Fabrication of burst pressure competent vascular grafts via electrospinning: Effects of microstructure

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Abstract: In this work, electrospun tubes of interest for vascular tissue engineering were fabricated and evaluated for burst pressure and suture retention strength (SRS) in the same context as tensile strength providing a direct, novel comparison. Tubes could be fabricated displaying average burst pressures up to 4000 mmHg—well above the standard of 2000 mmHg—and SRS values matching those of relevant natural tissues. Surprisingly, highly oriented fiber and maximal tensile properties are not absolutely necessary to attain clinically adequate burst pressures. The ability to resist bursting is clearly related to both initial solution solids loading and electrospinning deposition time. We make novel in situ observations of the relative microstructural characteristics of failure during bursting, and connect this to the conditions used to fabricate the graft. Processes typically thought to promote fiber alignment are, in fact, highly condition-dependent and do not always provide superior properties. In fact, electrospun structures displaying no discernable alignment could achieve burst pressures regarded clinically sufficient. The properties of individual electrospun fiber clearly do not fully dictate macroscale properties. Normal background levels of point bonding are enhanced by increased rotational speeds, and can have effects on properties more dominant than those of alignment. © 2008 Wiley Periodicals, Inc. J Biomed Mater Res 88A: 923–934, 2009

Key words: biodegradable; electrospinning; bioengineering; burst pressure; biomaterials

INTRODUCTION

Coronary vascular disease is the leading cause of morbidity and mortality in the US; over 2 million surgical or percutaneous procedures are performed annually.1 Although vascular bypass grafting remains the mainstay revascularization treatment for ischemic heart disease and peripheral vascular disease, many patients do not have healthy vessels suitable for harvest. In addition, the patient population undergoing repeat coronary revascularization is increasing; ~15% of these patients will require alternative conduits.2 Pre-existing conditions may limit the availability of suitable autogenous vessels for complete coronary revascularization. Thus, there is an increased need for alternative, synthetic, small-diameter vascular grafts. Unfortunately, the use of synthetic materials is limited to grafts larger than 5–6 mm, because of the frequency of occlusion observed with small-diameter prosthetics.3,4

In 1952, Voorhees and Ah likely developed the first synthetic fiber-based vascular graft when they constructed vascular prostheses out of Vinyon N.5 Since that time, many researchers have pursued production of the "ideal" synthetic vascular graft. Although many materials have been considered as grafts, Dacron (polyethylene terephthalate) and expanded polytetrafluoroethylene (ePTFE) grafts have emerged as the primary synthetics in clinical use.6 Although large diameter grafts provide long-term patency rates,7 smaller diameter (<5 mm) grafts suffer early thrombotic complications and late myointimal hyperplasias, often leading to total graft occlusion.8 In fact, less than 50% of small diameter femoropopliteal grafts remain patent 5 years postimplantation.4

In the face of these limitations, investigators have turned their attention toward seeding vascular grafts with endothelial cells to improve long-term patency. Such tissue engineering approaches
are actively being pursued to add the only truly nonthrombogenic surface—the human endothelium—to the luminal wall of otherwise synthetic grafts. By exploiting the ability of endothelial cells to inhibit the full range of blood response to any synthetic surface, investigators hope to develop the first true cellular therapy treating cardiovascular disease. Herring and Glover made the first report of such a seeded graft in 1978. In subsequent studies, it was realized that while cell source and seeding technique are important to ultimate seeding efficiency, the substrate/scaffold upon which the cells are seeded is a primary determinant of optimal graft endothelialization and subsequent success in vivo.

A well-designed vascular scaffold must also meet two specific mechanical requirements to be effective: (1) it must retain microstructural integrity and stability after implantation and (2) it must provide sufficient biomechanical support. In producing substrates for vascular engineering, adequate burst pressure is a critical goal. The imposed requirement of 2000 mmHg is mandated by both clinical concerns and the fact that the arteries targeted for replacement do not always exhibit appropriate biomechanical properties. The concept of fiber alignment is of interest and extracellular matrix (ECM) deposition, and we have considerable experience with both this and electrospinning.

To our knowledge, no previous studies of electrospun PCL have achieved the 2000 mmHg burst pressures required for clinical application as vascular grafts. Studies have shown that electrospun nanofibrous structures have better mechanical properties than do structures composed of larger diameter fibers. Fortunately, this process can easily be used to fabricate tubular constructs suitable for subsequent cell seeding. Unfortunately, these constructs do not always exhibit appropriate biomechanical properties. The concept of fiber alignment is often put forward as a means of improving mechanical behavior. Little is known, however, about the microstructural characteristics of these "aligned" structures and how these may control burst pressure and suture retention. Here, we report our procedures for generating electrospun grafts having appropriate burst pressures and their microstructural behavior under loading regimes considered clinically relevant.

MATERIALS AND METHODS

Electrospun PCL

Solutions (12, 18, and 25 wt %) of poly(ε-caprolactone) (PCL; Sigma-Aldrich, Mw = 65,000) in acetone (Mallinckrodt Chemicals) and 6.7 wt % PCL in hexafluoropropanol (HFIP, Mallinckrodt Chemicals) were prepared by heating the solvents to 50°C, while continuously stirring to fully dissolve introduced PCL. The use of 6.7 wt % PCL in HFIP follows after the work of Zhang et al., who used 10 w/v % PCL in TFE; conversion to wt % in HFIP results in the 6.7% value. After cooling to room temperature, the solutions were deposited onto a target using a 20-gauge blunt tip needle under a 24-kV potential. A 15-cm tip-to-mandrel distance and a 20 mL/h flow rate were employed. For sheet tensile specimens, the collector plate was a 6.35 cm × 6.35 cm square covered with aluminum foil. For the tangential and longitudinal tensile specimens, a grounded 5-mm tip needle under a 24-kV potential. A 15-cm tip-to-mandrel distance was used to collect fibers. These two speeds, 0.65 and 1.66 m/s, are both much lower than the >4 m/s needed to observe statistical differences in ultimate tensile strength (UTS). For burst pressure and suture retention strength (SRS) samples, a grounded 5-mm diameter mandrel was spun at 2500 rpm (a linear tangential velocity of 0.65 m/s) and used to collect fibers. These two speeds, 0.65 and 1.66 m/s, are both much lower than the >4 m/s needed to observe statistical differences in ultimate tensile strength (UTS). For burst pressure and suture retention strength (SRS) samples, a grounded 5-mm diameter mandrel was spun at 2500 rpm (a linear tangential velocity of 1.66 m/s), while collecting fibers. The total deposition time for the tensile specimens was 10 min. For burst pressure samples, it ranged from 5 to 10 min, unless otherwise indicated. For SRS samples, deposition required 5 min. Both the tubes used to evaluate burst pressure and the tubes used to produce tensile specimens were generated from 7.6-cm long tubes. Uniformity of tube wall thicknesses along the length of the mandrel was evaluated by spinning samples (n = 5) of each composition for 7 min and recording their diameter with a laser micrometer (TLA, LS-7030). The scans consisted of >700 diametric measurement along an 8-cm length of the tubes, providing the following averages and standard deviations: 618 ± 58 μm (12 wt % in acetone), 900 ± 60 μm (18 wt % in acetone), 1057 ± 172 μm (25 wt % in acetone), 331 ± 90 μm (6.7 w/v % in HFIP). The SRS tubes were 3-cm long. All samples were placed in a vacuum overnight to ensure that the residual solvent levels in the electrospun polymer were below toxic levels.

The 12, 18, and 25 wt % PCL spun from acetone are hereafter referred to as 12, 18, and 25 wt % PCL. The 6.7 wt % PCL spun from HFIP is hereafter referred to as 6.7 wt % HFIP PCL.

Tensile strength

Tensile dog bones with a gauge length of 20 mm and a width of 2.4 mm were cut by placing the sheet between two 2-mm thick aluminum templates. A Bard-Parker No. 15 surgical blade was used to cut the straight edges, while a 3-mm dermal punch was used to cut the radii. Great care was taken to ensure that no tearing or smearing of the electrospun PCL occurred. Tensile sample thickness was measured using a digital micrometer: the gauge...
length of each specimen was confined between two glass microscope slides and the total thickness determined. Subtraction of the thicknesses of the individual glass slides provided an average gauge length thickness. The tensile properties were determined utilizing a 1-kg load cell (model 31, Sensotec) and a strain rate of 5 mm/min on an Instron load frame (model 1322) using lightweight carbon fiber grips (A2-166 Fiber Clamp Assembly, Instron). The sample size \( n \) per condition was five. To calculate the modulus from the sheet, tangential, and longitudinal tensile specimens, stress and strain values between 0 and 10% strain (~30 data points) were used.

**Burst pressure**

Using the appropriate electrospun tubes, an angioplasty balloon having the matching diameter was first evacuated and then inserted into the graft. The balloon was then filled with water at a rate of 20 mL/min and the graft allowed to lengthen freely until bursting occurred.\(^5\) The balloon + graft diameter was actively measured using a laser micrometer (Keyence LS-7001 high accuracy CCD); the inflation pressure was also recorded using a transducer (Honeywell 40PC150G) connected to a data acquisition board (National Instruments Co., Austin, TX) during filling. The data was recorded using a LabView.vi system. The burst pressure was defined as the highest pressure reached before failure up to the 5000 mmHg limit for this system. An \( n = 5 \) was used for each data point.

**As-strained burst pressure microstructures**

To examine the strained electrospun microstructure in the as-inflated condition, the angioplasty balloon was first inserted into each graft and inflated until the sample reached ~40% diametric strain. The sample did not fail, but displayed visible regions of strain and deformation. A stopcock was then used to maintain the pressure inside the balloon and the combination placed into a plastic bag that was then inserted into a circulating water bath at 50 °C for 10 min. This thermal exposure caused extensive “necking” to develop between the fibers\(^5\) and prohibited free fiber motion, while also relaxing diametric strains incurred during testing. The samples were then placed on double-sided carbon tape, and gold coated and prepared for scanning electron microscopy (SEM) [see Section “Scanning electron microscopy (SEM)”].

**Suture retention strength (SRS)**

To measure the force necessary to pull a suture through the wall of an electrospun graft, two types of cuts were used: “straight across” and “oblique.”\(^2\) The straight across method uses a graft cut normal to the long axis producing sections ~18 mm in length. Three silk sutures (6.0 Ethicon with a cutting needle) are inserted 2 mm from the end of the graft at 90° angles, looped, and tied with seven knots.\(^9\) The suture loop and the other end of the graft are secured to the grips of the tensile machine using a 22.7-kg (50 lb) load cell (Test Resources, MTestW R system) and pulled at 50 mm/min, until the suture pulled through the vessel wall. The maximum force required is the SRS. The oblique method is similar, but the graft is cut at 45° to the long axis and single sutures are located at the base (heel) of the cut, the toe of the cut, and at 90° to the toe. The electrospun samples were oriented such that the sutures pulled out parallel to the longitudinal axis of the tube (perpendicular to any potential orientation).

**Scanning electron microscopy (SEM)**

Determination of microstructural change (or its absence) required that these samples be examined in an SEM (FEI Sirion) at accelerating voltages of 5–15 kV. All samples were coated with gold (Pelco Model 3 Sputter Coater 9100). Great care was taken to make sure that the fibers were not excessively heated during sputter coating. In all cases, measurements of fiber orientation and diameter were made utilizing 100–200 fibers, following the procedures given in the pioneering work of Inai et al.\(^5\)

**RESULTS**

**Tensile testing**

Figure 1 shows that UTS is highly dependent upon the solids loading of the acetone, as each concentration produces a clearly characteristic value. The 12 wt % loading average is ~37.9 and 58.4% of the average 18 and 25 wt % values, respectively. The choice of HFIP as a solvent produces the more “classic” behavior expected from electrospun fiber aligned by the action of a rotating mandrel. In comparison to 18 wt % acetone, 6.7 wt % PCL in HFIP in the tangential direction shows a dramatic 3- to 4-fold increase in UTS. The longitudinal direction displays a strength value even lower than either the 12

![Figure 1. UTS of the PCL-solvent compositions. T = tangential (in the direction of the intended orientation); L = longitudinal (transverse to the intended orientation); S = sheet (unoriented).](image-url)
or the 18 wt % average, while the sheet condition is only 49% of the longitudinal value for 18 wt %.

In contrast, the strain data (Fig. 2) shows more dramatic variations. The 12 wt % data is remarkably uniform around 100% strain regardless of direction/condition. Relative to itself, the 18 wt % displays a substantial increase in the longitudinal versus the tangential direction (from 235 to 505%). Interestingly, the sheet condition displays a value very similar to that of the tangential direction. All three acetone conditions show relatively large standard deviations.

The 25 wt % data, in turn, is similar to the 12 wt % data in both value and consistency although the sheet condition, surprisingly, provides for greater total strain. Concurrently, the strain data shows that the 6.7 wt % HFIP data produces a tight clustering at an average of 50% strain in the tangential direction, a wider range of strains (~130 to 270%) in the longitudinal direction, and 75–110% strain in the sheet condition. As expected for highly aligned materials, total elongation to failure decreases compared to the 18 wt % PCL composition.

Figure 3 shows that acetone compositions (12, 18, and 25 wt % PCL) result in clustering of the modulus values not present in the 6.7 wt % PCL HFIP data (see inset) that varies widely. The tangential 6.7 wt % PCL HFIP data has a modulus identical to the sheet data, but its UTS is more than twice as large. Not surprisingly, the longitudinal direction displays a lower average for 6.7 wt % PCL HFIP. The longitudinal data consistently show the lowest values for each spinning parameter. Sheet tensile specimens had the highest average value regardless of spinning parameter. Quantitative data reporting both modulus and average UTS is found in Table I.

Figure 3. Modulus versus solvent identity and solids loading. Acetone compositions (12, 18, and 25 wt % PCL) result in clustering not present in 6.7 wt % PCL HFIP data (see inset) that varies widely. Not surprisingly, the longitudinal direction displays a lower average for 6.7 wt % PCL HFIP. The tangential 6.7 wt % PCL HFIP data is plotted in the upper right-hand corner of the inset) has a modulus identical to the sheet data but its UTS is more than twice as large.
Microstructure

Figure 4 shows that for 12 wt % PCL spun onto a mandrel [Fig. 4(a)], slight beading occurs and is not accompanied by measurable alignment [Fig. 5(a)]. Figure 5 shows that for 18 wt % PCL spun onto a mandrel [Fig. 4(b)], alignment is more obvious and can be measured [Fig. 5(b)]. In contrast to Figure 4(a), no obvious beading results from these spinning conditions. Upon close examination, point bonding between fibers can be observed but is, as usual, difficult to quantify. Figure 5 also shows that for 25 wt % PCL [Fig. 4(c)] spun onto a rotating mandrel, only limited (if any) alignment can be measured [Fig. 5(c)]. In contrast to Figure 4, obvious point bonding results from these spinning conditions. Figure 5 also shows that for 6.7 wt % PCL in HFIP [Fig. 4(d)] spun onto the rotating mandrel, clear evidence of alignment is observed [Fig. 5(d)]. Point bonding—resulting from the use of a too-short source-ground distance—apparently compromises the effects of orientation resulting from these spinning conditions. The average fiber diameters for 12, 18, and 25 wt % PCL in acetone are 610, 1370, and 2280 nm, respectively. The average diameter of the 6.7 w/v % PCL in HFIP fibers is 790 nm.

Burst pressure results

Figure 6 shows steady progress toward the standard goal of 2000 mmHg[13,18] resulting in spinning conditions that either meet or exceed this goal. In electrospun scaffolds, the ability to resist bursting is related to both solids loading and deposition time. However, burst pressure clearly does not follow the

<table>
<thead>
<tr>
<th>Solution (wt % PCL/Solvent)</th>
<th>Orientation</th>
<th>E (MPa)</th>
<th>UTS (MPa)</th>
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<tbody>
<tr>
<td>12/acetone</td>
<td>Tangential</td>
<td>3.95 ± 0.82</td>
<td>1.63 ± 0.17</td>
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<tr>
<td></td>
<td>Longitudinal</td>
<td>3.49 ± 0.56</td>
<td>1.25 ± 0.36</td>
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<tr>
<td></td>
<td>Sheet/random</td>
<td>4.96 ± 1.30</td>
<td>1.8 ± 0.36</td>
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<tr>
<td>18/acetone</td>
<td>Tangential</td>
<td>16.65 ± 2.62</td>
<td>4.48 ± 0.54</td>
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<tr>
<td></td>
<td>Longitudinal</td>
<td>15.61 ± 0.63</td>
<td>3.94 ± 0.40</td>
</tr>
<tr>
<td></td>
<td>Sheet/random</td>
<td>17.60 ± 3.35</td>
<td>4.13 ± 0.51</td>
</tr>
<tr>
<td>25/acetone</td>
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<td>17.00 ± 1.44</td>
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<tr>
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<td>17.07 ± 1.57</td>
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<td>Sheet/random</td>
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<td>3.42 ± 0.19</td>
</tr>
<tr>
<td>6.7/HFIP</td>
<td>Tangential</td>
<td>32.94 ± 6.62</td>
<td>14.22 ± 0.90</td>
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<tr>
<td></td>
<td>Longitudinal</td>
<td>6.38 ± 1.87</td>
<td>2.48 ± 0.44</td>
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<tr>
<td></td>
<td>Sheet/random</td>
<td>33.18 ± 3.31</td>
<td>5.42 ± 0.50</td>
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Figure 4. SEM images of mandrel-deposited electrospun PCL fibers. a) 12 wt %; b) 18 wt %; c) 25 wt %; d) 6.7 wt % HFIP PCL. The tangential direction is the vertical direction in all but b) where the tangential direction is horizontal.
trends observed during tensile testing, in that the 7-min 25 wt % (displaying a UTS of 2.5 MPa in the tensile direction) clearly resists bursting more effectively than the 7-min 18 wt % (4.5 MPa in the tensile direction). To date, HFIP displays the greatest resistance to bursting, and this is more consistent with the tensile results. Slight sintering51 of the 12 and 18 wt % fibers to produce more point bonding between the individual fibers does not result in significant increases in burst pressure.

Figure 7 shows a low magnification result of in situ microstructural analysis of the 18 wt % PCL tubular grafts subjected to diametric strain utilizing the burst pressure apparatus. This reveals a size-based hierarchical arrangement of fibers and defects in the strained microstructure. Figure 8(a) shows the typical in situ response the acetone-based composi-

Figure 6. Burst pressure versus electrospinning conditions. Only the 25 wt % PCL and 6.7 wt % PCL HFIP cases achieved clinically desirable burst pressures.
tions, in which strain is accommodated by the generation of a network structure where the edges of expanding defects are composed of larger diameter fiber apparently collected into continuous "braided" structures. This low magnification view reveals what appears to be a large (350–500 μm) defect growing within the microstructure. Within this defect, alignment of smaller diameter fibers is obvious. Given the broad distribution of fiber diameters that are generated during electrospinning, this could be due to higher levels of point bonding that logically should exist due to relatively prolonged retention of solvent in these larger diameter fibers.

In the HFIP-generated fiber [Fig. 8(b)], in contrast, increases in pressure are accommodated by increased alignment rather than the generation of individual defects. Visual observations of failure show that HFIP-based fiber grafts undergo catastrophic failure with no obvious point of initiation. The acetone-based compositions always exhibit a momentary local failure (in which the internal balloon briefly extrudes through the wall of the graft) that then propagates along the longitudinal axis of the tube.

Suture retention strength

Figure 9 shows a comparison of the SRS of electrospun tubes and relevant (porcine carotid and aortic arterial) vascular tissues. The 18 and 25 wt % tubes, in both the straight and oblique cut cases, failed at an average value of 3.7N similar to or greater than what has been observed elsewhere and statistically identical to or greater than the natural tissues. Interestingly, the 12 wt % tubes had a much lower SRS—0.56N—while the 6.7 wt % PCL in HFIP samples were statistically identical to the 18 and 25 wt % cases. The HFIP samples, however, possessed a much broader range (almost 4N in the straight cut case) of values. The 12 wt % tubes displayed a much more narrow range of SRS values.

Of equal importance is that when the interface between synthetic and natural tissue was tested, failure always occurred in the arterial wall rather than in the graft. This demonstrated that the graft SRS is more than adequate. SEM [Fig. 10(a)] shows an image of the multifilament structures penetrating both sides of a graft-tissue interface. Figure 10(b) shows a representative image of an electrospun structure following suture pullout. The sutures appear to rip cleanly through the electrospun matrix.
DISCUSSION

The generation of scaffolds having tailored, biomimetic geometries (across multiple scales) has become an increasingly active area of research.\textsuperscript{43,48,54–61} Electrospinning is an ideal method for achieving this in three-dimensional form, partly due to the ease with which it produces nonwoven nano- to micro-sized fibrous scaffolds having 70–90% relative porosity. Although electrospinning as a technique for vascular applications was first demonstrated in 1978,\textsuperscript{35} many other fabrication techniques producing synthetic forms of tissue-engineered blood vessels have been studied: collagen gels,\textsuperscript{19,62–64} polymer scaffolds\textsuperscript{11,12,18,65–69}, and self-assembly.\textsuperscript{13,70} In spite of the successes of these alternative methods, electrospinning has enjoyed recent popularity due its inherent simplicity and affordability.

Electrospinning techniques have previously been used for synthetic graft fabrication. Xu et al. electrospun a block copolymer, poly(\textit{\alpha}-lactide-caprolactone (P[\textit{\alpha}-LLA-CL]) (75:25)) and demonstrated that it had greater tensile strength and elongation than vascular tissue,\textsuperscript{46} Boland et al. electrospun both collagen and elastin into tubes having appropriate physiological diameters,\textsuperscript{71} Jeong et al. spun novel tubes consisting of a porous collagen matrix and an electrospun PLGA layer that substantially improved the mechanical strength of the collagen scaffold.\textsuperscript{72} Stitzel et al. fabricated composite collagen, elastin, and poly(lactic-co-glycolic acid) (PLGA) scaffolds that displayed burst pressures of up to 1425 mmHg.\textsuperscript{73} The majority of these investigations included some level of mechanical property characterization, but said little regarding the nature of the observed failures did not describe behavior at the microstructural level. We set out to more fully understand what factors, exactly, allow a mechanically capable graft to be electrospun. This relies heavily on previous work regarding point bonding concepts\textsuperscript{51,54} potentially important in electrospun structures having a fortunate biomimetic resemblance to biologically produced ECM’s.

Creating small diameter synthetic vascular grafts involves a number of challenges. The first challenge is to fabricate a structural matrix that possesses mechanical strength and flexibility sufficient to withstand high physiological pressures \textit{in vivo} without being prone to aneurysm and graft dilation. Engineering-based approaches to such mechanical property goals often rely solely on tensile evaluations. In the context of vascular tissue engineering, however, burst pressure and SRS are more clinically relevant as they better describe a surgeon’s immediate concerns regarding any new graft material. The former provides a measure of how likely these grafts can resist the blood pressures generated within the body. The prerequisite clinical benchmarks are well-described.\textsuperscript{17,18,74} SRS, on the other hand, concerns a practical but equally important point: can these structures successfully retain the sutures used to hold the graft in place? We evaluate three parameters (tensile strength, burst pressure, and SRS) in the same context providing a direct, novel comparison of these different properties.

The microstructure of these engineered vascular graft prostheses also has an important role in modulating tissue ingrowth. Electrospun structures feature a morphological similarity to the extracellular matrix of natural tissues being characterized by a wide pore diameter distribution, high porosity and, as we show here, effective mechanical properties. The nanofibrous structures produced by the electrospinning
process have a relatively (compared to standard Dacron) high surface area to volume ratio. Based on simple estimates of relative fiber diameter difference (~10 μm for Dacron; ~1 μm for electrospun PCL) we estimate that the surface area of electrospun fiber is roughly 12 times greater thus making much more substrate available for cell attachment.

The mechanical properties of electrospun structures shown in Figures 1–3 reveal general trends in UTS, elongation, and modulus versus spinning conditions. The 12 wt % acetone material displays the lowest values in all three categories. Its tangential and longitudinal directions and the sheet condition are statistically indistinguishable. There appears to be little mechanical advantage in attempting to produce alignment from this particular solids loading. SEM (Fig. 4) shows that this condition contains point bonding that may compromise any potential mechanical property gains.

The 18 wt % longitudinal case shows the greatest elongation (up to 600%, Fig. 2), while its tangential direction and sheet condition are much lower and statistically indistinguishable. Strain in the longitudinal direction runs perpendicular to any existing alignment and generates a cellular arrangement of fibers, allowing maximum extension at minimal levels of applied force. Both the UTS and the moduli, however, are not statistically different between the three cases. This then suggests that point bonding is significant enough to negate advantages gained by the alignment evident in the 18 wt % samples (Figs. 4 and 5).

The 25 wt % case shows no difference in either modulus, UTS, or elongation in the two directions. Interestingly, however, the sheet condition is both stronger and able to undergo greater elongation than either the tangential or the longitudinal directions. This suggests that the tubular deposition process can, in this circumstance, generate greater amounts of point bonding that compromise UTS and elongation. Indeed, Figure 4 makes it clear that this composition is at the edge of “spinnability” and results in a strongly networked structure. Further increases in solids loading would likely lead to the generation of a largely solid polymer film interspersed with fiber-like features.

The use of HFIP introduces a solvent having greater volatility and higher dielectric strength, factors known to improve “spinnability.” This composition results in the classically expected effects of alignment (Fig. 5) on modulus (Fig. 3), UTS (Fig. 1), and elongation (Fig. 2) for both the tangential and longitudinal directions. The tangential direction has more than six times the strength of the longitudinal direction and more than three times that of the 18 wt % acetone case, the best performing acetone-based composition. The longitudinal direction shows both greater elongation and UTS as low as the 12 wt % acetone cases. What is more startling, however, is the behavior of the sheet condition comprised of completely unoriented fiber. Its modulus is identical to that of the highly aligned (Fig. 3) fiber. Elongation is only slightly higher than the tangential case, in spite of the absence of alignment. Although decreases in crystallinity versus increasing mandrel speed are well-established, taken by itself this phenomenon would be expected to result in bulk property decreases; however, the opposite has been observed. We suggest that individual electrospun fiber properties do not efficiently translate to the macroscale. In fact, the literature in this area reveals some ambiguity regarding the net efficiency of increased mandrel speed as a means of improving bulk properties. Net properties have been observed to either decrease or remain unchanged in the tangential/circumferential and transverse/longitudinal directions. Our observations of increased point bonding with increases in rotational speed provide evidence that point bonding can have more dominant effects on properties than alignment.

Figure 6 shows that we met or exceeded the goal set out at the beginning of this investigation. There appears to be an inherent thickness dependence, as increased deposition time provides greater burst pressure resistance. The desired thickness in any vascular application, however, varies greatly with physiological location and we plan to investigate this in more detail. The fact that burst pressure and tensile strength do not necessarily follow one another reflects the fact that the stress states are different. Tensile strain is applied uniaxially while bursting strain is biaxial, or applied in more than one direction at once. The latter more efficiently locates defects in the microstructure (Fig. 7) and leads to earlier failure. If these inherent defect populations could be quantified in terms of their size and ranked in descending order, we suspect that they would follow 18 wt % > 25 wt % > HFIP. This would largely explain the differences observed in Figure 6.

These observations prove that electrospun tubes can exhibit burst pressures that equal or surpass surgical standards. Figures 7 and 8 suggest that the mechanisms leading to failure in these structures involves (1) isolation of stress to areas of the scaffold having smaller diameters and thus greater compliance and thus easier expansion of pre-existing defects; (2) localized alignment of fiber in an arbitrary direction; and (3) fracture of these strained, highly aligned fibers leading to catastrophic failure in the longitudinal direction.

The SRS data clearly show that electrospun tubes can match or exceed the suture retention capabilities of relevant natural tissues. Only the 12 wt % acetone
data are clearly weaker, an observation that fits the trend generated by the tensile, modulus, and burst pressure data. SEM (Fig. 10) shows that pull through of the suture occurs by relatively clean fracture of the matrix without significant permanent alignment or deformation of the surrounding fiber.

We are well aware that compliance matching is another factor critical to avoiding long-term occlusion. Compliance measurements on these electrospun tubes are ongoing.

CONCLUSIONS

For any near term application of electrospun structures as vascular replacements, ensuring adequate mechanical properties is of paramount importance. By careful control of conditions, electrospun PCL tubes having burst pressures > 2000 mmHg could be routinely synthesized. In the course of this study, we have injected biomimetic elements of vascular design by targeting fiber alignment to achieve improved mechanical properties. The expected positive effects of alignment can, however, be compromised by increased fiber-to-fiber point bonding. Electrospun structures displaying no discernable alignment could achieve clinically sufficient burst pressures. We link the microstructural characteristics of these burst pressure competent electrospun tubes to more standard mechanical properties. In the future, we will address another “missing link,” the connection between compliance and microstructure.

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