methods

In an attempt to standardize the process, two pathologists looked at each specimen under ×400 power field (Fig. 5a). They counted three fields per sample. The sample spanned from the muscle to the skin side of the graft. The number of capillaries that contained erythrocytes was counted. There were six to nine fields per material per timeframe analyzed. The AMS InteXen LP had the most vascularization at 84 days \((p<0.03)\). There was an interesting observation in the Pelvisoft material. There was a difference in the capillary count based on location of the pores of the graft material. Within the pores, there were more capillaries than within the graft material only. When comparing fields with graft material only (no pores), the capillary count was higher in the InteXen and InteXen LP than in Pelvisoft.

Fibroblast count methods

A similar technique was utilized looking at fibroblast counts under ×1,000 field (Fig. 5b). Four fields per sample were utilized, and the number of fibroblasts was counted. There were approximately 8–12 samples per material per timeframe. At 84 days, the number of fibroblasts was fairly uniform in all materials (Fig. 5c). Even without the pores in the Bard Pelvisoft, the fibroblast count was similar. There was a peak in fibroblast count noted at 16 days in all materials, probably secondary to the inflammatory response. The 16-day peak in fibroblast count has been seen in plastic surgery literature, so it is not a novel finding.

Cellular infiltration methods

When the aspect of host cellular infiltration is examined, there are several areas of interest. First, it is important to determine if there are host cells infiltrating the materials and, secondly, one should identify where this cellular infiltration occurs in the grafts. To answer these questions, a grid model was employed to attempt to examine the issue of cellular infiltration in various regions of the graft. The graft was divided into four regions. The area of the graft adjacent to the muscle was named outer area 1. The area of the graft adjacent to the skin of the animal was named outer area 2. These outer regions represented the superficial surfaces of the graft. The central areas were named central area 1 and central area 2 and represented the inner aspects of the graft, not directly adjacent to host tissue. This facilitated determination of cellular infiltration into the center of the graft compared to the peripheral surfaces of the graft. An infiltration ratio was calculated by examining the combined number of cells in the two central fields expressed as a percentage of the total. If one only examines cell count and not cellular location, it may not be known if infiltration is occurring in the central portion of the graft. There were only two to three samples per material at the 84-day timeframe. When the cellular infiltration data is examined, the Bard Pelvisoft had the highest cellular infiltration within the pores at early time intervals. The InteXen LP had the most even distribution of cells within the material. The conventional InteXen graft and the Bard Pelvisoft graft without pores had lower cellular infiltration within the central areas of the grafts.

Material integrity methods

A 30-square grid was utilized to measure the graft materials (Fig. 6). When each graft was implanted, it filled the entire grid, based on the size of the graft, which was controlled at the time of implantation. The grid was then re-applied to the graft at the time it was explanted. The number of squares that was occupied by the graft was then counted to determine the surface area. A complete graft at time zero, at the time of implantation, completely occupied the whole grid (30 squares). The integrity of the graft at the time it was explanted was calculated as the percentage of the squares remaining out of the 30. A major concern with this method is that this technique measures surface area only.
and not thickness as well. Thus, one can contend it is a measure of surface area and not of graft integrity, which may influence graft strength. In the absence of a standardized model, this technique needs further review.

The Pelvisoft and the InteXen LP had a higher degree of surface area persistence at the end of the 84-day timeframe. The InteXen LP seemed to have the most manual integrity or surface area at the end of the 84-day timeframe compared to the more conventional InteXen graft (Fig. 7).

**Conclusion**

This study appeared to show that Bard Pelvisoft and AMS InteXen LP maintain integrity to a similar degree. These materials seemed to facilitate cellular ingrowth, and the cells are evenly distributed throughout material. The porous nature of the Pelvisoft enhanced tissue penetration into all areas of this graft material, whereas the InteXen LP had actual graft infiltration in all areas. These materials do seem to facilitate both vascularization and remodeling in this particular model, as they contained the highest number of capillaries and competitive fibroblast counts.

**Animal studies and InteXen**

Our literature review found very few studies with other animal models using InteXen materials. The group from the Mayo Clinic led by Daniel Elliott presented several laboratory studies. They initially reported that cadaveric fascia and porcine dermis have a higher degree of inflammatory rind and inflammation, when compared to autologous fascia [2]. Cadaveric fascia and porcine dermis had decreased stiffness and tensile strength compared to autologous fascia [3]. These findings were observed in a rabbit model and not in a rat model as in our study. The authors concluded that the higher degree of inflammation in graft substitutes may contribute to higher degrees of graft deterioration over time.

**Human studies**

There are few human studies demonstrating efficacy and safety with porcine dermis. Gomelsky and Dmochowski examined a group of 70 patients with a mean follow-up of 2 years looking at porcine dermis interposition graft (InteXen) for repair of high-grade anterior compartment defects. Ninety-one percent were “dry” after the sling procedure, and nine patients had recurrent cystoceles (six Ba=0 and three Ba=+2). One patient had a vaginal wound separation. These authors concluded that the porcine dermis represents a successful and safe treatment option for repairing high-grade anterior compartment defects [4].

**Porcine dermis: potential pitfalls**

It has been reported that using porcine dermis encapsulation can occur, and this has been seen with Pelvicil [5] and InteXen [6]. There are some unknown factors that may inhibit remodeling in some hosts and will have impaired and/or variable host response [1]. More studies are needed to determine whether it is cross-linking, terminal irradiation, or some process related to the porcine dermis.

**InteXen and InteXen LP: future study**

This is an area where much more animal data is needed. There is very little animal data looking at non-cross-linked porcine dermis. There are several clinical protocols in various stages of development, particularly looking at graft interposition in prolapse surgery. However, more con-
trolled animal data is needed. In addition, much information can be obtained from a systematic examination of material explanted from patients who require re-operation after graft implantation procedures. This data may provide meaningful insight into the host–graft interaction.

References


Tissue engineering a clinically useful extracellular matrix biomaterial

Abstract Implantable biomaterials are one of the most useful tools in the surgeon’s armamentarium, yet there is much room for improvement. Chronic pain, tissue erosion, and late infections are just a few of the serious complications that can occur with conventional, inert materials. In contrast, tissue-inductive materials exist today. Combinations of biologically important molecules for directing cell growth and providing structural stability can be found in naturally occurring extracellular matrices. These “soft-tissue skeletons” of Mother Nature can be harvested, processed, and provided in a medically safe and biologically active form for repairing many different tissues in the human body. The future of surgical practice may well be determined by how well these new implant materials recreate the tissues they replace.

Keywords Tissue engineering · Interactive biomaterials · Remodeling materials

Introduction

Tissue engineering is the discipline of medical science dedicated to recapitulating living tissue structure and function using fundamental principles of both engineering and biology. Although a relatively new discipline, tissue engineering is likely to affect many aspects of medicine. The scope, methods and promise of tissue engineering may differ from investigator to investigator, but most experts agree that its foundation relies on three basic elements: cells, scaffolds, and signals [1]. These three elements are used in combination to produce tissue-engineered products (TEPs) that can be used to treat human disease (Fig. 1). Perhaps the first and most well known TEP is “synthetic skin” or “living skin equivalents.” Three such products are currently available today in the US market for skin wound healing.

Arguably, TEPs do not have to contain all of the three foundational elements to be either beneficial to the patient or to be considered to be “tissue engineered.” If the ultimate goal of all TEPs is to recapitulate living and functional tissue within the patient, then the definition of a TEP may be considerably broader than just living cellular constructs. In fact, some of the key questions often posed by researchers in this field are:

“How can we stimulate the patient to regrow his or her own tissues?”
“Must all three elements be delivered to a wound or surgical site?”
“Why can’t the patient provide one or more of the key elements?”

This paper will address these questions in light of recent developments in tissue-engineered biomaterials for surgical use.

Natural extracellular matrix biomaterials

Structure and composition

The extracellular matrix (ECM) is both complex and tissue-specific. Its components are tailored to, and respond to, the physiologic requirements of the local tissue. The ECM derived from the basement membrane of the urinary bladder or the blood vessels is vastly different from the ECM derived from the submucosa or dermis. For example, ECM derived from the basement membrane provides
structural support for the endothelium or epithelium and consists primarily of laminin, entactin, heparin sulfate proteoglycan, and type IV collagen. In contrast, submucosal ECMs and dermal ECMs, adjacent to structures rich in epithelial cells undergoing constant and rapid turnover, supply the epithelial tissue with nutrients through a large vascular network and provide a structural support upon which the cells reside. These ECMs consist of a wide variety of proteins and carbohydrates including type I collagen, glycosaminoglycans, glycoproteins, and growth factors, and are less structurally dense than basement membranes. Although many people have made valiant attempts to synthesize ECMs [2, 3], only Mother Nature has done it with such eloquence and specificity.

Each of the components of the ECM serves specific functions. Collagen provides strength to the ECM, which is then easily translated into tissue integrity. Glycosaminoglycans, glycoproteins, and growth factors provide context that can be used by the residing cells to help maintain the homeostatic environment, assist in orchestrating tissue inflammation and repair, and modulate the localized innate immune response. To extract or purify collagen from an ECM is to harvest a frame with no finish. Likewise, to extract the bioactive proteins without collagen is to harvest finish without frame.

Natural ECMs are very fragile entities with respect to composition. Even though they exist in various tissues populated by the very cells that created them, removal of those cells during the production of a TEP often proves difficult without irreversibly altering the structure and/or composition of the matrix. While cells can be extracted from the submucosa without grossly altering the natural structure and composition of the ECM, even relatively weak solvents, organic salts, rogue enzymes, and contaminating microorganisms can significantly alter ECM composition. [4, 5] Additionally, harsh chemicals, such as detergents and cross-linkers, can completely remove or render the signals present “unreadable” (Cook Biotech, unpublished). When starting from natural tissues, only a complete isolation of an ECM can provide both a scaffold for tissue regrowth and the needed signals for cellular instruction in the same ratios, conformations, and spatial orientations as those present in nature.

ECM materials can be isolated from a number of various tissues. The submucosa of the small intestine (Fig. 2), the submucosa of urinary bladder, the pericardium, the basement membrane of the liver, the decellularized Achilles tendon, arteries, veins, the ureter, and the renal capsule have been purified and used as sources of ECM materials. Of these tissues, the tissue graft material derived from the small-intestinal submucosa (SIS) of pigs is the most extensively characterized.

SIS is a plentiful and commercially available ECM that has been proven to retain much of the original structure and composition of the natural, intact ECM [6–9]. SIS is obtained from the tunica submucosa of the small intestine and is the layer of connective tissue arranged immediately under the mucosa layer. It is a 100- to 200-μm-thick ECM. In the living intestine, the submucosa supports the mucosal structures and is secreted and maintained by connective tissue fibroblasts. This tissue layer supports the growth and differentiation of the rapidly renewing and metabolically active mucosal and glandular cells, while maintaining a connective tissue structure that gives the intestine its integrity and strength. The commercial preparation of SIS is an acellular matrix with much of the chemical and structural components of the ECM left intact. In a wide variety of tissue beds, animal models, and human clinical studies, SIS has proven to be an extremely useful biomaterial for tissue growth and structural repair. When implanted surgically, it stimulates angiogenesis and connective and epithelial tissue growth and differentiation, as well as deposition, organization, and maturation of ECM components that are functionally and histologically appropriate to the site of implantation [6].

Fig. 1 The basic elements of tissue engineering are scaffolds, cells, and signals. These elements can be combined in various ways to make tissue-engineered products.

Fig. 2 A scanning electron micrograph of the cross-section of a single sheet of freeze-dried small intestinal submucosa. Clearly, there are pores and interstices present that can serve as entry points and migration paths for invading cells.
Mechanism of action

Surgical biomaterials “act,” at least as a start, by means of the surgeon’s placement. Over time, however, the fate of a given implant can be very different, depending on its structure and composition. For example, a monofilament, absorbable suture will undergo rapid degradation after implantation and leave nothing but the tissues it attached behind. This complete degradation can be a positive feature for some medical implants, but for recapitulating living tissue structure and function, this transient fate is not adequate.

The very notion of the empty degradation of a scaffold is counter to the meaning of the word “scaffold.” Scaffolds can be degradable or permanent, but by definition, a scaffold must support the ingrowth of cells into the implant. Nondegradable scaffolds allow or “conduct” cells into their matrix, so they can be said to be tissue conductive. Scaffolds that both conduct cells into their matrix and “induce” them to proliferate, differentiate into different cell types, and deposit a new matrix, can be said to be tissue inductive.

Tissue-inductive scaffolds, such as SIS, undergo controlled degradation simultaneously as they induce tissue reconstruction (Fig. 3). To say that these materials simply degrade and “go away” is to only tell half of the story. The other half, and perhaps the most important half, is the induced reconstruction of new patient tissue in a site-specific and intelligent way—dubbed “smart tissue remodeling” [8]. The ingrowth of fibroblasts, the rapid angiogenesis, the differentiation of myofibroblasts, and the resurfacing by epithelial cells all speak to the intelligent way in which SIS becomes “self” to the patient.

Many of today’s other implant materials undergo strong localized inflammation, foreign body recognition, and walling-off or encapsulation. This occurs with both purely synthetic materials and highly adulterated (stripped or cross-linked) natural materials. These materials are not scaffolds. They are long-term foreign bodies that may or may not degrade to a future point of failure, may eventually erode through adjacent tissues, or may get infected without the patient’s ability to defend them.

Importance of bioactivity

Although natural ECM is very complex in composition, it was only recently considered that such a material could be processed for medical purposes while maintaining such complexity. Likewise, natural ECM bioactivity, which helps maintain tissue homeostasis and orchestrates tissue inflammation and repair through the release of growth factors, is a foreign concept in the literature on biomaterials. Bioactivity is not a characteristic typically used to describe surgical biomaterials, but it is important for tissue-inductive scaffolds and can be easily measured using cells in culture or by measuring active blood-vessel growth in animal models.

Much of the inherent bioactivity of the natural ECM is due to the presence of growth factors within the matrix. Generally, growth factors are thought to be fragile proteins that undergo degradation almost immediately after they are secreted into the ECM by the cells that produce them. Growth factors are heat-labile proteins; they are easily cleaved by ubiquitous enzymes and are very sensitive to hydration in their purest forms. For example, when delivered to a patient in purified form to stimulate tissue repair, growth factors are rapidly degraded if their release is not rigorously controlled. Similarly, when growth factors are subjected to sterilization procedures in a nonbound state, their bioactivity is rapidly lost. Several studies have indicated, however, that growth factor stability can be dramatically enhanced when the growth factor is bound to a carrier matrix, such as demineralized bone [10] or chitosan fleece, [11] or if the growth factor remains in the matrix, bound to its binding proteins or to heparan sulfate proteoglycans. In the case of the basic fibroblast growth factor (FGF), its stability is markedly increased in the presence of some zwitterionic detergents, heparin, or other polyanions present in the ECM.

Engineering bioactivity into a TEP is therefore a process that can be strategically controlled during product manufacture. For example, growth factors can be tethered into medical biomaterials as a means of stabilizing them from degradation immediately after surgical implant. Delivering sustained bioactivity from a medical biomaterial is possible because linking molecules used to tether the growth factors to the device can be designed with different degrees of thermal and/or enzymatic sensitivities. For medical biomaterials isolated from natural ECM, this tethering process has been solved by Mother Nature. In the natural ECM, growth factors are deposited by cells and bound to stabilizing binding molecules, such as heparin, glycosaminoglycans, or other proteins. For example, transforming growth factor beta (TGF-β) is secreted by cells as a propeptide and is stored in the ECM as part of a large latent complex bound to a latent-TGF-β-binding protein. TGF-β activation occurs when protease-sensitive
regions of the complex are cleaved, releasing active TGF-β into the extracellular milieu.

SIS has been shown to retain a wide variety of natural ECM components even after decellularization, viral kill, lyophilization and sterilization. Hodde and Hiles showed that extractable and bioactive FGF-2 is retained in processed and sterilized SIS even after 24 months on the shelf [7]. Further, this study was done with material that had undergone a complete viral kill program as described by Hodde prior to lyophilization and sterilization [12]. SIS has also been shown to contain fibronectin in significant quantities [9], a known, highly bioactive molecule important to cell attachment and ECM signaling [13].

**Why remodeling?**

Long-term repair, not a long-term foreign body

Implantable plastic, metal, and other nonresorbable-material-based devices are considered permanent implants, but are they truly biocompatible? And if they get infected or erode through adjacent tissues, are they truly permanent? In any case, these materials are recognized by the patient’s cells as foreign bodies during their entire period of implantation. In fact, Klinge et al. showed that polypropylene mesh used for hernia repair elicits a chronic foreign body response in humans that remains measurable for many years [14].

An actively remodeling biomaterial can avoid long-term complications associated with a chronic foreign body response. Instead of acting as a nidus for constant immune cell activation, a repository for persistent bacterial contamination, or a source of eventual implant loosening and resulting implant failure, tissue scaffolds can incite cellular adaptation, ingrowth, and repopulation that become part of the natural wound-healing process. Inflammation occurs but is transient as part of the normal sequence of tissue repair events. Both animal and human studies have shown that remodeling biomaterials, such as SIS, are completely integrated into the host tissue, are turned over, and no longer act as foreign bodies [15–17]. Avoiding long-term pain is also important, and Ansaloni et al. showed that pain scores after inguinal hernia repair were significantly lower in patients receiving SIS than in patients receiving polypropylene mesh [18].

An unexplained complication that occurs often with many biomaterials and can interfere with tissue remodeling is seroma. Some researchers have reported near 100% seroma formation with microporous materials; still, others point to the overuse of electrocautery as a causative factor. The pathogenesis of seroma is not known, but it has been reported for virtually all materials, including ECM biomaterials. Very little research has been published on the mechanisms of seroma formation, but it is clear that an untapped serous collection has the potential to separate a remodeling material from its source of cellular ingrowth.

Of course, of prime interest to the surgeon is whether these newly formed tissues truly affect a long-term structural repair. Even the most tissue-inductive scaffold will fail as a viable medical implant if the repair that it creates has no longevity or permanence. Long-term, 2-year studies in dogs have shown that natural ECM biomaterials maintain their strength over time even as they become incorporated and remodeled into naturally functioning fascial tissue. In human clinical use, Franklin et al. have shown that SIS-repaired ventral hernias show good outcomes for more than 2 years, with the formation of normally appearing tissues [19]. Further, Rutner et al. have shown that SIS placed in the urethral sling location provides successful outcomes (significantly improved continence) for more than 4 years without long-term complications [20].

Scaffolds with signals can recapitulate tissues

There is a large and growing body of evidence that in situ tissue engineering, where the patient receiving the implantation provides a large part of the elements for success, is both possible and desirable for many tissue types. Specifically, the implantation of scaffolds with signals but void of cells provides for the growth of new tissues that resemble vascular conduits, bladder walls, collateral ligaments, and other tissues from the macroscopic level to the microscopic level.

More than 300 publications now exist relating to the medical utility of SIS; these include studies in vitro, in animal models, and in a variety of human applications. Site-specific remodeling occurs and has been well documented by histology in the canine aorta [16], the rat dermis [21], the canine urinary bladder [22], and even full-circle as a graft for bowel reconstruction [23].

![Fig. 4 Successful outcomes from the use of implanted medical products depend on a balance between the procedure used, the patient health and the product applied. Tissue-engineered products often rely even more heavily on this delicate balance for best performance](image)
Success is possible, but not assured

As more is learned about engineering replacement tissues, harnessing the body’s ability to heal itself appears to be strongly affected by three key facets: the product, the patient, and the procedure (Fig. 4). For example, even the best designed prosthesis can be implanted with a technique that virtually assures its failure; even the most advanced graft, like an organ transplant, cannot survive if the patient’s metabolism, immune acceptance, or physicality cannot support it; and even the best patient and the best surgeon cannot overcome a fundamentally flawed product.

Tissue-inductive biomaterials are no exception to this paradigm. Material design, surgical technique, and patient health all play crucial roles in the success of the grafts. As a result, some patients and some surgical approaches are not suitable for combination with tissue scaffold materials. It is clear, however, that better tissue attachment, better access to blood supply, a metabolically active subject, and a “use it or lose it” approach are all beneficial to tissue remodeling and successful outcomes with these materials.

**Conclusions**

ECM including cellular signals but void of cells can be an effective scaffold for tissue repair. SIS is an acellular, complex ECM that contains many noncollagenous components, an open structure, measurable bioactivity, and a proven track record for serving as a tissue-inductive scaffold both in animals and in humans.

**References**

Evaluation of a unique bovine collagen matrix for soft tissue repair and reinforcement

Abstract Veritas® Collagen Matrix, a product of Synovis Surgical Innovations, is derived from bovine pericardium. It can be used for a number of applications including body wall repair and replacement. In this study, we evaluated its efficacy as an adhesion barrier in a rabbit model of uterine horn surgery. When Veritas® was placed on the uterine horn stump it reduced the incidence of adhesions by 50% (n.s.) compared with untreated controls. Histologic analysis of recovered material showed that the surface was covered with a monolayer of mesothelial-like cells. In addition, there was an infiltration of host cells into the matrix of the product, which suggests a replacement of the material with host tissue.

Keywords Antiadhesion barrier · Animal model · Uterine horn

Introduction

The Surgical Research Laboratory at Tufts, New England Medical Center has worked with the medical device industry as a subcontractor for a number of years in evaluating materials for soft tissue repair and reinforcement. Many of these materials have been synthetic, but in recent years, there has been increased interest in materials derived from biological materials. One of these materials is Veritas® collagen matrix (Synovis Surgical Innovations), which is derived from bovine pericardium. The use of bovine tissue has been associated with bovine spongiform encephalopathy (BSE) and variant Creutzfeldt–Jakob disease (vCJD) transmission. However, the safety of Veritas® is evidenced by the facts outlined in Table 1.

Abdominal and/or pelvic adhesions arising from a variety of conditions including endometriosis, inflammatory disease, and abdominal and/or gynecologic surgery are major contributors to chronic pelvic pain, intestinal obstruction, and infertility. The incidence of adhesions is quite high (55–95%) in women after laparotomy. As knowledge related to the pathophysiology of adhesion formation has increased, so has interest in developing adjuncts to surgery that will reduce the incidence of adhesions and their associated clinical sequelae.

Veritas® may possess “antiadhesive” properties (i.e., it does not adhere readily to tissues of the chest wall or abdomen under conditions that promote such adherence).

Material and methods

Unilateral hysterectomy surgery and subsequent implantation of Veritas® as an “antiadhesion barrier” was performed on adult female rabbits (2.5–4.5 kg weight). A total of five (5) animals underwent hysterectomy without implantation of Veritas® and served as controls. In an additional ten (10) animals, Veritas® was positioned over the hysterectomy site and was held in place by a suture placed at each end. In all cases, the abdomen was closed and animals were monitored for a period of 14, 15, or 29 days as indicated in Table 2. There were no postoperative complications in any of the animals used in this study.

Results

Assessment of adhesion formation

At the time of explantation, a digital video record of the implant site was created. Careful dissection was accompanied by an assessment of the location and severity of adhesions observed in each animal. Scoring of adhesion severity follows the Adhesion Scoring Group system published in 1994 (grade 0 = none, grade 1 = filmy, avascular adhesions, grade 2 = dense and/or vascular, grade
3 = cohesive adhesion). After assessment, each implant was surgically removed, along with attached adhesions when possible, and processed for histologic analysis.

As seen in Table 2, adhesions, frequently of grades 2 and 3, to bowel, bladder, and uterus were predictable results of hysterectomy in five (5) control animals when assessed after 14, 15, or 29 days. Conversely, of the ten (10) animals undergoing hysterectomy followed by implantation of Veritas® as an antiadhesion barrier, only five (5) were observed to have adhesions after 15 or 29 days. Of these, only two were classified as grade 3 adhesion; one of these was peculiar in that the implant was adherent directly to the peritoneal aspect of the abdominal incision. A sixth animal (USDA #B3) did exhibit a single adhesion, to the uterine stump which had not been completely covered by Veritas® at the time of implantation.

There was not a statistically significant difference in adhesion formation between the control animals (100%) and the Veritas®-treated animals (50%). However, this study did produce a large amount of materials for histologic analysis.

Table 2  Adhesion formation after rabbit hysterectomy in control and Veritas®-treated animals

<table>
<thead>
<tr>
<th>Time</th>
<th>Adhesions</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td></td>
</tr>
<tr>
<td></td>
<td>USDA #B1 14 days +</td>
<td>Adhesion of bladder “fat” to incision and mesentery to stump of uterine horn (no grade recorded)</td>
</tr>
<tr>
<td></td>
<td>USDA #B2 14 days +</td>
<td>Adhesion of bladder fat to suture line, of bladder to uterine stump, and mesentery to uterine stump (all grade 3)</td>
</tr>
<tr>
<td></td>
<td>USDA #39 29 days +</td>
<td>Adhesion to small bowel (grade 3), adhesion to ovary (grade 3), and adhesion to bladder (grade 1)</td>
</tr>
<tr>
<td></td>
<td>USDA #47 15 days +</td>
<td>Adhesion to small bowel (grade 3) and adhesion to bladder serosa (grade 2)</td>
</tr>
<tr>
<td></td>
<td>USDA #53 29 days +</td>
<td>Adhesions to fat (minor) and adhesion to ovary (grade 2)</td>
</tr>
<tr>
<td></td>
<td>Total 5/5=100%</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Treated</td>
<td></td>
</tr>
<tr>
<td></td>
<td>USDA #B3 14 days −</td>
<td>Note: The implant did NOT completely cover the uterine stump</td>
</tr>
<tr>
<td></td>
<td>USDA #B4 14 days +</td>
<td>Adhesion of bladder to implant (grade 3), bladder to mesentery (grade 1)</td>
</tr>
<tr>
<td></td>
<td>USDA #40 15 days +</td>
<td>Adhesion of implant to incision (grade 3)</td>
</tr>
<tr>
<td></td>
<td>USDA #41 15 days −</td>
<td></td>
</tr>
<tr>
<td></td>
<td>USDA #42 15 days −</td>
<td></td>
</tr>
<tr>
<td></td>
<td>USDA #48 15 days −</td>
<td></td>
</tr>
<tr>
<td></td>
<td>USDA #49 15 days +</td>
<td>Adhesion to bladder serosa (no grade)</td>
</tr>
<tr>
<td></td>
<td>USDA #50 15 days −</td>
<td></td>
</tr>
<tr>
<td></td>
<td>USDA #51 29 days +</td>
<td>Adhesion to bladder (grades 1–2), Adhesion to cecum (grade 2), Thin fibrous adhesion to large bowel</td>
</tr>
<tr>
<td></td>
<td>USDA #52 29 days +</td>
<td>Adhesion to mesentery (grade 2), small bowel adhesion to serosa (grade 2)</td>
</tr>
</tbody>
</table>

| Total 5/10=50% |
Histologic analysis

H&E stained sections of Veritas® explants reveal that this material is composed of a mixture of amorphous and fibrillar collagen of limited porosity (Fig. 1). The material itself is largely acellular except where cells from the surrounding tissue (e.g., fibroblasts and/or mesothelial cells) have repopulated the matrix or surface. It is important that each of the explants appeared to have a complete (or nearly complete) covering of host cells that in some instances was a single layer thick (i.e., a monolayer). While it is not possible to identify these cells histologically, they may be mesothelial cells that have repopulated the surface of the implant by direct migration from surrounding tissues. It is important that mesothelial cells possess fibrinolytic properties similar to those of vascular endothelial cells and this may contribute to the relative lack of adhesions observed on Veritas®.

Infiltration of host cells, most likely fibroblasts, was a consistent histologic finding and is illustrated in Fig. 2. Individual host cells, identified by their darkly stained nuclei, can be seen within the implant. The depth of infiltrating cells is indicated by white arrow. A braided suture can be seen in the upper-right hand corner of this micrograph. This host cell infiltration of the implant, which is a critical component of its eventual “remodeling”, was most pronounced at the lateral edges of the implant where natural separation of the collagen bundles offers less resistance to cell migration (Fig. 3). It is worth noting that no implant fragmentation or degradation was observed.

Fig. 1 H&E stained section of Veritas® explant 14 days post-implantation. Note the amorphous, acellular nature of explant and the “lining” of host cells—at some points a single layer thick—that represents a continuation of the visceral peritoneum. Original magnification =×200

Fig. 2 H&E stained section of Veritas® explant 14 days post-implantation. At the lateral edge of the implant, extensive host cell infiltration was observed. This infiltration follows natural planes separating collagen bundles and forms a path by which cells can penetrate deeply into the implant. Again, a mild inflammatory response is observed and adjacent host tissue is composed of preexisting fat and newly formed connective tissue. It is important that there is no evidence of implant “degradation”. Original magnification =×200

Fig. 3 H&E stained section of Veritas® explant 14 days post-implantation. In this section, host tissue adjacent to the implant exhibits heightened vascularity (white arrow) and cellularity characteristic of healing tissue. Some infiltration of the implant by host cells is evident. A modest inflammatory response is also present. A foreign body giant cell next to a “void” which may have contained suture material (black arrow) is present. Original magnification =×200

Fig. 4 H&E stained section of Veritas® explant 29 days post-implantation. In this section, the acellular implant is visible on the right and the vascularized host tissue is on the left. The black line delineates a zone of remodeling characterized by the gradual replacement of implant material with new collagen laid down by infiltrating host fibroblasts. Original magnification =×100
This is consistent with a relative absence of inflammation at the implant host tissue (Fig. 4). Increased cellularity and vascularity were observed and these would be expected as part of the normal healing response, but the number of acute (i.e., polymorphonuclear leukocytes) and/or chronic (monocytes, macrophages) inflammatory cells was relatively small. An occasional foreign body giant cell, generally associated with chronic inflammation, was observed. In many cases, as in Fig. 4, these may well have been related to the presence of remnant suture material rather than with the implant itself.

While the bulk of the implants were retrieved after 14–15 days, two implants were left in place for 29 days to collect information regarding Veritas® remodeling at the 1-month time-point. In Fig. 5, host tissue (on the left) is separated from the acellular implant (on the extreme right) by a “transitional zone” characterized by active remodeling of implant. In this zone, alternating bands of implant collagen (distinguishable by their relative acellularity) are interspersed with new host tissue. Implant angiogenesis (i.e., the development of new blood vessels within the implant itself) is a consistent finding in 29 days (as well as 14 or 15 days) implants.

Conclusions and discussion

The unilateral hysterectomy model would appear to be a reasonable model in which to study abdominal (pelvic) adhesions. Control animals predictably develop singular and/or multiple adhesions following surgical removal of a uterine horn, and the imposition of a physical barrier (in this case the Veritas® collagen matrix) can be shown to reduce adhesion formation frequency. A 14-day time frame appears to provide an appropriate endpoint for detecting these adhesions and for testing the efficacy of adhesion-preventing materials such as Veritas®.

While the difference in the frequency of adhesion formation in the control and Veritas® groups in this study was not found to be statistically significant, there was, nonetheless, a clear trend in the data supporting the hypothesis that use of Veritas® as a physical barrier in abdominal surgery can reduce the formation of adhesions.

This, notwithstanding, histologic analysis of implants from this study provides important insight into the host response (i.e., the “biocompatibility” of Veritas® when used in the abdomen). Veritas® is extremely well tolerated by host tissue, and forms a stable interface that is largely free of inflammation. Moreover, a gradual infiltration of the implant by host cells from all surfaces can be expected over time. This infiltration is most pronounced at the lateral edges of the implant and at the interface between the implant and viscera (in this case the uterine stump). The “peritoneal” surface (i.e., the free surface in contact with other abdominal organs) is repopulated by a covering of host cells that at some points is a single layer thick within 14 days of implantation. It is possible that this covering represents the reestablishment of the normal peritoneal lining composed of mesothelial cells and that this is responsible for the adhesion-preventing properties of Veritas®.

As anticipated, implant angiogenesis and remodeling are more extensive in the 29-day explants. Based on the results of previous studies that demonstrate after a period of 3–6 months the difficulty in locating the Veritas® implant materials, it is expected that this remodeling will continue over time.
Cadaveric fascia harvested from tensor fascia lata was popularized in 1997 for use in pubovaginal sling procedures and sacrocolpopexies. Before this time, high density synthetic meshes and autologous fascial grafts had been used. Cadaveric fascia had been used for decades in orthopedic reconstructive surgeries, and its successes were widely reported in multiple case series [1]. Cadaveric fascial grafts evolved, partly in response to the dissatisfaction of pelvic surgeons with the existing graft materials. Specifically, most synthetic materials had significant erosion rates up to 10% for most materials and higher for polytetrafluoroethylene [2–5]. In addition, some of the meshes were not compatible with the material properties of the vagina resulting in a stiff rigid texture. Other complications associated with synthetic meshes included dyspareunia, infection, and in rare cases, the formation of sinus tracts and fistulas necessitating removal of the graft material [6].

Autologous fascial grafts from the patient’s own rectus fascia or tensor fascia lata offered a compatible biological profile with no risk of infectious transmission and similar material properties to the vaginal wall. However, these benefits were at the expense of significant donor site morbidity, prolonged recovery times due to pain at the harvest site, poor cosmesis at the donor site, and increased operating time to obtain the graft. Thus, cadaveric fascia lata allograft was easily introduced into the market as a graft associated with a low risk of erosion, absence of donor site morbidity, shorter operating times, more rapid recovery, and efficacy purportedly similar to that of autologous fascia [7, 8]. As a result, cadaveric tensor fascia lata became widely used in pubovaginal slings and other reconstructive pelvic surgeries, particularly, sacrocolpopexies. Both patient and surgeon outcomes in various case series confirmed minimal erosion rates and high patient satisfaction [9, 10]. By 2002, surgeons began using cadaveric fascia in anterior colporrhaphies [11, 12]. At this time, the limited data on clinical outcomes using allograft were demonstrating equivalence to autograft [8, 13]. Moreover, allograft had been used for more than 20 years in clinical practice and was thought to be both safe and stable over time [1, 14, 15].

Cadaveric fascia lata is obtained from the deep fascia that forms a sheath around the muscles of the thigh. It is composed of parallel bundles of collagen fibers and minimal cellular material. The durability and quality of the graft and its antigenicity is highly dependent on the procedure used to process and preserve the graft [16–18]. Cadaveric fascia lata is processed with the intention of decreasing antigenicity and therefore, the potential for immune sensitization and to decrease the transmission of infectious disease from donor to host. Surgeons who use fascia lata allograft should make sure that the tissue bank supplying their product adheres to the guidelines outlined by the Food and Drug administration and the American Association of Tissue Banks. In addition, the surgeon should understand potential mechanisms, whereby, the effect of the processing technique could potentially compromise its biomechanical properties.

In general, tissue processing and preservation can be divided into three steps. The first step involves screening the donor for risk factors including a medical and social history and serological screening for HIV, Hep B, Hep C, and HTLV-1. The tissue should be harvested under aseptic conditions and microbiological cultures obtained. The second step referred to as “tissue processing” is designed to eliminate infectious material from the graft and antigenic material. Tissue processing varies according to the manufacturer. LifeNet uses a patented wash referred to as Allowash which purportedly removes cells, bacteria, myobacteria, viruses, fungi, and spores. The graft is then freeze-dried. Mentor (Santa Barbara, CA) has patented a multistep solvent dehydration method in which the graft is subjected to serial treatments with sodium hydroxide, hydrogen peroxide, and organic solvents (suspend Tuto-
plast). In addition to the antigenic and infectious substances removed by other methods, the solvent dehydration method also removes prions (the infectious particle that transmits Creutzfeldt–Jakob disease). After the graft is packaged, the company has the option of terminally sterilizing the package with gamma-irradiation resulting in minimal infection transmission risk. The risk of HIV transmission from any soft tissue allograft has been estimated to be less than one in 8 million cases [19, 20].

Within each of the processing techniques are variables, which may compromise the ultimate quality of the graft that is implanted. For example, there is evidence that freeze-drying may have deleterious effects on the biomechanical properties of the graft and substantially decrease its tensile strength [21]. Independent of the effects of freeze-drying, the method of rehydration can affect the quality of the graft significantly. Most companies recommend that the graft be rehydrated for at least 1 h before placement in the patient. Incomplete hydration can lead to trapping of water between the collagen fibers as they expand resulting in a decrease in structural integrity and inferior biomechanical properties.

Irradiation is a highly effective method to sterilize any material of infectious substances. However, at very high levels, radiation can destroy the tertiary structure of collagen, thereby, compromising the tensile strength and stiffness of a graft. For example, <20 kGy is considered the ideal amount of radiation to preserve collagen structure but will only kill bacteria and not viruses. Over 30 kGy is considered completely virucidal but detrimental to collagen. As a compromise most companies use 20–25 kGy and do not rely on radiation as the sole source of graft sterilization.

Finally, donor factors likely play a role in the ultimate graft that is implanted. Ideally, a graft would be obtained from a young well-developed, well-nourished, physically active male with good muscle mass and no defects in collagen synthesis or assembly. Unfortunately, most grafts are obtained from a largely sedentary, poorly nourished elderly population with age-related sarcopenia and attenuated fascia. Indeed, multiple studies have suggested that there may be considerable variation within a company according to the patient from which the graft was harvested and within the graft according to the methods of preparation [22].

Recently, a series of retrospective case control studies have provided evidence that processed fascia lata may not be as effective as autologous fascia in regard to functional outcome after use in a pubovaginal sling procedure. McBride and colleagues [23] compared subjective quality of life measures and objective evidence of urodynamic stress incontinence that had been obtained in 71 women after placement of an autologous fascial sling (N=39) compared to a Tutoplast sling (solvent dehydrated, gamma-irradiated, N=32) at 24 months. Although, subjectively, there was no difference between groups, objective stress incontinence was reported as 0% in autograft vs 42% in allograft (P=0.007). Similarly, Soergel and associates compared failure rates in women who had undergone an autologous fascial sling (N=33) vs those who had undergone a sling procedure using freeze-dried and gamma-irradiated allograft (N=12) [22, 24]. The group found that 67% of women with allograft and 21% of women with autograft had urodynamic stress incontinence at 6 months indicating that allograft produced inferior outcomes.

For sacrocolpexy outcomes, fascia allograft has been compared only to mesh. There have been no studies comparing outcomes using autograft and allograft fascia. This is most likely a reflection of a lower likelihood of obtaining an autograft of sufficient length and width for use in a sacrocolpexy. Additionally, lighter weight polypropylene meshes were being introduced into the market with purportedly lower erosion rates than the traditional high-density meshes [5]. Culligan et al. performed a randomized controlled trial in which 54 women who had undergone a sacrocolpexy with Trelex polypropylene mesh were compared with 46 women who were treated with Tutoplast [24]. The primary outcome measure was anatomical, defined as a failure if prolapse of any compartment was present beyond Stage II as defined by the Pelvic Organ Prolapse Quantification Exam [25] at 6 months after surgery. The investigators found a significantly higher failure rate in the allograft group (32%) vs the mesh group (9%, P=0.007). Gregory et al. found similar anatomical failure rates when comparing mesh (Mersilene or Marlex) to allograft (freeze-dried, gamma-irradiated) [26]. Thus, the literature provides evidence that if given a choice between mesh and allograft in a sacrocolpexy, in regard to anatomical outcomes, mesh is superior.

Very little is known of graft physiology, once it is implanted, that may contribute to differences in clinical outcomes. In a limited retrospective case series of 35 women undergoing a sling with allograft, Fitzgerald et al. performed a histological analysis of the slings from seven women who complained of recurrent stress urinary incontinence within 6 months of surgery [27]. All of the allografts used were freeze-dried and gamma-irradiated. The authors attempted to excise the slings and found that the slings could not be retrieved in five patients and had “grossly degenerated” in two others contributing to a “material failure rate” of 20%. In such a limited study, multiple factors related to tissue remodeling and surgical procurement could account for these findings including a process the authors refer to as “autolysis.” This term, based on tissue transplantation literature, refers to the breakdown of graft material due to incompatible graft and host antigens. Further studies using animal models will be needed to determine, at molecular and cellular levels, whether processed tissue, indeed, contains sufficient material to induce a host immunologic reaction.

There have been several studies in which the biomechanical properties of various graft materials have been compared using a rabbit model. Dora et al. grafted the anterior abdominal wall of 15 rabbits with a series of grafts including fascia lata allograft (Tutoplast), porcine intestinal mucosa (SIS), polypropylene mesh, and rectus fascia allograft [28]. The investigators performed a load to failure test to generate stress strain curves at 12 weeks after
implantation. Both allograft and SIS demonstrated a decrease in tensile strength and stiffness, while there was no change in these parameters in allograft. In contrast, the stiffness of polypropylene mesh increased without a change in strength. Walter et al. obtained six fascia lata allografts (freeze-dried and gamma-irradiated) from three separate lots and implanted them into the rectovaginal space of six rabbits [29]. A tensiometer was used to generate stress–strain curves. In addition to high variability among the graft lots, the authors found a 90% decrease in tensile strength at 12 weeks after implantation. Both studies provide evidence for a biomechanical basis of the inferior outcomes seen after surgery.

In summary, the data in the literature, to date, suggest that allograft is inferior to autograft in functional outcomes for pubovaginal sling. However, if using allograft, a graft processed by solvent dehydration appears to be superior to freeze-dried. The basis for the differences in outcomes may be related to host remodeling of the grafts after placement. For sacrocolpopexy, polypropylene mesh has been shown to be superior to fascia lata. Although clearly limited data exists, studies using an animal model have shown that the biomechanical properties of polypropylene mesh may improve once implanted in the host [28, 29]. Clearly, the structural and biomechanical properties that contribute to the “ideal graft material” are not currently known. More investigations are needed to improve our currently limited understanding of the fate of a graft once implanted in the host at the molecular and cellular level.

References

Clinical implications of the biology of grafts: conclusions of the 2005 IUGA Grafts Roundtable

G. Willy Davila · Harold Drutz · Jan Deprest

Published online: 6 May 2006
© International Urogynecology Journal 2006

Abstract With few exceptions, the current expansion of graft utilization in pelvic reconstructive surgery is not a product of evidence-based medicine. Abdominal sacrocolpopexy and suburethral sling procedures are two situations under which synthetic graft utilization is indicated, based on randomized prospective trials and reported clinical outcomes. Otherwise, indications and contraindications for graft utilization are unclear. Current published data on the biology of synthetic and biologic grafts are limited and overall not very helpful to the reconstructive surgeon who is faced with the selection of a graft for use during a reconstructive procedure. This Roundtable presented the opportunity for a series of basic science researchers to present their data to a group of reconstructive surgeons and provide publishable background information on the various currently available grafts. The occurrence of healing abnormalities after graft implantation is becoming increasingly recognized as a potentially serious problem. To date, definitions and a classification system for healing abnormalities do not exist. Based on the input from basic scientists and experienced surgeons, a simple classification is suggested based on the site of healing abnormality, timing relative to graft implantation, presence of inflammatory changes, and the viscera into which the graft is exposed. Many opportunities for clinical and basic science research exist. As the use of grafts in reconstructive surgery is expanded, surgeons are encouraged to familiarize themselves with currently published data, and determine whether a graft should, or should not be, utilized during a reconstructive procedure, and if so, the type of graft best indicated in each specific clinical situation.

Conclusions of the Roundtable

The Grafts Roundtable provided a unique opportunity for an exchange of knowledge and ideas between basic science researchers and pelvic reconstructive surgeons. Although industry and current clinical trends suggest that graft materials could be used in all cases of reconstructive pelvic surgery, the Roundtable attendees acknowledged that presently there is no evidence-based medicine to justify a move in this direction. For the use of a graft to be considered, various factors should be present, including: the presenting compartment being at least to the introitus or beyond, the prolapse having a significant negative impact on the patient’s quality of life, and inadequacy in the quality of the patient’s endogenous tissues for a non-grafted repair. Abdominal sacrocolpopexy and suburethral sling procedures represent clinical situations where prospective randomized trials as well as published prospective series have demonstrated the need for a graft and the superiority of a synthetic graft over a biologic graft. Apart from these two situations, the reported indications for graft utilization are not clear and not universally recognized.

It was the consensus of this group that not all patients undergoing a reconstructive procedure for genital prolapse require the use of a graft. As more literature is published, it is likely that clearer graft utilization indications will eventually develop. In all likelihood, indications for graft utilization will be better delineated and limited to specific clinical situations. The risk/benefit ratio of graft utilization, as with any clinical intervention, will be clarified as short- and long-term benefits of graft use is balanced with short-

G. W. Davila
Cleveland Clinic Florida, 2950 Cleveland Clinic Blvd, Weston, FL 33331, USA
Tel.: +1-954-6596209
Fax: +1-954-6595587

H. Drutz
Dept. of Ob/Gyn, Section of Urogynecology, University of Toronto/Mount Sinai Hospital, 700 University Ave, Ste 3097, Toronto, Ontario Canada, M5G1Z5
e-mail: hdrutz@mtsinaion.ca

J. Deprest
Faculteit Geneeskunde KU Leuven, Centre for Surgical Technologies, Minderbroederstraat 17 3000-Leuven, Belgium
e-mail: eurofoetus@eurofoetus.org
and long-term risks of graft implantation. Moreover, the technique for graft implantation might have as important a role in long-term success as the actual makeup of the graft itself. This adds a significant variable, which requires further study. New techniques, such as transobturator and infracoccygeal routes for graft placement, have not yet been demonstrated to improve the outcomes of reconstructive pelvic surgery, whether grafted or not.

Currently accepted relative contraindications to the use of biomaterials have been suggested (Table 1). However, it was also appreciated that some of the above conditions represent a patient at a high risk for failure of a traditional prolapse repair, and that these situations might present a case where the use of a graft would be recommended by some surgeons. Resolution of some of these contraindications may occur with medical therapy (i.e., treatment of severe atrophy with local estrogen or vaginal infection with antibiotics). Others, however, are not reversible and may actually be progressive leading to an increasingly high risk of healing abnormalities (i.e., pelvic radiation with progressive devascularization). In many of these high-risk situations, graft utilization should be individualized, and preference may be given to the use of a biologic graft.

During this Roundtable, a series of observations were made based on the previous presentations by basic science researchers and the surgeons’ clinical experiences. These include:

1. Integration of a graft into host tissue is important. The implanted graft should allow for prompt collagen ingrowth and neovascularization. This should occur without the occurrence of infection or significant inflammatory reaction. A limited amount of inflammatory reaction is necessary to promote neovascularization and collagen in-growth. Grafts that are poorly integrated include microporous synthetic grafts and biologic grafts treated with chemical cross-linking. These grafts may become encapsulated, leading to hardening, shrinkage, and other graft changes, which may subsequently lead to dyspareunia, alteration of normal anatomy, and increased risk of failure. Attempts at improving integration of cross-linked biologic grafts, such as by placing holes within the graft, appear to improve their utility. However, long-term data are lacking. Microporous synthetic grafts are more likely to become infected and, thus, should not be utilized in the pelvis. Currently available monofilament, macroporous synthetic grafts, and non-cross-linked biologic grafts appear to be well-integrated into host tissues. A potential drawback to biologic non-cross-linked grafts relates to rapid metabolism and enzymatic degradation of the graft, which may occur before appropriate integration in a small percentage of patients.

2. Currently existing graft classification systems do not apply to the use of grafts in the pelvis. The Amid classification published in 1997 is frequently quoted as the accepted classification system for synthetic grafts (see Dwyer and Deprest contributions). As only Type 1 mesh is recommended for use in the pelvis, this classification does not, in general, apply to the use of synthetic grafts in the pelvis. There are currently multiple subtypes of Type 1 mesh, with markedly variable physical characteristics, including softness, weave, elasticity, and pore size, among others. There are, thus, great and clinically significant differences among Type 1 synthetic mesh materials. To improve clarity when referring to Type 1 mesh, other physical characteristics of the mesh should be described. Clear examples of these characteristics include directionality and distortion with the stretch, which can play a significant role in the outcomes of a suburethral sling procedure.

3. Evaluation of biologic grafts poses a more complex clinical challenge. Unlike synthetic grafts where composition and physical characteristics are the only significant variables, biologic grafts have a number of other characteristics, which should be considered when selecting a graft for surgical implantation.

   a. The biologic source may have a significant impact on how the graft behaves after implantation. Unpredictable graft quality has led to a decline in the use of human tissue banked grafts in preference of xenografts of more predictable integrity. Whether a porcine, bovine, or other animal source is preferable for use in the pelvis is unclear. Whether a dermal, pericardial, dural, or other anatomic source is preferable is also unclear. A great amount of work is needed in clarifying the preferred source of a biologic graft.

   b. Graft preparation is highly variable. Chemical denaturation of a graft eliminates cellular components which may predispose to immune reactivity. However, it may also weaken the integrity of a graft and alter its integration into host tissues. Irradiation of biologic grafts has been demonstrated to weaken a graft and reduce its integrity. Less is known about the impact of chemical treatment of a graft in preparation for implantation. The current concept that a non-cross-linked graft is preferable for use in the pelvis is based primarily on a theoretical basis. Comparative trials will be necessary to determine the optimal pre-implantation treatment of a biologic graft.

Table 1  Relative contraindications to the use of biomaterials

| 1. History of previous pelvic radiation |
| 2. Severe urogenital atrophy |
| 3. Immunosuppressed patient |
| 4. Presence of active pelvic or vaginal infection |
| 5. Patient currently on systemic steroids |
| 6. Host factors including: |
| a. Poorly controlled diabetes |
| b. Morbid obesity |
| c. Heavy smokers |
4. Rat biology does not always equal human biology. Animal models have been developed for evaluation of hernia repair techniques and the use of grafts in hernia surgery. These models may be helpful in evaluating tensile strength and graft incorporation. However, their utility in assessing likelihood of graft infection has no clinical correlation, as certain animal models, such as rats, are not susceptible to tissue infections. Tissue reaction to an implanted graft will also be different in an animal model as compared to a human model. The recognized occurrence of subcutaneous seromas in patients implanted with a biologic collagen matrix has not been reported in animal models and, thus, requires clinical experience in humans to be better understood. Thus, translation of animal biology to human biology is imprecise.

5. Healing abnormalities after graft implantation have various etiologies. Perhaps the most troublesome aspects of the use of grafts in reconstructive surgery relates to the development of healing abnormalities, such as “erosion,” “rejection,” and “infection.” The etiology of these healing abnormalities is poorly understood. We will not be able to minimize their occurrence until we have a better idea of their etiology. Contributing factors likely include bacterial contamination and infection, immune reaction to the graft, and physical interference with the normal healing process. Most healing abnormalities are noted with the use of synthetic polypropylene grafts. These grafts are not apt to get infected, are relatively inert, and do not elicit a significant immune reaction. Other factors must, therefore, be involved in these healing abnormalities. Technical implantation factors, such as separation of a suture line and individual host factors (patients with sensitive skin and/or multiple allergies) are likely significant contributing factors. Much basic work is required in achieving a better understanding of the etiology and management of healing abnormalities.

6. The characteristics of the ideal implant material are unclear. Some were proposed and discussed at this Roundtable (Table 2). It was accepted that currently, the perfect biomaterial does not exist. Also, it was acknowledged that currently available laboratory testing and animal models used in researching biomaterials do not adequately reflect the same environmental challenges that the female pelvis endures during a lifetime.

7. A healing abnormality classification is needed. It was agreed that a standardized classification system would be helpful in describing healing problems and designating a degree of severity. Current terminology is vague and there is lack of consensus as to the definition of the utilized terms. Terms such as “erosion”, “rejection,” and “exposure” are used frequently. These terms are poorly defined in relation to healing abnormalities associated with the usage of grafts. The participants, therefore, suggested not using these terms and rather adhering to a classification, which would describe a healing abnormality based on four factors:

- Time related to implantation
- Site of healing abnormality relative to suture line
- Presence of inflammatory tissue
- Viscera affected

This results in a classification of simple vs complex healing abnormalities, which may also help the surgeon determine a therapeutic plan (Table 3). A simple healing abnormality may require treatment with local estrogen or simple excision of the exposed mesh in the office setting. In some situations, a brief outpatient procedure in the operating room may be required. A complex healing abnormality may require increased diligence, including usage of systemic or local antibiotics, local treatment with chemical or electrocautery, or surgical exploration with the removal of a significant portion or the entire implanted graft. Based on the participants’ clinical experience, the vast majority of healing abnormalities are considered simple, based on the above classification. Typically, a Type 1 soft synthetic mesh is not associated with a complex healing abnormality, unless the mesh is visible in the bladder or rectum. The above classification will require clinical scrutiny and validation before widespread acceptance. We consider it a first step in standardizing the

---

**Table 2** “Ideal” biomaterial

<table>
<thead>
<tr>
<th>Requirement</th>
<th>Ideal Biomaterial</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inert</td>
<td>Sterile</td>
</tr>
<tr>
<td>Sterile</td>
<td>Non-carcinogenic</td>
</tr>
<tr>
<td>Mechanically durable</td>
<td>Cause no/minimal inflammatory or immune reaction</td>
</tr>
<tr>
<td>Inexpensive</td>
<td>Easy to use</td>
</tr>
<tr>
<td>Convenient</td>
<td>Readily available</td>
</tr>
<tr>
<td>Maintains implanted shape and configuration</td>
<td>(Synthetic) Withstand modification by body tissue</td>
</tr>
<tr>
<td>(Biologic) Resist enzymatic breakdown prior to established neovascularization and collagen in-growth</td>
<td></td>
</tr>
</tbody>
</table>

Timing relative to implantation  
Site relative to suture line  
Presence of inflammatory tissue  
Affected viscera  

<table>
<thead>
<tr>
<th></th>
<th>Simple</th>
<th>Complex</th>
</tr>
</thead>
<tbody>
<tr>
<td>Timing relative to</td>
<td>&lt;12</td>
<td>&gt;12 weeks</td>
</tr>
<tr>
<td>implantation</td>
<td>weeks</td>
<td></td>
</tr>
<tr>
<td>Site relative to</td>
<td>At</td>
<td>Other than at</td>
</tr>
<tr>
<td>suture line</td>
<td>suture</td>
<td>suture line</td>
</tr>
<tr>
<td>Presence of</td>
<td>None</td>
<td>Granulation</td>
</tr>
<tr>
<td>inflammatory tissue</td>
<td>Vagina</td>
<td>Bladder, rectum</td>
</tr>
<tr>
<td>Affected viscera</td>
<td></td>
<td>or other</td>
</tr>
</tbody>
</table>
implantation technique poses the most significant barrier to optimal graft utilization. Thus, improving surgeons’ knowledge is at the basis of improving outcomes from graft usage, and surgeons should be increasingly inquisitive when considering the use of a graft in reconstructive surgery.

Based on current knowledge, surgeons should not be expected to convert to the uniform use of synthetic and biologic materials for all cases of reconstructive pelvic surgery. It will be through cooperative efforts between surgeons, basic scientists, and industry that our knowledge base will increase to a degree where we will be able to make rational decisions regarding the appropriate usage of grafts in reconstructive pelvic surgery.