Chapter 7
Polymer Blends and Composites for Biomedical Applications

S.T. Lin, L. Kimble, and D. Bhattacharyya

Abstract The versatility of polymers has enabled to implement many new ideas in the biomedical field and continues to play a significant role in finding new and improved solutions in the exciting multidisciplinary fields of implants and scaffold tissue engineering. Polymers are rapidly replacing other traditional biomaterials such as metals and ceramics attributable to their versatility. Material properties of polymers will be discussed in a context specific to biomedical engineering here, which includes biocompatibility, biodegradation, biochemical and biomechanical behaviours. Biopolymers have been used as implantable materials for a broad range of biomedical applications, such as cardiology, cartilage, vasculature, bone, wound healing, drug delivery and prosthetic dentistry. Also biopolymers have become a primary material used for fabricating scaffolds in tissue engineering, a fast emerging area for tackling a critical issue in tissue and organ shortage. To address these two above-mentioned application areas, this chapter is divided into two sections: The first focuses on tissue scaffolds, including the design requirements, fabrication methods and cellular testing. The second discusses coronary stents and their development, investigating into the potential of biopolymer blends as a candidate for biodegradable coronary stents.

Keywords Polymer blends • Scaffold tissue engineering • Implant • Coronary stent • Composites

7.1 General Introduction

Biomaterial research has proposed a wide variety of applications in the field of tissue engineering and regenerative medicine, ranging from metals and ceramics to polymers, both naturally derived and synthetic materials. Among these, polymers have played a significant role and hold importance mainly due to their flexible and adjustable chemical and physical designs, being able to improve their performance

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through easily adaptable production techniques and the possibility of being tailored to suit the requirements of the application.

The versatility of polymers has inspired many new ideas in the biomedical field and continues to play a significant role in finding new and improved solutions in the exciting multidisciplinary field of implants and tissue engineering. Due to the interdisciplinary nature of this field, to develop and enhance further research, the combined knowledge from biological sciences, chemistry and engineering must be utilised to construct indispensable innovations for improving human health.

The term *biomaterials* was defined in the second consensus conference on definitions in biomaterials science, of the European Society for Biomaterials [1], as 'a material intended to interface with biological systems to evaluate, treat, augment or replace any tissue, organ, or function of the body'. The use of biomaterials dates back to more than 2000 years ago, when gold was extensively used in dentistry [2]. Other applications of biomaterials were used by the old civilisations of Romans, Egyptians, Indians and Chinese people for thousands of years for artificial eyes, ears, teeth and noses for aesthetic purposes or reconstruction of missing or defective parts of the body [3].

Since the middle of the last century, advancements allowed the first biodegradable polymeric sutures to be approved in the 1960s [4]. Another application of biomaterials using poly(2-hydroxyethyl methacrylate) (polyHEMA) was developed by Wichterle, also in the 1960s, for contact lens applications [5].

Advancements over the centuries have improved surgical techniques, sterilisation methods and biomaterials knowledge, which have enabled the development of new methods and devices to improve human health. Biomaterials have been used for implants such as sutures, bone and joint replacements, vascular grafts, heart valves and dental implants and also for medical devices such as pacemakers, biosensors, artificial hearts and tubes [3]. Significant developments in other areas of regenerative medicine has also extensively used biomaterials for repairing damaged tissue such as cartilage, bone, muscle, skin and blood vessels [6].

Polymers are rapidly replacing other traditional material choices such as metals and ceramics in biomedical applications due to their versatility. Applications for biopolymers include neural, cartilage, vascular, bone, wound dressing, drug delivery systems and dental applications. Also biopolymers have been the primary materials used for scaffold fabrication in tissue engineering applications [7]. Polymers can be classified into natural and synthetic polymers. Natural polymers are often considered to be biocompatible due to their natural compositions, and it is no surprise that they were the first used in clinical applications [8]. Natural polymers include collagen, gelatin, elastin, actin, keratin, chitosan, chitin, cellulose, silk and hyaluronic acid. Synthetic polymers include polyethylene, polypropylene, polyvinylchloride, polyurethanes, silicone, rubbers, hydrogels and polytetrafluoroethylene (PTFE).

Armentano et al. [9] has classified biopolymers into two main groups comprising of biodegradable polymers and biosourced polymers. Biodegradable polymers have been used extensively in medical applications and are defined as being capable of breaking down into molecules found in the environment such as carbon dioxide,

carbon and methane, under the enzymatic action of microorganisms, in a defined period of time (European standard EN 13432).

Biodegradable polymers can be further classified into two major groups: (1) naturally derived materials and (2) synthetic materials. Favourable characteristics of naturally derived materials include their biological compatibility which supports cell adhesion and function. However, the drawbacks of poor mechanical properties and their limited supply leading to costly factors encourage alterations or other alternatives to be found. On the other hand, synthetic materials possess hydrophobic surfaces and low cell-recognition signals, whereas their mechanical strength, manufacturing versatility and overall controllable characteristics have encouraged more research to focus on the many new possibilities of synthetic materials to be used [7]. Apart from reconstruction of tissue, polymeric biomaterials are also important in the applications such as bone cement, degradable sutures, bone screws and dental devices.

Biocompatibility is a primary requirement for biomaterials and is a critical characteristic that must be satisfied before it can be used in the body. Cell–material interactions are also crucial when biomaterials are applied clinically. Major factors including surface topography, surface chemistry, mechanical properties and biological cues can all influence cell behaviour and end response to their interactions with biomaterials. Careful design of biomaterials for specific applications can control and modulate the cell adhesion, proliferation, migration and differentiation. By using polymeric biomaterials, it is now possible to adjust the material properties, by modifying chemical composition, varying the fabrication method and altering the physical structure. While many other biomaterials have also been studied intensively and can also be adjusted and designed for different applications, they may not possess as much flexibility as polymeric materials offer.

For a material to be qualified as a biomaterial, it must first satisfy the primary requirement of being biocompatible, which is the ability of a material to perform with an appropriate host response in a specific application, or the quality of not having toxic or injurious effects on biological systems [1]. The host response to a biomaterial implant can depend on a multitude of factors ranging from the chemical, physical, biological, biomechanical and degradation properties to the structure, dimensions and shape of the implant [8].

Another important factor of biomaterials is the biodegradability in numerous applications. Current trends predict that many permanent prostheses and related devices used for temporary biomedical applications will soon be replaced by biodegradable substitutes [8]. Biodegradable materials have become more and more popular with keen interest developing further to solve current challenges such as long-term biocompatibility issues as well as the technical and ethical issues associated with revision surgeries [8]. Both natural and synthetic polymers can be biodegradable. Although degradable natural polymers such as collagen have been used for thousands of years for biomedical applications, synthetic polymers have started to gain popularity over the years since the latter half of the 1960s [10].

Note that while there are advantages of biological materials over synthetic materials such as biodegradability, favourable cell adhesion properties and having

similar mechanical properties to that of natural tissue, there are several major deficiencies of using natural biological materials such as viral infections, antigenicity, unstable material supply and batch inconsistency [10]. These unfavourable deficiencies have led to the decrease in the demand of biological materials and instead have increased the need for synthetic materials with improved properties and flexibility.

Other advantages of synthetic polymers include their uniformity and their predictability, which can be tailored and used to design specific products to fulfil specific applications. The properties of polymers affect the physical structure and thermal and chemical characteristics [11]. The majority of biodegradable polymers available on the market for regenerative medicine and tissue engineering applications are based on collagen or polyester materials. By combining different materials physically or chemically and using appropriate processing methods, many desired characteristics can be created to produce biocompatible products for biomedical applications.

Biodegradation of polymeric biomaterials works by allowing polymer erosion to occur. The mode of degradation can either involve the material degrading hydrolytically or enzymatically [10]. The majority of natural biological polymers rely on enzymatic degradation. However, hydrolytically degradable polymers are preferred for use in implants due to minimal variations compared to enzymatically degradable polymers [12]. Some of the most promising hydrolytically sensitive synthetic polymers developed for biomedical applications include poly(α-esters), polyurethanes, polyanhydrides and poly(ester amide)[8]. The earliest and most extensively studied class of biodegradable polymers is the poly(α-esters) group, and, among this group, the most extensively investigated polymers are the poly(α-hydroxy acid)s, which include poly(glycolic acid) and poly(lactic acid) [8]. Polyglycolide is highly crystalline (45–55 %) and thus possesses a high tensile modulus. It has been processed in a variety of methods including extrusion, injection and compression moulding, particulate leaching and solvent casting, to produce desirable structures for biomedical applications [13]. The first biodegradable synthetic suture called DEXON® was approved by the Food and Drug Administration (FDA) in 1969 and was based on polyglycolide. The degradation product of polyglycolide in the body is glycine, which can be excreted in the urine or converted into carbon dioxide and water via the citric acid cycle [14]. Polylactides exist in two different forms, poly (L-lactic acid) (PLLA) and poly(D-lactic acid) (PDLA), which are formed from L-lactide and D-lactide stereoisomers, respectively. Additionally, copolymers can be produced from L-lactide and D-lactide and are abbreviated PDLLA. PLLA has a crystallinity of approximately 37 %, a glass transition temperature of 60–65 °C and a melting temperature of approximately 175 °C [15]. PLLA is semi-crystalline, while PDLLA copolymers tend to be amorphous [16]. PLLA has a high elastic modulus of 3.2–3.7 GPa and strength of 55–60 MPa, while an equimolar PDLLA copolymer has an elastic modulus of 0.9 GPa and strength of 41 MPa[17]. Orthopaedic products such as BioScrew®, Bio-Anchor® and Phantom Suture Anchor® have been produced based on PLLA. PDLLA is preferred for developing drug delivery systems compared to PLLA, due to its lower strength and subsequent faster degradation rate.

Polylactides degrade into lactic acid, a normal human metabolic by-product which can be broken down into water and carbon dioxide via the citric acid cycle [14].

A range of poly(lactide-co-glycolide) (PLGA) polymers has also been developed and studied for a wide range of biomedical applications. Different ratios of glycolic acid and L-lactic acid can be used to obtain different characteristics. PLGA has been commercially used as meshes (Vicryl Mesh®), suture reinforcements, skin replacement materials and tissue scaffolding structures. Degradation rates of PLGA depend on such chemical parameters as glycolic acid and L-lactic acid ratios and molecular weight. PLGA has been FDA approved for use in humans, and its versatile processibility enables controllable shapes, structures and degradation rates to be formed.

Poly(caprolactone) (PCL) is a semi-crystalline polyester and is highly processible due to its solubility in a wide range of solvents. PCL has a melting temperature of 55–60 °C and has the ability to form miscible blends with a wide range of polymers. PCL has a relatively low tensile strength of 23 MPa but an extremely high elongation over 700 % [13]. The rate of degradation of PCL is much lower than PLA and needs approximately 2–3 years, making it suitable for longer-term drug delivery systems.

Polyurethanes possess excellent mechanical properties with good biocompatibility and structural versatility. This has attracted interest in their usage for biomedical applications which have been used as cardiac pace makers and vascular grafts. Polyanhydrides degrade by surface erosion due to their hydrophobic nature, which makes them suitable for controlled-release applications [13].

Poly(ester amide) has been developed to combine the excellent mechanical properties of polyamides and the biodegradability of polyesters [18]. Poly(ester amide) has been investigated as potential suture materials. CAMEO® is based on a poly(ester amide) blend which has been developed for site-specific delivery of small hydrophobic drug and peptides.

With the few examples given for the types of polymers and their versatility in terms of production method and overall properties, many possibilities can arise by combining the different polymers and other biomaterials available, to create endless innovations for biomedical applications.

The flourishing and rapidly advancing field of tissue engineering aims to replace, regenerate or improve damaged tissues and organs that have lost their functions. The three general strategies that have been adopted by tissue engineering include (1) implantation of isolated cells or cell substitutes; (2) delivery of tissue-inducing substances, such as growth factors, and placing cells onto or within matrices [19]; and (3) the use of tissue scaffolds, where cells are seeded onto the substrate to create implantable tissue constructs.

Tissue scaffolds are porous structures which provide mechanical support for cells, which play an important role in the regeneration of neotissue. Scaffolds mimic the natural extracellular matrix (ECM) and create an environment similar to that of the ECM in our bodies. The ECM is a noncellular component that exists within all tissues and organs. Each type of tissue has its own ECM composition and structure which are generated during tissue development through dynamic and

mutual conversations between the various cellular components. Scaffolds facilitate the regeneration of tissue by acting as a temporary ECM for cell attachment, proliferation, differentiation and subsequent growth until the tissue is completely restored.

Scaffolds have been researched and used in a number of areas such as bone, cartilage, skin, vascular tissues, neural tissues and vehicles for controlled drug delivery. The design and fabrication of tissue scaffolds are of vital importance to the tissue engineering and have become a major focus of biomaterial research recently. The most often used synthetic biopolymers for 3D scaffolds are saturated poly(α-hydroxy esters) which include PLA, PGA, PLGA and PCL. The mechanical properties and cellular adhesion and proliferation are enhanced through incorporation of nanoparticles into the synthetic particles [20].

Another important area of biomaterials is to make the implantable devices for restoring malfunction of the body. A typical example is the use of stents to mechanically support blood vessels inside the human body. Coronary arteries are the vessels commonly treated by stenting strategy due to the prevalence of coronary artery disease (CAD). CAD has become very widespread within developed regions of the world and is the leading cause of death in the US and Europe [21]. CAD is basically the build-up of plaque within the coronary arteries. Excessive plaque build-up leads to constriction of flow through the vessel, in which case the implantable stents need to be deployed to restore normal flow.

To address these two above-mentioned application areas, this chapter is divided into two major sections: The first will focus on tissue scaffolds, including the design requirements, fabrication methods and cellular testing. The second will discuss coronary stents and their development, investigating into the potential of PLLA/PBS blend as a candidate for biodegradable coronary stents.

7.2 Tissue Scaffolds

Tissue engineering and regenerative medicine combines the knowledge and advancements from multiple disciplines with the ultimate goal to create biological substitutes and replacements that can immensely improve the lives of human beings who suffer from loss or damage of body parts due to accidents and diseases. This field has grown to see the many potential applications that the fruits of research can be used for and has recognised the huge international interest that has contributed to this exciting area of improving the quality of lives for mankind.

There are nevertheless many challenges seen in the field of tissue engineering. Nerem [22] pointed out that the challenge of imitating nature to potentially solve tissue engineering issues in terms of donor tissue and organ shortages has three different categories that need to be addressed: *cell technology*, *construct technology* and integration of these into the living system. In summary, Nerem refers to *cell technology* as involving cell sourcing, manipulation of cell function and the effective use of stem cell technology. C*onstruct technology* includes engineered three-

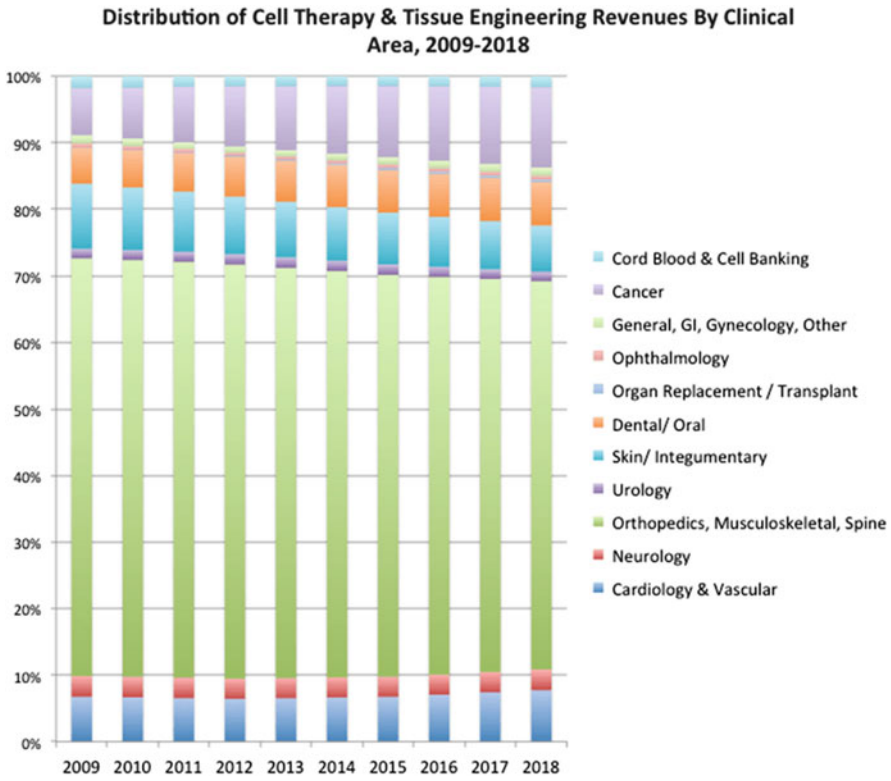

Fig. 7.1 'Distribution of cell therapy and tissue engineering revenues by clinical area, 2009–2018' (Report #S520) (Reprinted from Ref. [23], Copyright 2011, with permission from MedMarket Diligence, LLC)

dimensional architectures which either mimic a specific tissue or provide a delivery vehicle for cells.

Figure 7.1 shows the global market of tissue engineering, cell therapy and transplantation by clinical areas from 2009 to 2018. The orthopaedics, musculoskeletal and spine sector clearly dominates the market and has been predicted to continue to dominate the market right through to 2018.

Figure 7.2 shows the increase of global solid organ transplantations from 2009 to 2012 based on the Global Observatory on Donation and Transplantation (GODT) data, produced by the WHO-ONT collaboration [24]. Based on the GODT organisation, there was a 9.59 % increase in global organ transplantation activity from 2009 to 2012. Although the global activity of organ transplantation increases annually, the statistics for 2012 showed that only less than 10 % of global needs were satisfied. Kidneys are the most commonly transplanted organs worldwide, followed closely by the liver, heart, lung, pancreas and then small bowel among

Fig. 7.2 Global activity in solid organ transplantation from 2009–2012. (Plotted based on data from the WHO-ONT Global Observatory on Donation and Transplantation) [24]

Table 7.1 Global activity in solid organ transplantation from 2009–2012

	Kidney	Liver	Heart	Lung	Pancreas	Small bowel	Total
2009	72,100	21,175	5405	3650	2320	–	104,650
2010	73,179	21,602	5582	3927	2362	227	106,879
2011	76,118	23,721	5741	4278	2564	209	112,631
2012	77,818	23,986	5935	4359	2423	169	114,690

Data from the WHO-ONT Global Observatory on Donation and Transplantation [24]

other organs and tissues. The major problem is to access enough donations of tissues and organs for all the patients who need them.

Table 7.1 summarises global activity in solid organ transplantation from 2009 to 2012. Based on the incredible numbers of transplantations annually, even though it is only a small fraction of the amount that is actually required, the field of tissue engineering will continue to motivate researchers worldwide to engage in and be part of the ever increasing health demands of humanity and to constantly improve and develop existing technologies even further.

Another report on the tissue engineering and stem cell technologies by Jaklenec et al. [25] shows that the orthopaedic industry also lead the field in terms of sales for commercial products or services in 2011, taking up 50 % of sales.

As the largest sector of commercial product sales in the tissue engineering and stem cell being taken up by the orthopaedic and wound healing industry, it makes sense to explore more options in construct technology used in this area. An approach to designing tissue-like substitutes pioneered by Langer and Vacanti [19] is the cell-seeded polymeric scaffolds. The challenge here is to design a scaffold that will allow cells to create their own matrix. The structure and morphology of the scaffold is what determines the ‘environment’ that the cells are living in. Hence, it is a great design challenge to create the perfect suitable

environment to stimulate cell matrix deposition and subsequently proliferation, migration and differentiation. Research of suitable scaffolds is one thing; development of cost-effective manufacturing processes to produce these scaffolds is another aspect which many can often overlook. Ultimately, these engineered products will need to be conveniently and readily available to industry and clinicians. Hence, when designing tissue scaffolds, the ease of manufacture and increase of availability of manufactured devices are the important aspects to consider.

What is needed in the development of tissue scaffolds is an understanding of the environment in which specific cells are able to survive, and then recreate the environment using biocompatible and/or biodegradable materials for some specific applications. This has now drawn attention on finding the most suitable material for the specific requirements.

Different materials have different properties that may be better suited for use in specific applications. For example, the skin is a relatively soft tissue and therefore would not require a stiff material to support, whereas bone, on the other hand, needs to withstand much higher compressive stresses and would therefore require a much stiffer scaffold that is able to provide similar mechanical properties to the tissue which we are aiming to regenerate.

Each material would contain more favourable properties suited for specific applications compared to other materials. Since each tissue in the body has its particular role and therefore a specific set of favourable characteristics, it seems illogical to design just one super material that will cater to different purposes in tissue engineering [26]. By recognising that there are multiple aspects that need careful consideration to complete the process of designing a suitable scaffold, one should better understand the things required at each stage of research and development for delivering a product, thereby contributing to the field of tissue engineering. With this in mind, we will now focus on the topic of tissue scaffolds, as it is one of the major components required to regenerate a three-dimensional functional tissue.

7.2.1 Tissue Scaffold Materials

One of the major considerations of tissue scaffolds is the choice of material. The minimum requirement for any material is biocompatibility, both before and after degradation if the material is biodegradable. Biomaterials have recently had an impact on scaffold tissue engineering [27]. For example, by combining scaffolds with cells, the skin can be made and used for patients with burns. Various other applications involving a combination of polymers and cells such as corneas, cartilage, bone and liver have been in clinical trials [27, 28].

Many naturally occurring biological scaffolds that have great biocompatibility are available. However, one of the disadvantages of using these scaffolds is their poor mechanical properties. For this reason, synthetic polymers have emerged to solve this problem. Mechanical properties do play an important role especially for load-bearing applications. Polymers can be chosen based on the mechanical

properties required. They can also be processed in a way that may increase their mechanical properties. In addition to the favourable mechanical properties of polymers, their processing ability is another advantageous aspect, hence drawing popular demands of polymeric research in the biomedical field.

The question is which manufacturing method or process will allow the chosen bulk material to be processed into a structure suitable for the biological application that the scaffold is intended to be used for. Depending on the end application of scaffolds, different mechanical and chemical properties may be required. With so many aspects to consider, designing a scaffold with optimal characteristics such as desired strength, degradation, porosity, structure and surface, shapes and sizes is more manufacturable when using polymers [29].

7.2.2 Scaffold Design

7.2.2.1 General Structural and Morphology Requirements

Scaffolds are typically three-dimensional structures that mimic and promote an in vivo environment to support and enhance cell viability for regenerating tissues [30]. Scaffolds need to mimic the architecture of the natural extracellular matrix (ECM) environment to assist the body to heal [31]. When designing scaffolds, the following requirements are often considered:

1. Biomaterials of choice should take into account the physical and chemical properties required for the end application [32].
2. Structure and morphology of biomaterials should mimic the host tissue's structure and biological functions [33].
3. Scaffold must be biocompatible, both before and after degradation [34].
4. Scaffold must possess a porous, interconnected architecture which enables transportation of cells and cell nutrients as well as provides mechanical support during neotissue regeneration [35].
5. Scaffold surface should support cell adhesion, proliferation, migration and differentiation [36].
6. Interconnected pores with appropriate pore size to allow cell infiltration and vascularisation [37, 38].
7. Controlled biodegradability to support growth of new tissue [39, 40].

Cells need a structure to provide mechanical support while regenerating tissue. In the natural human body, the extracellular matrix (ECM) is what provides this support. The goal to imitate nature means that the natural extracellular matrix that is a composite of proteins, glycoproteins and proteoglycans is an important model that researchers are mimicking when designing scaffolds [41]. Collagen is a protein and a major component in the ECM which arranges into a fibrous network with diameters ranging from 50 to 500 nm [42, 43]. With the fibrous property that collagen possesses, ECMs have a porous structure which allows the cells to

proliferate and migrate into the ECM structure thereby differentiating into the required tissue. The architecture of ECM provides the environment that the cells grow in, which strongly influences the behaviour of the cells and the type of tissue it will differentiate into [15, 44, 45]. Generally, scaffolds must have a porous structure with sizes similar to that of the cells to be used for the particular application. By creating an imitation of the ECM using synthetic and artificial materials, limitations such as batch-to-batch variation and disease transmission associated with the materials from mammalian sources when using naturally derived biological materials can be avoided [27].

Porosity

Porosity is one of the most important aspects of a tissue scaffold as mentioned previously and is necessary in bone regeneration as shown by Kuboki et al. where direct osteogenesis occurred on the porous scaffolds [46]. Without these pores, the transport and migration of the cells and nutrients would simply be impossible and would not be assisting the cells to produce their own ECM. It is still a challenge to accurately characterise the porosity of scaffolds as there are many different types of nanostructures created from various manufacturing methods, which lead to very distinctive pore structures.

Different techniques to test and characterise the porosity may be required for different scaffolds fabricated. Some porosity characterisation techniques include mercury porosimetry [47], image analysis [48], gravimetry [49] and liquid displacement [50] methods based upon density and volume. Porosity looks at pore size, pore distribution and interconnectivity of the pores. All of these factors play their part in determining and characterising the porosity of the scaffold. In effect, a change in one could affect the other, so to study the porosity and alter the morphology and dimension, one needs to look at all the aspects that contribute to the porosity of the scaffold as a whole.

Pore Size

Pore size can directly influence the porosity of the scaffold, and in turn, affect the behaviour of the cells. Lower porosity stimulates osteogenesis, as the cell is forced to aggregate due to suppression of cell proliferation, while higher porosity allows the cells to infiltrate into the scaffold, creating more ingrowth [37]. There is, however, a compromise that has to be made for the latter, where mechanical properties maybe sacrificed if a higher porosity needs to be achieved. There is thus a limit to which this balance of porosity and functionality can be achieved, which also depends on the rate of tissue growth and the rate of scaffold degradation. It has also been shown in the previous work [51] that although specific surface area provided by scaffolds with small pores allows better initial cell adhesion, the effect

of larger pores which provide better cell and nutrient infiltration is more important as it better promotes cell proliferation and migration into the centre of the scaffold.

Interconnectivity

Depending on the type of scaffold and how it is produced, the interconnectivity of the material used for producing the porous scaffold can differ vastly. MFC scaffolds are a great example of how the interconnectivity of the structure can vary just by controlling the type of polymer used. In the MFC scaffolds, the fibrils can create an interconnected 3D network or create a fibrillar network by controlling the type of polymers used. Electrospun scaffolds, on the other hand, have a different type of connectivity mechanism, where the individual fibres intertwine and tangle with each other to create the nanofibrillar network. This kind of structure will undoubtedly allow the scaffold to have more elongation as the fibres will be allowed to stretch alongside each other. With this type of structure, the compressive strength may not be as high as that for the MFC scaffolds, where the fibrils are interconnected, forming a more rigid structure. From these examples, it can be seen that although the same type of material is used to create nanofibrous networks, the mechanism for which the nanofibres integrate to form a network is an important factor in determining and characterising the properties of the scaffold.

7.2.2.2 Cell–Scaffold Interactions

The ECM provides a structure which holds cells together to form tissues and organs. It provides anchorage and mechanical support for the cells and is also responsible for controlling and regulation of cell adhesion, spreading, proliferation, migration, differentiation and apoptosis by mechanical and biochemical communications with cells [52]. This bidirectional communication between cell and ECM influences the behaviour of each other and is important in determining how much ECM is synthesised or degraded and subsequently the fate of the cell. The realm of tissue engineering involves the use of biomaterials that can mimic the natural ECM to facilitate in cell proliferation and differentiation for the specific tissue requirements.

Cell adhesion is one of the first interactions that occur between the cell and the ECM followed by cell spreading onto the ECM. Migration of the cells is also an important aspect, especially during development and regeneration of tissues. Cell migration is encouraged by breaking the existing cell from the ECM bonds and formation of new bonds in other areas of the ECM. Cell proliferation and differentiation are also affected by the cell–ECM interaction, even though the precise mechanism for how they cooperate is still unknown. The current understanding of the behaviour of cells, proteins and other key biological factors will no doubt facilitate the amalgamation of cells and constructs to form and recreate new tissues that can be incorporated into the body.

7.2.2.3 Design Requirements for Orthopaedic Applications

Osteoinduction is part of the normal bone healing and formation of bone process, and for it to take place, osteoconduction, the growth of bone on a surface, does not just depend on biological factors but also the response of cells to implants. Successful osseointegration is achieved when there is a stable bone to implant contact where the anchorage will remain over a long period [53], enough to support the formation of new tissue. The four main components required for successful bone regeneration includes osteogenic cells, osteoconductive scaffolds, growth factors and the mechanical environment, which is referred to as the diamond concept [54, 55]. This shows that the development of osteoconductive scaffolds is a significant aspect, in which the synthesis of suitable tissue scaffolds is essential to accomplishing the regeneration of bone tissue.

It is important to be able to control the pore sizes as studies have shown that pore sizes affects cell behaviour and bone formation [37, 56]. The minimum pore size required for bone scaffolds is considered to be 100 μm due to cell size, migration requirements and biotransport [37]. However, pore sizes larger than 300 μm have been recommended to support formation of capillaries and new bone. This is important as vascularised tissue such as bone requires pores that are large enough for cell migration as well as vascular ingrowth [57]. Pore sizes around 1000 μm favour osteoblast phenotype expression, and pores around 500 μm allow more bone formation, so Sicchieri [56] has concluded that the ideal scaffold for bone tissue engineering should present pores in both sizes within the same scaffold.

7.2.3 Fabrication of Scaffolds and Assessment of Cell Viability

7.2.3.1 Scaffold Fabrication

Some common techniques as shown in Fig. 7.3 used to fabricate scaffolds include electrospinning, gas foaming [58], rapid prototyping [59], thermally induced phase separation (TIPS) [60], NFC, freeze drying [61], self-assembly [62] and solvent casting/particulate leaching [63].

One of the most important features that a tissue scaffold must possess is porosity. There are many different techniques for synthesising porous structures such as electrospinning, thermally induced phase separation, solvent casting and particulate leaching, porogen leaching, gas foaming and emulsification/freeze drying. Currently, the two main fabrication techniques used in bone tissue engineering are electrospinning and thermally induced phase separation [57]. These will be discussed further later. Recently, there has been research on a novel manufacturing method which uses a basic manufacturing process, extrusion, and further post-processing to fabricate porous scaffolds which have the potential to be completely solvent-free. This technique will also be discussed in more detail later.

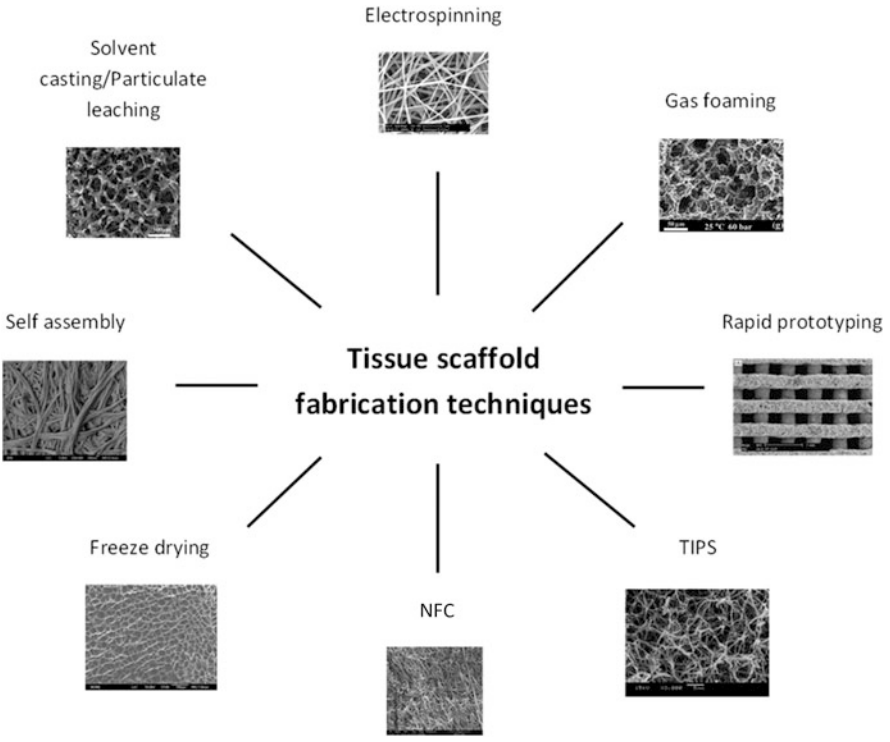

Fig. 7.3 Fabrication techniques for tissue scaffold: electrospinning, gas foaming (Reprinted from Ref. [58], Copyright 2011), rapid prototyping (Reprinted from Ref. [59], Copyright 2005), TIPS (Reprinted from Ref. [60], Copyright 2009), NFC, freeze drying (Reprinted from Ref. [61], Copyright 2009), self-assembly (Reprinted from Ref. [62], Copyright 2011), solvent casting or particulate leaching (Reprinted from Ref. [63], Copyright 2009). All with permission from Elsevier

Electrospinning

Electrospinning has received tremendous interest and been extensively researched in the past decade [64–66]. It has been recognised as a technique to produce nanofibres that have been widely studied for applications in the biomedical field [67–69]. Due to the similarity of the electrospun fibres to natural ECM, electrospinning has gained huge popularity, especially in the field of bone tissue engineering [70, 71]. Figure 7.4 shows a typical electrospun network which is composed of nanofibres overlapping and intertwined to form a fibrillar network with a very porous structure. A major drawback of electrospinning is the use of organic solvents, most of which are toxic to cells. A study by Lederer [72] showed that although the organic solvent content decreases after vacuum treatment of fibrillar scaffolds, there may still be traces that are undetectable, enough to negatively affect cell growth on the scaffolds. For this reason, there is a need for the scaffolds to be produced completely free of organic solvents.

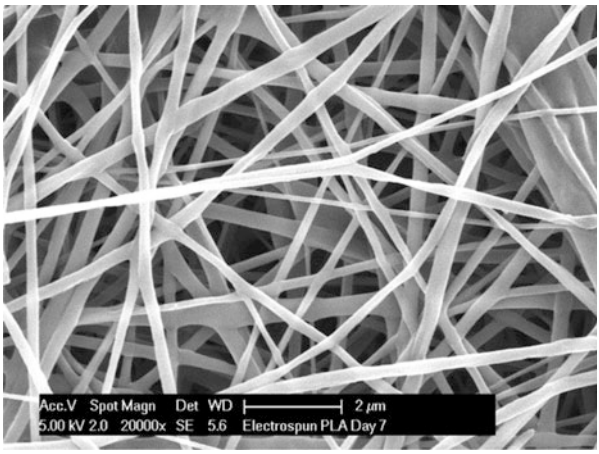

Fig. 7.4 Electrospun PLA scaffold

Thermally Induced Phase Separation (TIPS)

The TIPS technique produces a 3D nanofibrous scaffold by taking advantage of the thermodynamic instability of polymer solutions under certain conditions [33]. As described by Holzwarth [57], the TIPS process involves five steps: polymer dissolution, phase separation and gelatin, solvent extraction, freezing and freeze drying. When phase separation occurs, a polymer-rich phase with a higher concentration of polymer and a polymer lean phase with a lower concentration of polymer are formed. When the solvent is extracted and removed, the polymer-rich phase solidifies; different morphologies will form depending on the conditions of the system [33, 73]. Nanofibrous, synthetic biodegradable scaffolds have been fabricated using this method for use in tissue scaffold applications [74].

Nanofibrillar Composite Technique (NFC)

The nanofibrillar composite (NFC) technique utilises common engineering and commodity polymers to create nanofibrils of high strength and stiffness dispersed in an isotropic matrix. This technique is an emerging concept that employs melt blending of polymers to create an even distribution of insitu reinforcing nanofibrils through simple extrusion, drawing and post-processing [75, 76]. The NFC manufacturing method uses two thermodynamically immiscible polymers to create a fibrillar composite network. The polymers must have sufficient drawability to allow the formation of reinforcing fibrils and must be processed at a single temperature without the degradation of either polymer [75, 76]. One of the main advantages of this technique is that there is the opportunity for creating scaffolds which are completely free of organic solvents, by suitably selecting the isotropic matrix such that only water is required in the matrix removal process. Figure 7.5

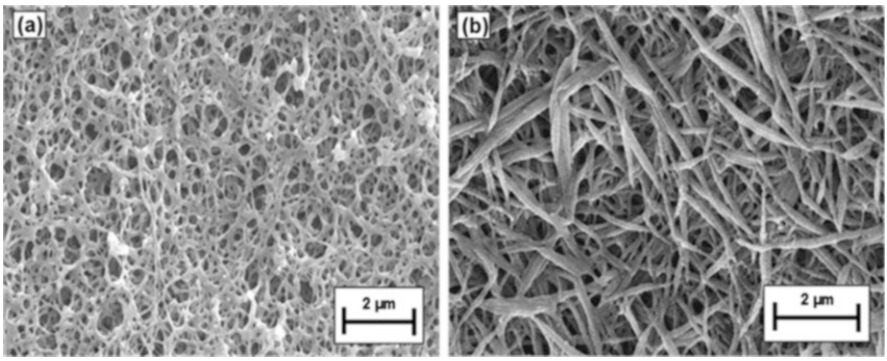

Fig. 7.5 Scaffolds manufactured using the NFC concept produced from (**a**) PLA and (**b**) PETG

shows the typical morphology of scaffolds manufactured using the MFC technique from (a) PLA and (b) PETG.

Natural and synthetic polymers have the advantages of processability, decent mechanical properties, immunological stability, manufacturability and the ease of being able to design the shape and architectures for accommodating the host tissues requirements. Where biodegradability is required, the type of polymer used can be chosen to fit the required degradation rate. This can be attributed to the large and diverse polymeric materials that have been available. Not only are they widely available, they are also simple to modify and can also be processed using a large variety of manufacturing techniques that are currently very well known and have been used for many different polymer processing. Some of the most common industrial manufacturing processes include injection moulding, blow moulding and extrusion.

Using extrusion as the main manufacturing process, polymers can be processed to produce nanoscale architectures. With the formation of nanofibres and nanopores, one of the applications which would welcome these nano-features is in the biomedical field. Tissue scaffolds require that the matrix should be highly porous to enable the cells to migrate and infiltrate the structure, transport of cell nutrients and removal of waste products and at the same time to maintain the overall mechanical properties of the synthetic ECM matrix to support cell growth.

The process of extrusion starts off with blending a mixture of polymer pellets which are thermodynamically immiscible so that the polymers will stay separate and preserve their individual properties. The polymers are melted inside a barrel which has either a single or twin screw to further blend the melted polymers. Once the blended polymers leave the die, the blended yarn is drawn by pulling and elongating the extrudate onto a winding drum which continuously rotates and pulls on the blended yarn. The final product produced from this extrusion process is a yarn of thread-like blended polymer composite.

To achieve nanoporous networks using extrusion as the main manufacturing process requires various steps in the post-extrusion section. Once a yarn of blended polymer composite is produced from the extrusion process, this yarn is transferred

to a frame which is in the shape of interest, in this case, a rectangular stainless steel frame. The transfer of the yarn can be done in various ways such as winding manually onto the frame or by using winders or lathes.

To create the porous structure that is required in tissue scaffolds, one of the components of the composite blend will need to be removed. When one component of the composite blend is removed, the remaining structure will be composed of the matrix that was not removed and the porous holes that were once filled up by the polymer that has been removed. There can be a wide variety of polymer blends which can be chosen depending on the type of properties needed. When a polymer which is water soluble is chosen as the polymer that will be removed to form the porosity of the network, then only water is required in the removal process.

7.2.3.2 Assessment of Cell Viability

To assess the cytocompatibility of cells on the scaffold material produced, mouse osteoblasts (bone-forming cells) were used in cell culture techniques to grow the cells on the materials over a period of time. Tenocytes were also used to try different cell types on the same scaffolds. Qualitative analysis was achieved by conducting live/dead staining of cells that have grown on the scaffold up to a total of 20 days and 14 days for osteoblasts and tenocytes, respectively.

One of the most important steps in the cell culture is sterilisation. A common problem with polymers is their sensitivity to heat. The method of sterilisation in most labs is autoclaving which uses pressurised steam to heat the material being sterilised. This is obviously a concern for polymers as the temperature at which lab equipment are sterilised at is typically 121 °C for 15–20 min depending on the size of the load. Polymers are sensitive to heat and typically have low glass transition temperatures. PLLA which is used in the NFC manufacturing process has a glass transition temperature of 55–60 °C. Glass transition temperatures for polyethylene terephthalate and polyvinyl alcohol are 70 °C and 85 °C, respectively. All polymers used in this project have glass transition temperatures below 100 °C.

For the purpose of cell culture, ethanol has been chosen to sterilise the materials which will be used to assess cell growth. Although ethanol technically does not sterilise, it does disinfect by killing microbial cells, but has no effect on spores. Ethanol works by denaturing proteins through a process that requires water; hence, ethanol must be diluted to 60–90 % to work effectively in disinfection of the materials. The manufactured NFC scaffolds were soaked in 70 % ethanol for a minimum of 30 min and left to dry at room temperature under sterile conditions.

UV radiation has damaging effects on cells and also makes a great tool for sterilisation. To further ensure the materials are sterile, they were exposed to UV radiation for 30 min each side. UV radiation was chosen partly due to its convenience and safety of use as it does not have high penetration so is safe to use in small areas such as laminar flow hoods.

In preparation for seeding cells onto the sterilised scaffolds, the materials were soaked overnight in the culture media used for maintenance of the cells. Tenocytes

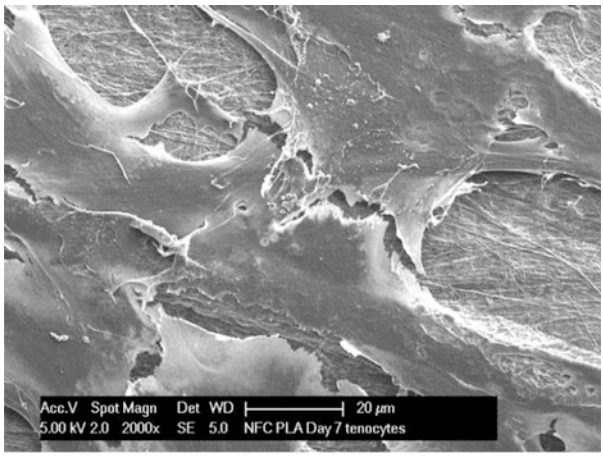

Fig. 7.6 Tenocytes grown for 7 days on PLA scaffold fabricated using the NFC technique

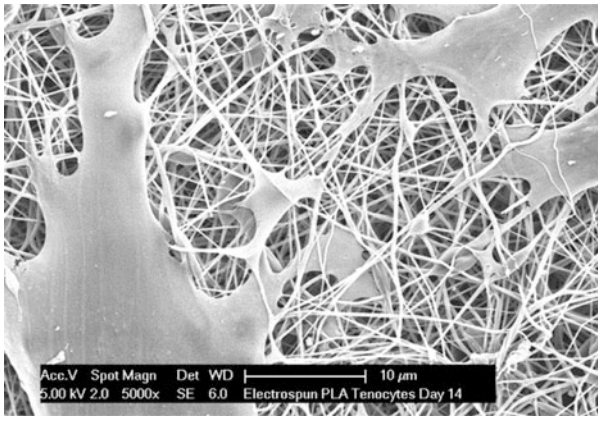

Fig. 7.7 Tenocytes grown on the electrospun scaffold for 14 days

were used to study the cytocompatability on the scaffolds. Figure 7.6 shows the tenocytes have spread across the surface of the PLA scaffold after 7 days of cell culture. The limited penetration of cells into the scaffold can be improved by increasing the porosity of the scaffold. Cell attachment on the surface of the scaffold demonstrates that the NFC fabrication technique has potential to produce scaffolds which can be completely solvent-free.

Figure 7.7 illustrates how the tenocytes have migrated beneath the top surface of the electrospun network on day 14 of cell culture. This shows the importance of having a porous interconnected structure, creating a three-dimensional space that allows cells to embed themselves within the scaffold. Traditional cell culture practice often uses 2D cell culture plates to study cells. As tissues are 3D structures, the significance of 3D cell culture studies have become paramount in mimicking the natural environment of host cells more closely. Being able to alter the pore size and

porosity of the scaffold through controlling manufacturing process is an important step in mimicking the ECM of the natural host tissue.

7.2.4 *Other Porous Scaffolds and Surface Modification Methods*

Several porous scaffolds using polymeric materials such as PLGA and PLLA and collagen have been developed and studied by the researchers from the National Institute for Materials Science (NIMS). They have developed funnel-like scaffolds using ice particulates, collagen scaffolds with micro-patterned biological molecules, ECM scaffolds derived from MSCs (chondrocytes and fibroblasts) and hybrid porous scaffolds (synthetic and natural). The surface of scaffolds can be modified to promote cell growth and can be designed to accommodate particular desired characteristics. One example of surface modification of scaffolds can be referred to the work of Lu et al. [77], where funnel-like PLGA–collagen hybrid scaffolds have demonstrated to facilitate cell seeding and homogeneous cell distribution, ECM production and chondrogenesis. These funnel-like collagen sponges were produced by using embossing ice particulates as a template. A diverse range of surface patterns can be formed on the scaffold surfaces by creating and adjusting the pattern of the ice particulates formed.

Cell functions such as angiogenesis have been manipulated by incorporating biological micro-patterned surfaces onto scaffolds. Guided blood vessel formation was achieved by preparing collagen sponges with micro-patterned vascular endothelial growth factor (VEGF). VEGF was micro-patterned in the 3D collagen sponges using micro-patterned collagen/VEGF ice lines, prepared by a dispersing machine [78]. VEGF–micro-patterned collagen sponges have demonstrated blood vessel regeneration after 6 weeks of implantation [78].

Porous ECM scaffolds have also been derived from mesenchymal stem cells (MSCs), chondrocytes and fibroblasts [81, 80]. Cells were initially cultured on poly (lactic-co-glycolic acid) (PLGA) templates until ECM was generated by the cells. The construct was then decellularised and the PLGA mesh was removed, leaving an ECM scaffold (Fig. 7.8). Using this method, three types of ECM scaffolds were produced using MSCs, chondrocytes and fibroblasts. MSCs and fibroblasts were cultured in these cell-derived ECM scaffolds to examine their potential as scaffolds for cartilage and skin tissue engineering. The MSC- and chondrocyte-derived ECM scaffolds supported cell adhesion, promoted cell proliferation and the production of ECM and demonstrated stronger stimulatory effects on the chondrogenesis of MSC. The fibroblast-derived ECM scaffolds were shown to help with fibroblast proliferation and production of ECM.

Hybrid scaffolds have also been developed to create an improved structure that combines the superior qualities of both scaffold materials. He et al. [81] developed a hybrid poly(L-lactic acid) (PLLA)-collagen hybrid sponge in the shape of a cup

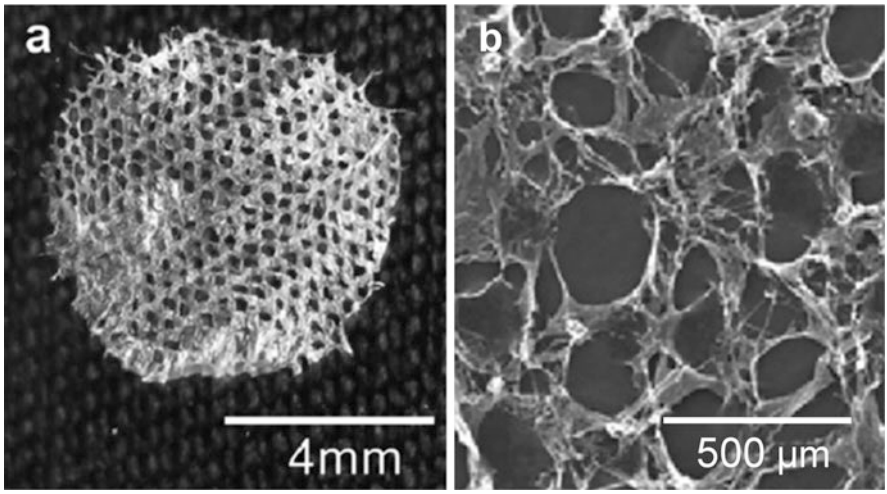

Fig. 7.8 Gross appearance of ECM scaffolds derived from MSCs (**a**) and SEM image of the scaffold (**b**). Reprinted from Ref. [80], Copyright 2010, with permission from Elsevier

with the PLLA sponge enclosing a collagen sponge in the centre. The cup-shaped PLLA sponge skeleton provided the hybrid sponge with high mechanical strength, high porosity and high cell retention and protection against cell leakage during cell seeding, while the collagen sponge in the centre contributed to high porosity and facilitated cell adhesion and distribution. Cartilage-like tissue has also been shown to form on the hybrid sponge when cultured with chondrocytes.

Another example of a hybrid scaffold was developed by Dai et al. [82], where the hybrid structure of the 3D scaffold combined the advantages of type I collagen and PLGA-knitted mesh, as shown in Fig. 7.9. The mesh provided the skeleton, while the collagen micro-sponges facilitated cell seeding and tissue formation. Bovine chondrocytes were cultured on these scaffolds and transplanted into mice for up to 8 weeks. The transplants showed homogeneous cell distribution, natural chondrocyte morphology and abundant cartilaginous ECM deposition. The mechanical strength of the engineered cartilage when compared to native articular cartilage reached up to at least 49 % in Young's modulus and 62 % in stiffness.

From these examples, it can be seen that not only is there a wide and varied choice of materials and processing methods available, but also there are many more possibilities by creating different combinations using different materials and processing methods available. There is also the biological aspect that will create even more possibilities, which needs to be taken into account. The inclusion of the biological aspects into the scaffolds, such as bioactive growth factors, will also open up another area to study which includes a large variety of incorporation methods into the scaffolds.

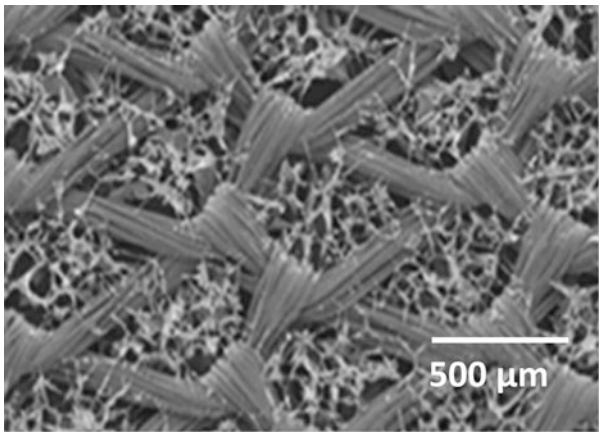

Fig. 7.9 Hybrid PLGA and type I collagen scaffold (Reprinted from Ref. [82], Copyright 2009, with permission from Elsevier)

7.2.5 Commercial and Industry Perspective

The interdisciplinary nature of the tissue engineering is one of the most exciting research areas that have led to rapid advancements as innovations in the related fields have emerged and expanded, while obstacles are being overcome. Tissue scaffold materials and fabrication techniques play a crucial role where significant progress has been achieved to provide structural support for successful tissue regeneration. Nature has provided brilliant examples to mimic, and together with technology, the society is on its way to recreating tissues and organs, with the hope of improving the quality of human lives.

It is no surprise that one of the fastest growing markets for tissue engineering and regenerative medicine products is to improve human health and longevity. To support this, the market has been creating new products based on biomaterial technologies, including both synthetic and naturally derived materials. Other technologies that have developed include genetically engineered materials, stem cell technology and cell culture technology, enabling development of even more innovative devices to enhance this interdisciplinary field of tissue engineering.

The end goal for research and development of tissue scaffolds is to improve the quality of life of humans in need of a tissue or organ replacement. One of the main methods in achieving this which has been discussed in this chapter involves creating scaffolds which mimic the ECM to provide structural support for cells to produce the required tissue naturally. For all the extensive research going on to be fully utilised, there must be a product that is produced for the patient to be able to receive the benefits that the innovations of science and engineering has created. There are numerous companies working to translate and commercialise the research, to create a product or service that can be of use for society. Commercial polymeric scaffolds that are available is summarised in Table 7.2, which has been extracted from [83].

Table 7.2 Commercial polymeric scaffolds extracted from Ref. [83]

Polymer	Biomedical application	Trade name
PGA	First biodegradable synthetic suture in 1969	DEXON
	Bone internal fixation devices	Biofix
PLLA	Orthopaedic fixation devices	Bio-Anchor, Meniscal Stinger
		The Clearfix Meniscal Dart
	High-strength fibres (FDA approved in 1971)	DEXON
	Ligament replacement of augmentation devices	Dacron
	Blood vessel conduits	
	Human immunodeficiency virus or correction of facial fat loss	
PLDLA	Bioresorbable implant material	Resomer
PLGA	Multifilament suture	Vicryl, Vicryl Rapide and CRVL
	Skin graft	Vicryl Mesh
PLGA collagen	Tissue regeneration membrane	Cytoplast Resorb
PLGA	Drug delivery vehicle	Lupron Depot
	First commercially developed monofilament suture (1980)	PDS
PDA	Orthopaedic applications	Pins
PCL	Long-term contraceptive device	Capronor
PDLLA-CL	Monofilament suture	MONOCRYL
PGCL, *PLCL*, *PETG*	Drug delivery	SynBioSys
PCLTMC &PGCL	Flexible suture materials	Maxon
	Orthopaedic tacks and screws	Acufex
PHBHV	Bone pins, plates, drug delivery	
PEU	Tissue engineering application	DegraPol
LDI-based PU	Orthopaedic applications and bone cement	Polynova
PEAs	Site-specific delivery of small hydrophobic drugs & peptides	CAMEO
POE	Tissue adhesives	Dermabond
	Bilayer skin substitute	INTEGRA Dermal
		Regeneration Template
	Wound dressings	Biobrane and AlloDerm
Collagen	Bioengineered skin equivalents	TransCyte
HA	Wound dressing application	HYAFF
	Synthetic bone graft	Ossigel
HMW viscous HA	Corneal transplantation and glaucoma surgery	Amvisc and Amvisc Plus
Viscous HA	Relieve pain and improve joint mobility for osteoarthritis	Synvisc, Orthovisc

With the promising products that are currently in use on the market as well as many other companies investing in this area of research [25], the growth of the tissue engineering field will continue to flourish in the hopes of creating even more life-saving technologies.

7.2.6 Summary

The interdisciplinary nature of the tissue engineering is one of the most exciting research areas that have led to rapid advancements as innovations in the related fields have emerged and expanded, while obstacles are being overcome. Tissue scaffold materials and fabrication techniques play a crucial role where significant progress has been achieved to provide structural support for successful tissue regeneration. Nature has provided brilliant examples for us to mimic, and together with technology, we are on the way to recreating tissues and organs, with the hope of improving the quality of lives for humans.

7.3 Stents

The human body is an extraordinarily complex system comprised of many parts and systems all working in unison to maintain and sustain health. Being mostly fluid, it is not surprising that there are various tubes within the body with functions ranging from delivery of nutrients to transmission of chemical signals and removal of metabolic waste. Sometimes these vessels may become compromised, thus losing their ability to perform their roles. In such cases, medical intervention can help to restore function by providing the vessels or tubes with support via the use of stents. In general, a stent is *a splint placed temporarily inside a duct, canal or blood vessel to aid healing or relieve an obstruction*, according to the Oxford English Dictionary [84]. The practice of implanting or deploying a stent into a vessel or tube is referred to as ‘stenting’.

Coronary arteries are the vessels commonly treated by stenting because of the prevalence of coronary artery disease (CAD). CAD has become very widespread within developed regions of the world and is the leading cause of death in the US and Europe [21]. From here onwards, ‘stents’ and ‘stenting’ refer to coronary artery stents and the implantation of stents into coronary arteries, respectively. CAD is basically the build-up of plaque within the coronary arteries, as shown in Fig. 7.10. Excessive plaque build-up leads to obstruction of flow through the vessel, in which case the vessel must be treated to restore normal flow.

CAD may be treated in several ways, namely bypass surgery, angioplasty or stenting. Bypass surgery was first introduced in 1968, before angioplasty and stenting were developed [4]. During bypass surgery, a section of healthy artery is extracted from one of the legs of the patient and is used to bypass the blocked

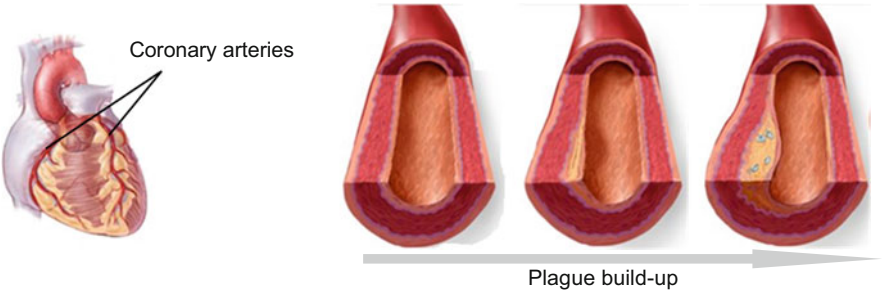

Fig. 7.10 Position of coronary arteries (*left*) and progression of plaque build-up within a coronary artery (*right*). Images adapted from www.medmovies.com

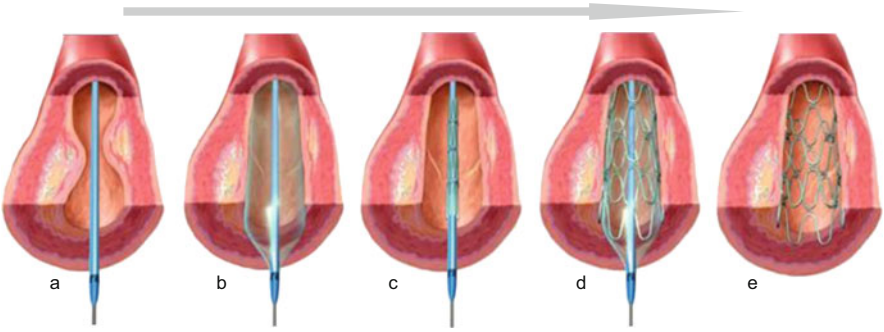

Fig. 7.11 Stenting procedure: (**a**) catheter insertion, (**b**) balloon expansion, (**c**) insertion of catheter with stent mounted on the end after removal of the balloon catheter, (**d**) stent deployment, (**e**) stented vessel. Images adapted from www.medmovies.com

section of a coronary artery, hence named bypass surgery (also known as coronary artery bypass grafting, CABG). In 1977, percutaneous transluminal coronary angioplasty (PTCA) was introduced. PTCA is a less invasive technique than CABG and begins with insertion of a catheter into the target vessel. Once the catheter is in position, a balloon on the end of it is expanded to open the blockage caused by the plaque (illustrated by a–b of Fig. 7.11), then the balloon is deflated and the catheter is removed. Unfortunately, the rate of restenosis (re-narrowing of the vessel) after PTCA is high; thus, stenting of coronary arteries after PTCA was introduced. The use of stenting began in 1986, and the procedure begins with PTCA, and then a second catheter, with a stent mounted over the end, is inserted into the vessel. Once the end of the catheter is located at the target region, the stent is deployed by balloon expansion, as shown in c–d of Fig. 7.11. The balloon is then deflated and the catheter is removed, leaving the stent in place to provide support to the vessel (Fig. 7.11e).

7.3.1 Introduction to Coronary Stents

7.3.1.1 Bare Metal Stents

The first stents were relatively simple in nature when compared with stents of today and were primarily made of medical-grade stainless steel [85] (316 L stainless steel) and less commonly of cobalt–chromium or nickel–titanium alloys. Stents of this nature fit into the category of 'bare metal stents' (BMSs). Although stainless steel has excellent mechanical properties, it contains small amounts of nickel, molybdenum and manganese to which some people are allergic [86], thus heightening the risk of restenosis (i.e. re-narrowing of the vessel) in some patients [87]. Restenosis occurs via an inflammatory reaction at the stented site of the vessel which causes smooth muscle cells to migrate to the surface of the inner wall of the vessel and proliferate. This proliferation (i.e. multiplication and spreading) of the cells is what causes the vessel to re-narrow [88]. Furthermore, blood platelets (also known as thrombocytes) have a tendency to stick to BMSs which can cause a clot, or thrombus, to form at the stented site, a phenomenon called thrombosis. This is extremely dangerous since a thrombus may break away from the stent and cause a heart attack [89, 90]. Stent thrombosis occurring soon (within 30 days) after implantation is common in the case of BMSs, although the introduction of anti-platelet therapy for patients who received BMS implantations greatly reduced occurrence rates [90].

7.3.1.2 Coated Metal Stents

Due to the common occurrence of restenosis and thrombosis after BMS implantation [91], a need was seen to alter the surface characteristics of BMSs without changing their mechanical properties, since it seemed that restenosis resulted mainly from the body's chemical interaction with BMSs. Experimentation with coating BMSs began, and a new category of stents was formed called 'coated metal stents' (CMSs). Several different coatings have been applied to stents, including platinum, carbon, gold, silicon carbide, phosphorylcholine, polymers and titanium–nitride–oxide [89, 92], in an attempt to lower the restenosis and thrombosis rates. The results of coating stents were mixed, and in some cases, such as in the case of gold-coated stents, restenosis rates were even higher than those of uncoated stents [93].

7.3.1.3 Drug-Eluting Stents

The first stent coatings were passive, and the train of thought was to make stents as benign and stable as possible to prevent undesirable responses within patents' vessels. Coated stents eventually evolved by way of a new train of thought: what if the coating could play an active role in preventing the body's responses

responsible for complications after stent implantation? Thus this strategy began the era of drug-eluting stents (DESs), which are metal stents loaded with drugs which are gradually released to provide local treatment to prevent restenosis. DESs are a significant improvement over BMSs and CMSs, but a problem which persists is the presence of a permanent, foreign object in the body. Furthermore, studies on the outcomes of treatment with DESs have revealed that late stent thrombosis rates are sometimes even higher than those of BMSs [94], and the cause may be that endothelialisation of the stent struts is delayed by drug elution [95].

7.3.1.4 Recap and the Next Phase of Stent Evolution: Biodegradable Stents

Taking a step back and considering the challenges faced by stent technology throughout its evolution from BMSs to DESs, it is noted that the initial complications faced after BMS implantation were acute, occurring soon after implantation, and were addressed as stents evolved to DESs. However, complications still persist but tend to occur later after implantation, for example, late stent thrombosis. Naturally, it is expected that the next evolutionary step is able to address this. It is recognised that stented vessels need support for only up to 6 months while they heal [96], so development of stents which degrade and essentially disappear after their jobs are done began; these stents are known as biodegradable or bioabsorbable stents. Biodegradable stents offer the advantage of completely disappearing after their service, making any future intervention less complicated. Furthermore, there is no need for rest-of-life pharmaceutical treatment as is the case for permanent stents in which case aspirin is taken indefinitely after treatment [93, 97].

Figure 7.12 shows a summary of the evolution of CAD treatment and highlights the key issues addressed and remaining with each evolutionary step. Before biodegradable stent development is discussed from a materials perspective, let us lay the foundation for this by looking at stents from an engineering point of view.

7.3.2 Stents: An Engineering Point of View

In order to better understand the challenges of biodegradable stent development, it is useful to first think about what a stent must endure during its lifetime.

7.3.2.1 Stent Deployment: The Need for Ductility

As a starting point to describe what is required of stent materials, imagine that a stent has been navigated to the target site of a vessel needing support. Now, the stent is deployed via balloon expansion in which the stent is mounted is inflated, as shown in Fig. 7.13a, b. Keep in mind that the stent is expanded to a diameter greater than the desired final diameter. Once expansion of the stent is complete, the balloon

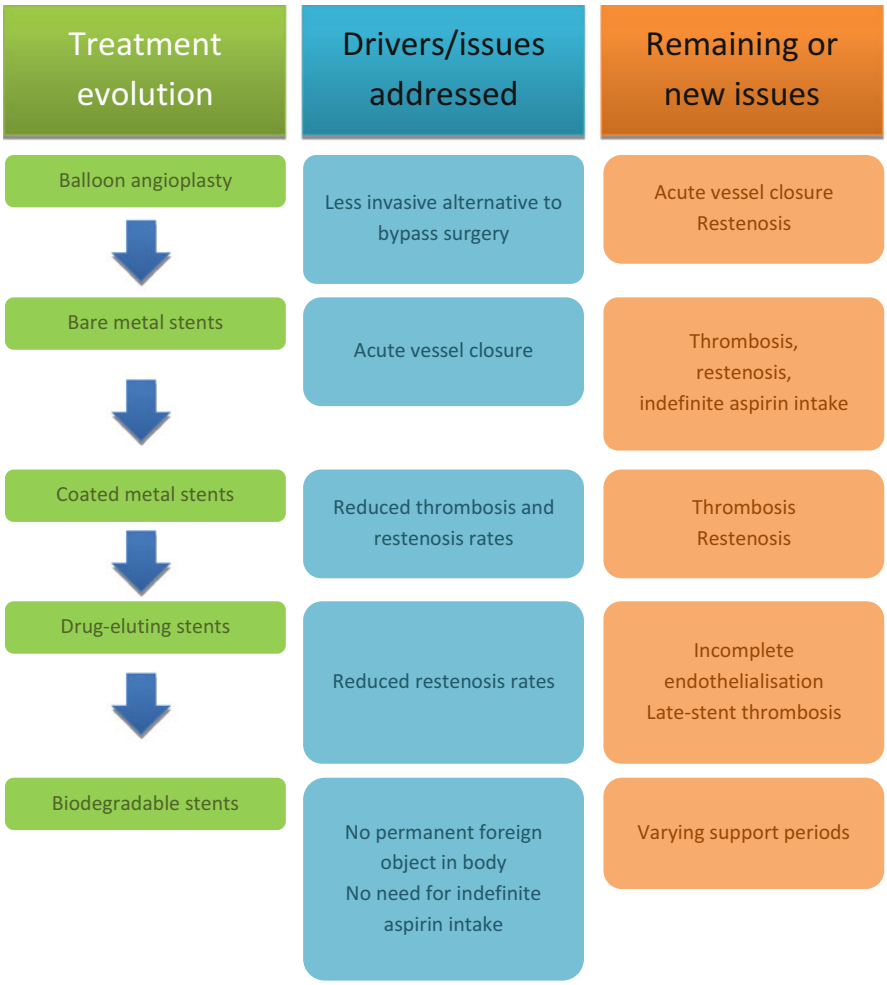

Fig. 7.12 The evolution of stents and the issues addressed with each step in progress as well as issues which remained or new issues which arose

is deflated and withdrawn as shown in Fig. 7.13c. While the balloon is deflated, the stent diameter reduces somewhat because the elastic strains within the stent material are recovered, which is why the stent had to be expanded beyond the desired final diameter. Once the balloon has been deflated and withdrawn, the stent remains in place to support the vessel, Fig. 7.13d.

From a material point of view, the important fact to note is that the stent has to undergo permanent deformation during the deployment process. This permanent deformation leads to the first of many material requirements: ductility. Ductile materials can be subjected to large strains before breaking, while brittle materials fracture instead of deforming and stretching.

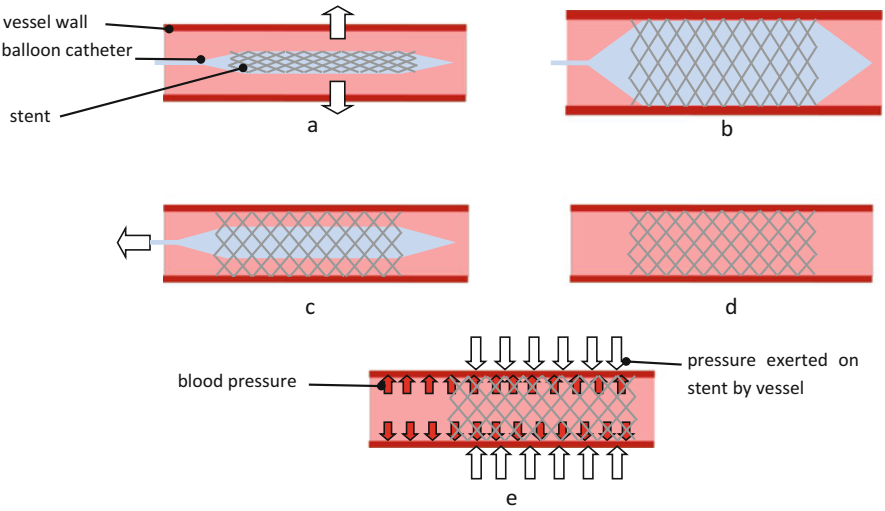

Fig. 7.13 Stent delivery by balloon expansion (**a**–**d**) and schematic of stent loading conditions (**e**). Stent on balloon catheter reaches target site and expansion begins (**a**), stent expanded beyond final diameter to ensure sufficient permanent deformation of stent material (**b**), balloon deflated and removed (**c**), stented vessel (**d**). The implanted stent experiences a radial crushing load exerted by the vessel which fluctuates since blood pressure in the vessel (which opposes pressure exerted by the vessel) fluctuates during each cardiac cycle

7.3.2.2 Importance of Creep Behaviour After Implantation

Once the stent has been deployed, it provides support to the healing vessel to prevent it from collapsing. This implies that the vessel exerts an inward pressure on the stent which must be supported until the vessel has healed sufficiently, meaning that the stent needs to endure pressure for an extended period of time. With long-term loading comes the possibility of creep, which is the gradual deformation of a material under load even if the stresses caused by this load are well below the maximum load the material can handle. In the case of a stent, creep would cause a gradual collapse, resulting in re-narrowing of the vessel. Creep is an especially important consideration for biodegradable stents for two main reasons, the first being that the majority of biodegradable stents are made of thermoplastic polymers which are prone to creep deformation and the second being the effect of degradation on material performance.

It is fairly challenging to design a stent with creep because of the numerous variables involved. For example, if a stent collapses slightly through creep deformation and the vessel experiences a corresponding narrowing, the pressure exerted on the stent decreases because the vessel is now less ‘stretched’ than it was immediately after stent implantation. Therefore the creep rate would slow down since the lower the stresses in a material, the lower the creep rate.

Furthermore, as a vessel heals, we should expect that the pressure it exerts on a stent decreases as it gradually regains the ability to support itself. This implies that

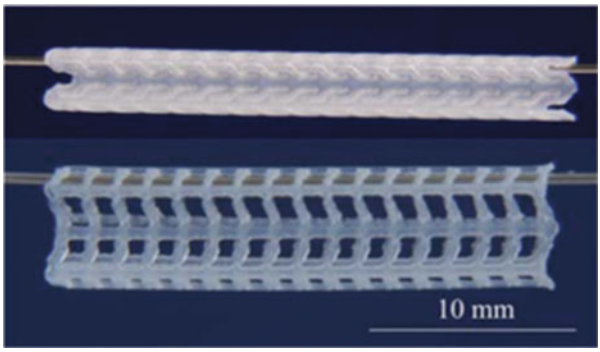

Fig. 7.14 A stent prototype made of a PLLA/P4HB blend. The stent is shown before (*top*) and after (*bottom*) balloon expansion [98]. The stent diameter remains enlarged because of permanent deformation of the PLLA/P4HB during balloon expansion

resistance to creep deformation is most critical in the early stages after stent implantation, but there seems to be a gap in the knowledge of the healing profile of a stented vessel, from an engineering point of view at least (Fig. 7.14).

7.3.2.3 Material Fatigue Considerations

With every heartbeat, blood pressure in coronary arteries fluctuates [99], and some branches also experience significant curvature changes as they bend and unbend during the cardiac cycle [100]. Blood pressure fluctuations result in subsequent vessel diameter fluctuations which would, in turn, cause the pressure exerted on an implanted stent to fluctuate. Furthermore, a stent implanted in a region where the vessel's curvature changes significantly would be forced to bend and unbend with each heartbeat. Both of these phenomena result in the presence of cyclic stresses in the stent, which raises the concern of fatigue failure. In the case of biodegradable stents, fatigue failure is destined to occur at some point when the stent material integrity has diminished enough, but care must be taken to prevent premature fatigue failures.

7.3.2.4 Material Degradation: A Critical Variable

The material considerations discussed so far apply to both permanent stents and biodegradable stents, but a consideration unique to biodegradable stents is the effect of degradation on the mechanical characteristics of the material. In order to design a biodegradable stent, we first need to understand how the properties of a potential stent material will change with time as it degrades. It is a complex process because there is still much to be learned about the rate at which support required by a healing vessel decreases, whether or not it is linear and how much it varies from patient to patient depending on age, medical conditions, etc. Perhaps when there is more knowledge in this area, biodegradable stents could be tailored to meet the needs of specific patients.

7.3.2.5 Engineering Solutions vs. Clinical Implications

At first glance, some of the challenges of stent design seem trivial to solve in engineering, but these trivial solutions may have negative clinical implications. For example, making stent struts thicker and wider enhances the support provided to the vessel but results in more restriction of blood flow as well as increased risk of small side branches of the vessel being blocked. Furthermore, endothelialisation would be more difficult in the case of thicker struts. To enhance stent performance while minimising negative impacts on clinical outcomes, stent materials must be developed to have desirable mechanical properties. The next section will outline progress of biodegradable stent material development and compare the various materials which have been on, or are currently under, investigation for biodegradable stents.

7.3.3 Materials Under Investigation for Biodegradable Stents

The engineering point of view provides insights into the mechanical requirements of biodegradable stent materials, and some of the medical requirements were discussed under stent evolution. With these in mind, the challenge of developing materials with suitable properties for the application may be getting clearer. Table 7.3 summarises some of the requirements of stents and stent materials.

Because of the array of demands of biodegradable stent materials, it is not surprising that researchers are experimenting with a wide range of materials with a broad spectrum of properties. To illustrate this, a selection of biodegradable stents is shown in Table 7.4. Note that these stents in Table 7.4 have been through clinical trials. The radial support period of these stents varies from days (in the case of a magnesium stent) through to 6 months for a poly(L-lactic acid) (PLLA) stent.

As an example of the pros and cons of different materials, the magnesium and polymer stents can be compared further. The magnesium stent has the distinct

Table 7.3 Stent and stent material requirements

Requirement	References
Biocompatibility of stent material and its degradation products	[101–103]
Low thrombogenicity to prevent stent thrombosis	[85]
The material must retain enough strength for the stent to provide sufficient support for 6 months	[103–105]
Fragments of material must not be released into the bloodstream during degradation	[103]
Radio-opaque for tracking during delivery	[85,103]
Stent must be able to withstand a minimum crush pressure of 100 kPa	[103]
The material must have sufficient flexibility to withstand being bent to get through tortuous vessels during delivery	[85,101]
The material must deform plastically upon balloon expansion to avoid recoil	[103]

Table 7.4 Comparison of biodegradable stents [104]

Stent	Material	Drug eluting?	Drug eluted	Radial support period	Absorption time
Igaki-Tamai	PLLA	No	–	6 months	2 years
BVS	PLLA	Yes	Everolimus	Cohort A – weeks Cohort B – 3 months	2 years
REVA	Tyrosine-derived polycarbonate	No	–	3–6 months	2 years
Magnesium stent	Magnesium alloy	No	–	Days or weeks	4 months
BTI	Salicylate linker	Yes	Sirolimus and salicylic acid	3 months	6 months

disadvantage of a short period of vessel support because it degrades fairly quickly. However, an advantage over the polymeric stents is that magnesium has far superior elastic modulus, so it can be designed to have thinner, narrower struts than polymeric stents while still offering good vessel support. Another advantage of magnesium is the short time for total absorption – just 4 months versus 2 years for a PLLA stent – but, again, this comes at the cost of a short vessel support period.

Magnesium is not the only metal which has been considered. Iron stents have been under investigation too. Iron takes longer to degrade in the body than magnesium, and it has superior mechanical properties to polymers, but it seems that researchers have steered away from it because of toxicity concerns [106].

7.3.3.1 Prominence of PLLA

PLLA is a very prominent material in biodegradable stent material research, both in the case of medical device companies and university research groups. Abbott Vascular (USA), Igaki Medical Planning Company (Japan), Elixir Medical Corporation (USA), ART (France) and possibly others have developed PLLA-based stents, some of which have been through clinical trials. In fact, in the first clinical trial a biodegradable stent in humans was done with Igaki-Tamai stents, which are made of PLLA.

PLLA is attractive because of its relatively high strength and stiffness when compared with other biodegradable polymers. Furthermore, it has a low enough degradation rate that stents made of PLLA can provide support for 6 months. However, PLLA suffers from brittleness which heightens the risk of stent strut fracture during deployment. For this reason, researchers have been investigating ways of modifying PLLA to be more ductile.

7.3.3.2 Modification of PLLA

There are various means of modifying polymers to alter the properties and to enhance ductility, methods such as plasticiser addition, blending with rubbery polymers and copolymerisation.

Attempts at modifying PLLA were made by Grabow et al., who blended PLLA with poly(4-hydroxybutyrate) (P4HB), as well as poly(caprolactone) (PCL), and attempted to produce biodegradable stents from these blends [36, 37]. The ductility of the blends was far greater than that of PLLA, but there were significant reductions in other mechanical properties. In the case of the PLLA/P4HB blend, tensile strength was reduced by 20 %, while stiffness was reduced by 50 %, which is undesirable since sufficient stiffness is a high priority for stenting applications. The PLLA/PCL blend exhibited an increase in strength but also a 50 % decrease in stiffness, as well as a major loss of creep resistance when compared with neat PLLA. Thus the PLLA/PCL blend was found to be unsuitable for application in biodegradable stents because stents made from this blend would possibly collapse prematurely due to low stiffness and insufficient resistance to creep.

Other work done on enhancing the ductility of PLLA involved blending with either poly(butylene succinate) (PBS) or poly(butylene succinate-co-L-lactate) (PBSL). While the work was a general study not aimed at application for biodegradable stents, the results were indicative and promising. In this study, Shibata et al. showed that the tensile elongation at break of PLLA is significantly enhanced by blending it with PBS or PBSL [107]. Furthermore, the losses of strength and stiffness were not as extreme as those exhibited by the PLLA/P4HB and PLLA/PCL blends investigated by Grabow et al. [36, 37]. For this reason, these blends are of considerable interest to the manufacturers of biodegradable stents, but there is still a lot which is unknown about them; for example, how does PBS addition influence the creep resistance of PLLA and how will degradation affect the blends' strength, stiffness and creep resistance?

7.3.3.3 PLLA/PBS: A Potential Candidate for Biodegradable Stents

The ductility of PLLA/PBS blends makes them promising candidates for the manufacture of biodegradable stents. This is the motivation for recent work which has focussed on characterising the changes in material behaviour of PLLA/PBS blends as they degrade.

Specimen Preparation PLLA and PBS were melt-blended in various ratios with up to 25 % PBS using a twin screw extruder. The blends were subsequently used to produce dog bone-type specimens via injection moulding.

Degradation Specimens were submerged in phosphate-buffered saline, with a pH of 7.4, in a temperature-controlled water bath which was used to maintain a

temperature of 37 °C. The degradation period spanned 24 weeks, and specimens were removed at intervals between 0 and 24 weeks for testing.

Tensile and Creep Tests Tensile and constant-load creep tests were performed on non-degraded and degraded specimens. Tensile tests were performed in an Instron universal testing machine at a crosshead extension rate of 50 mm/min, while loads for creep tests were applied by dead weights hanging from the specimen being tested. All tests were performed in an environmental chamber at 37 °C, and degraded specimens were tested immediately after removal from the degradation medium to ensure that they were tested in a saturated state to obtain relevant results.

Results of Tensile Tests Four different PLLA/PBS blends, as well as neat PLLA and neat PBS, were compared to determine the effects of degradation and PBS content on the changes in mechanical characteristics of PLLA/PBS blends. The changes in Young's moduli of the blend specimens as they degrade are of interest. As can be seen in Fig. 7.15, the moduli of the blends decrease gradually from 0 to 24 weeks of degradation, while those of neat PLLA and neat PBS actually increase slightly during the first 8 weeks and then stabilise.

The implications of the gradual decreases in moduli of PLLA/PBS blends during degradation could be positive if we consider that as a stented vessel heals, the amount of support it requires should decrease gradually. Figure 7.16 shows the microstructures of the PLLA/PBS specimens before degrading and after degrading. The latter becomes more porous structurally, lowering its mechanical properties.

Results of Creep Tests Creep tests were done on the specimens of one blend – 75/25 PLLA/PBS and neat PLLA, for comparison. The results of creep tests at different stresses on non-degraded specimens revealed that the initial creep rates of

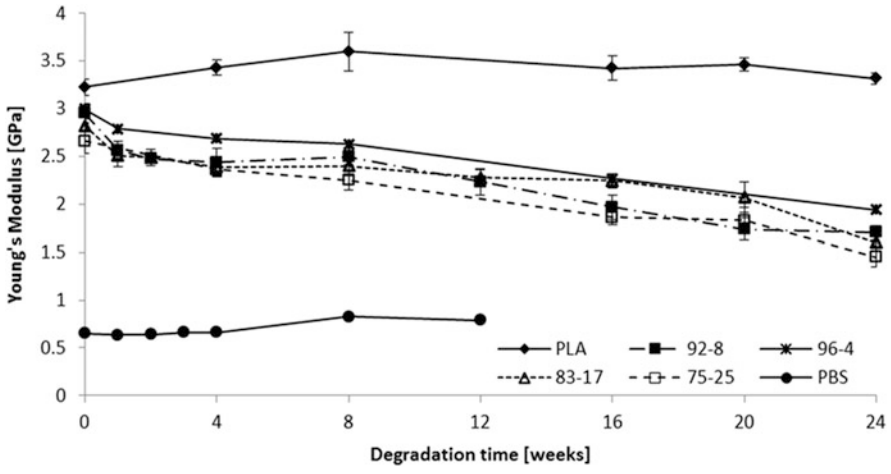

Fig. 7.15 Young's moduli of samples of PLLA/PBS, PLLA and PBS degraded for various periods of time. Error bars represent standard deviations of samples of five specimens each

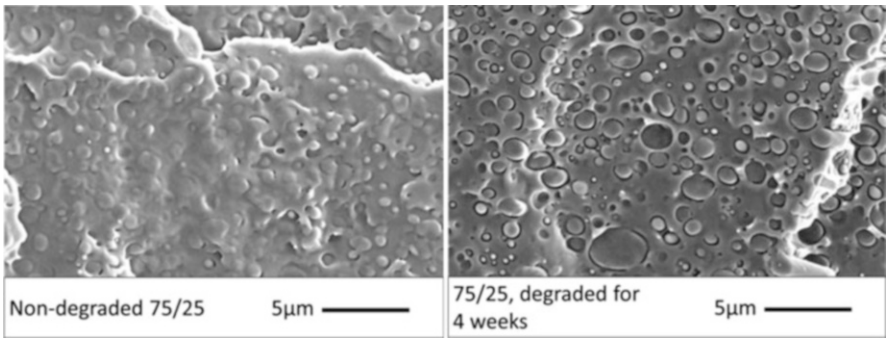

Fig. 7.16 Scanning electron microscope images of cryo-fractured 75/25 PLLA/PBS specimens: non-degraded (*left*) and degraded for 4 weeks (*right*) [108]

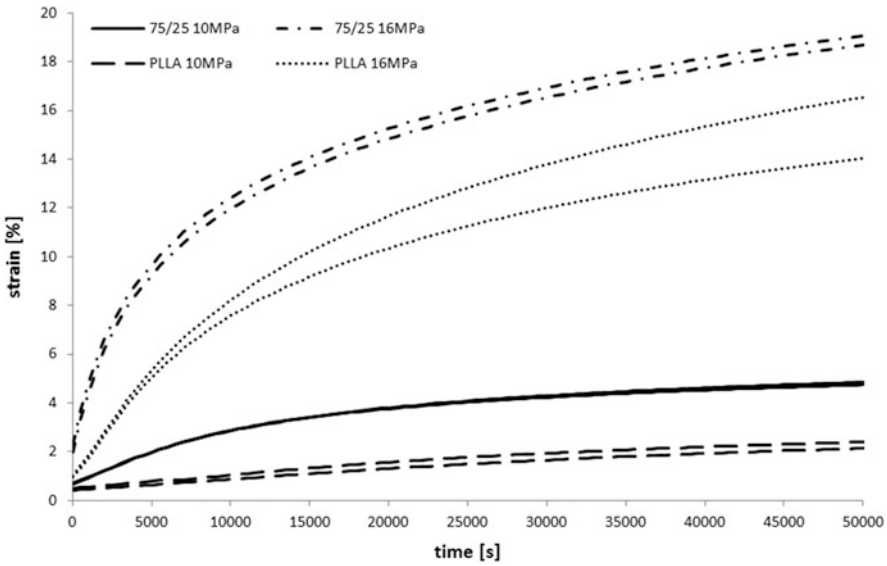

Fig. 7.17 Strain–time plot showing the first 50,000 s of constant-load creep tests performed on PLLA and 75/25 specimens. Results of two specimens from each material type are shown for each load. Stresses indicated are based on initial cross-sectional area (i.e. engineering stresses). All tests were performed at 37 °C

the 75/25 specimens were 3–4 times higher than those of neat PLLA, but physical ageing which occurred during creep tests slowed the creep rates significantly. Once creep rates stabilised after decreasing with physical ageing, the creep rates of the 75/25 blend and neat PLLA were almost the same for a given stress, as shown in Fig. 7.17.

The higher creep rates of the PLLA/PBS blend are not favourable for the application but are an expected result stemming from ductility enhancement.

Annealing was considered as a means of enhancing creep resistance, and the results from creep tests on 75/25 blend specimens annealed for 1 week at 45 °C are compared with the results of as-moulded specimens in Fig. 7.18. It is clear that annealing significantly enhances creep resistance, where the annealed 75/25 specimens exhibit the creep rates well below those of the neat PLLA. However, tensile testing of specimens annealed under the same conditions showed that the ductility enhancement from PBS addition is lost. Shorter annealing time retain some ductility but will result in a less significant boost in creep resistance, so further experiments can be done to find a balance between ductility and creep resistance.

The results of creep tests of non-degraded specimens provide important insights into the behaviour of PLLA/PBS blends, but critical to the application as biodegradable stent materials is understanding of how the creep resistance changes as the material degrades. Thus creep tests were also performed on degraded specimens. Again, a 75/25 PLLA/PBS blend was compared with neat PLLA. All the tests were done at a constant load equivalent to an engineering stress of ~10 MPa. Interestingly, the neat PLLA shows increases in creep resistance up until 8 weeks of degradation and then decreases in creep resistance from 8 to 16 weeks of degradation, as can be seen in Fig. 7.19 (left). The increase in creep resistance during the first 8 weeks of degradation can be attributed to physical ageing.

The 75/25 blend shows a trend of decreasing creep resistance with increasing degradation time. This may, at first, seem like a definite disadvantage, but it must be kept in mind that while a stented vessel heals, it will gradually exert less and less

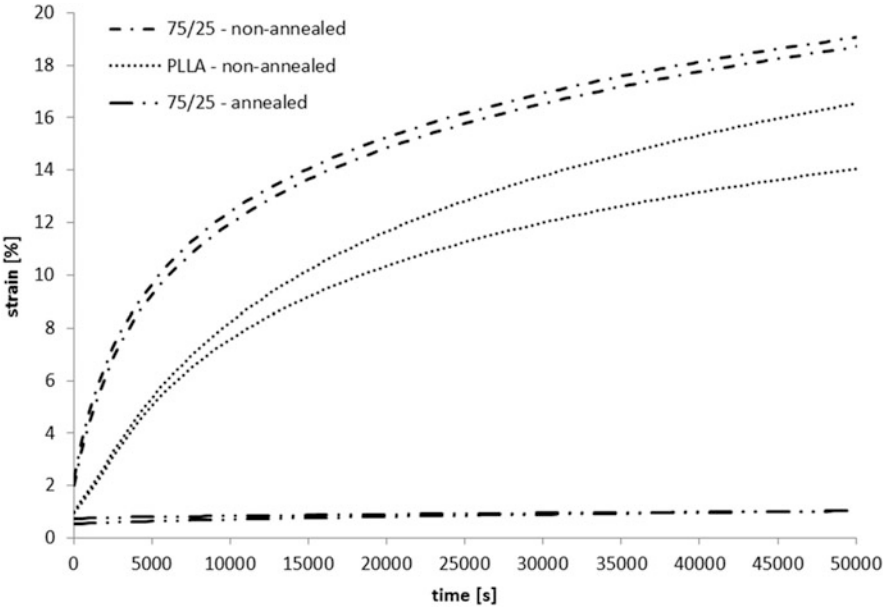

Fig. 7.18 Strain–time plots of 16 MPa creep tests of annealed 75/25 and as-moulded 75/25 and PLLA specimens. Each curve represents the results of an individual specimen

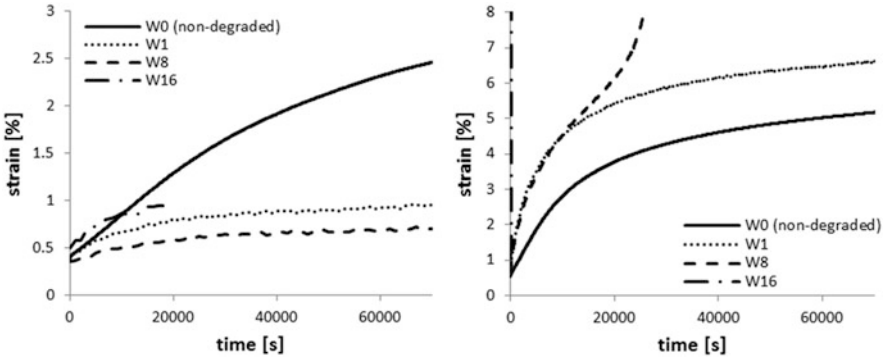

Fig. 7.19 Strain–time plots from constant-load creep tests (engineering stress ~10 MPa) of non-degraded and degraded PLLA (*left*) and 75/25 PLLA/PBS (*right*). All tests were performed at 37 °C

pressure on the stent, so a gradual decrease in the stent's ability to support the vessel is not necessarily disadvantageous. The ideal rate of decrease in support provided by a stent is not well understood, as far as the authors are aware.

7.3.4 Summary

Blending PLLA with PBS greatly enhances ductility, which is attractive for application as a biodegradable stent material. This work serves to elucidate the effects of degradation on the mechanical performance of PLLA/PBS blends in order to better understand how biodegradable stents made of these blends would perform. Results show that PLLA/PBS blends exhibit gradual decreases in their moduli as degradation progresses, while neat PLLA showed no loss over the same 24 week period. A 75/25 blend showed a gradual decrease in creep resistance with increasing degradation time, while PLLA exhibited increasing creep resistance during the first 8 weeks of degradation and a decrease from 8 to 16 weeks. The implications of the results are that a stent made of a PLLA/PBS blend would lose its ability to provide support to a healing vessel at a higher rate than a neat PLLA stent, but this may not necessarily be a negative implication since a stented vessel should require a decreasing level of support as it heals.

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