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Fabrication and Characterization of Medical Grade Polyurethane Composite Catheters for Near-Infrared Imaging

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Abstract

Peripherally inserted central catheters (PICCs) are hollow polymeric tubes that transport nutrients, blood and medications to neonates. To determine proper PICC placement, frequent X-ray imaging of neonates is performed. Because X-rays pose severe health risks to neonates, safer alternatives are needed. We hypothesize that near infrared (NIR) polymer composites can be fabricated into catheters by incorporating a fluorescent dye (IRDye 800CW) and visualized using NIR imaging. To fabricate catheters, polymer and dye are dry mixed and pressed, sectioned, and extruded to produce hollow tubes. We analyzed surface roughness, stiffness, dye retention, NIR contrast intensity, and biocompatibility. The extrusion process did not significantly alter the mechanical properties of the polymer composites. Over a period of 23 days, only $6.35 \pm 5.08\%$ dye leached out of catheters. The addition of 0.025 wt% dye resulted in a 14-fold contrast enhancement producing clear PICC images at 1 cm under a tissue equivalent. The addition of IRDye 800CW did not alter the biocompatibility of the polymer and did not increase adhesion of cells to the surface. We successfully demonstrated that catheters can be imaged without the use of harmful radiation and still maintain the same properties as the unaltered medical grade equivalent.

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Keywords

Catheter; Cell adhesion; Polyurethane; Fluorescence; Mechanical Testing

1. Introduction

Catheters offer a variety of uses in the clinical setting, including the delivery of chemical agents (such as drugs and imaging dyes), nutrients and blood to patients (1). Peripherally inserted central catheters (PICCs), which are inserted into veins not in the chest or abdomen, are widely used in neonatal and pediatric intensive care units (ICUs) for long-term delivery of therapeutics with lower infection rates compared to central venous catheters (2-4). However, the long-term placement of PICCs increases the likelihood of migration of the catheter from the target location, resulting in adverse effects to the patient (2,5). These side effects include vascular perforation (pierced blood vessel), venous thrombosis (blocked blood vessel), and pericardial tamponade (pressure on the heart), all of which can result in death (2,6). In addition to PICC migration, insertion can be difficult and often requires multiple adjustments in order for the tip of the catheter to be correctly placed (3). Only 66% of catheters are inserted correctly the first time and 2-10.5% of catheters dislodge throughout the course of implantation (3,6). To determine and monitor the location of the catheter, clinicians utilize X-ray imaging. Despite X-ray being the gold standard, neonates are particularly at an increased risk from prolonged radiation exposure involved in X-ray imaging, including proclivity to develop lymphoma and other forms of cancer at a later stage of their life (7-12). Thus, there is a clear medical need for catheters that can be imaged without the use of ionizing radiation in order to avoid any inherent risks to the developing child.

An attractive alternative to X-ray imaging is near infrared (NIR) imaging that allows images to be acquired without harmful side effects (13). The main tissue components that absorb light are hemoglobin and melanin which have high absorption bands at wavelengths shorter than 600 nm and water which begins to absorb significant amounts of light at wavelengths above 1150 nm (14,15). Thus, there is a window (between ~ 650 nm – 950 nm) where biological tissue components do not absorb significant light, allowing imaging at depths ranging from 1 - 4 cm (13,16,17). In this article, we report the fabrication of NIR fluorescent enhanced catheters through the integration of a near-infrared sensitive agent, IRDye 800CW, within a polymer matrix. The objective of this study is to demonstrate fluorescent-polymer composites as improved PICC materials, which we anticipate will provide physicians with a safe and effective substitute to imaging catheters without the use of ionizing radiation.

We provide details of the integration of medical grade thermoplastic polyurethane (TPU) with IRDye 800CW extruded as a PICC. Surface and mechanical testing results are reported to show the influence of the fluorescent agent incorporated within the TPU matrix. To test the safety of these altered PICCs in a biological setting, biocompatibility studies were conducted to analyze any adverse effects of the new PICC on endothelial cells.

2. Materials and Methods

2.1 Materials

Aromatic polyether-based medical grade TPU pellets (Texin RxT90A) was provided as a gift from Bayer Material Science (Pittsburgh, PA). IRDye 800CW Carboxylate infrared dye was obtained from LI-COR Biosciences (Lincoln, NE). Phosphate buffered saline powder (PBS, pH 7.4) was purchased from Fisher Scientific and a 1× solution was prepared in milli-Q deionized water (EMD Millipore). A fabricated medical grade PICC (Hospital TPU) was provided as a gift from Cook® Medical (Winston Salem NC, USA). Human Umbilical Vein Endothelial (HUVEC) cells and complete endothelial growth medium (EGM Bulletkit) were obtained from Lonza and prepared according to manufacturer's instructions. Alamar Blue, Calcein AM and Propidium Iodide were purchased from Fisher Scientific.

2.2 Thermal Analysis Characterization and Catheter Fabrication

The thermal degradation temperatures were analyzed to verify that both the TPU and IRDye 800CW would not decompose during the extrusion process. The temperature at which the samples began to decrease sharply in weight was determined to be the onset of degradation. Thermal degradation temperatures were evaluated using a Q50 Thermogravimetric Analyzer (TGA) (TA Instruments, New Castle, DE). Analysis was conducted in nitrogen gas at 20°C/min (n=3).

Thin films of TPU with and without IRDye 800CW (TPU Composite and Plain TPU) were fabricated using a hydraulic platen press (PHI, City of Industry, CA). As illustrated in Fig. 1, 5 grams of TPU with 0.025 wt% IRDye 800CW was pressed for 30 seconds, sectioned into 5 mm squares, and fed into a Haake Minilab Micro Compounder (Thermo Fisher Scientific, Waltham, MA). Catheters were extruded at 100 rpm at 195°C using a custom die fabricated via additive manufacturing (Solid Concepts Inc., Austin, TX). Extruded sections of Plain TPU and TPU Composites were imaged and outer diameter measurements were obtained using calipers (n=3). Inner diameter measurements were obtained using scanning electron microscopy (SEM), and thickness measurements were calculated by subtracting the inner radius from the outer radius.

2.3 Surface Analysis and Mechanical Testing

Scanning electron microscopy (Field Emission SEM, LEO Zeiss 1550, Tokyo, Japan) was used to examine the outer surface and cross-sectional features of the catheters. Outer surfaces and cross-sectional features were imaged before and after retention studies of the extruded tubes. Atomic force microscopy (Veeco MultiMode AFM, Plainview, NY) was used to obtain quantitative outer surface roughness measurements of the Hospital TPU, Plain TPU, TPU Composite, and Leached TPU Composite samples. Surface roughness was measured using contact mode (n=3). Tensile testing was performed using an Instron 5500R (Instron, Norwood, MA) at a cross head speed of 50 mm/min on Hospital TPU, Plain TPU, TPU Composite, and Leached TPU Composite (n=3) samples. To prevent slipping, an Instron clamp with grooved indentations was used. Uniaxial tensile testing was performed on all samples until material failure. The elastic modulus was determined to be the slope

from the linear low strain region (0 to 10%) of the curve. The point of fracture was determined to be the ultimate tensile strength (UTS).

2.4 Retention Studies, Fluorescence Imaging, and Photodegradation Analysis

To simulate the long-term effect of being implanted *in vivo*, catheters were leached in PBS for 23 days to determine the amount of dye retained within the matrix. TPU Composite tubes were cut into thin slices, weighed, and added to a black 96 well plate containing 200 μ l PBS. Leaching of IRDye 800CW from the TPU Composite (n=8) was analyzed under physiological conditions (pH $\sim$ 7.4, 37°C, with gentle agitation) in a water bath. The water bath was covered to prevent photobleaching. Each day, tube slices were transferred to the successive well containing fresh PBS, and the previous day's saline was analyzed using a microplate reader (BioTek Multi-Mode, Winooski, VT) with excitation at 765 nm, emission at 794 nm, and sensitivity at 100. To determine the amount of IRDye 800CW retained, a calibration curve containing serial dilutions of IRDye 800CW in PBS was used (0 to 0.00030 wt%) ($R^2 = 0.99$).

In order to ensure measurements were sensitive, uniform, and low in noise interference, imaging was performed using a LI-COR Pearl® Impulse NIR Imaging System (Lincoln, NE) with analysis conducted in LI-COR Pearl® Impulse Software to compute the signal-to-noise ratio (SNR) (Mean of Sample/Standard deviation of Background). All imaging was performed using a thermoelectrically cooled charged cooled detection camera with the following specifications: laser wavelength was 785 nm, resolution was 85 μ m, and acquisition speed was less than 30 seconds per scan. To determine the optimal loading concentration, thin films of TPU containing 0.025, 0.075 and 0.125 wt% IRDye 800CW were placed in the LI-COR Pearl® and imaged. Superflab® tissue mimic (Radiation Products Design Inc. Albertville, MN) was placed on top of the thin films (up to 2 cm thick) to determine the imaging resolution.

For fluorescence imaging of Plain TPU and TPU Composite samples, Plain TPU (n=1) and TPU Composite (n=4) was placed in the LI-COR Pearl®, automatic shape drawing of each sample occurred at 0 cm (without Superflab®) and imaged using the specifications described above. The shapes were copied during successive imaging under 1, 2, and 3 cm of Superflab®. Minor rotation of the shapes was performed between the 0 cm and 1 cm images if deemed necessary due to slight rearrangement that occurred when applying the first layer of Superflab®. Leached TPU Composite (n=4) was hydrated in PBS (24 hours) to simulate physiological conditions and imaged as described. Contrast enhancement factors were calculated by dividing the SNR of TPU Composite and Leached TPU Composite by the SNR of Plain TPU. Standard deviations were calculated from SNR of the four samples and scaled by the background noise.

For investigation of photodegradation, TPU Composite tubes (containing 0.025% IRDye 800CW) were placed 6 inches beneath a 13-watt halogen light source for eight days. Samples were removed (0 hr, 1 hr, 4 day, 8 day), scanned in the LI-COR Pearl® using the specifications above, and SNR values computed as described. Error bars represent variation within a sample as the LI-COR system determines the signal at each pixel within each individual sample.

2.5 Biocompatibility Studies

Biocompatibility studies were conducted to determine the toxicity of TPU Composite in direct contact with endothelial cells as well as the adhesion of endothelial cells to the TPU Composite. Pressed films (Plain TPU and TPU Composite) were sterilized by washing in 1× PBS for 24 hours under constant agitation (Argos Rotoflex), followed by a 30 minute soak in 100% ethanol and two 1 hour rinses with sterile PBS. Biocompatibility studies were based off work previously completed by Chan-Chan et al. (18). Briefly, 12-well cell bind plates were seeded with HUVEC cells (passage 4-10, cultured in complete endothelial growth medium EGM Bulletkit, Lonza) at a density of 100 cells/cm² for 12 hours (37°C, 5% CO₂) to allow for adhesion. Films (19 mm) were placed in direct contact with the cells and incubated for an additional 72 hours, with daily replacement of media. Toxicity was quantitatively analyzed with alamar blue according to manufacturer's protocol. Briefly, 100 µL of alamar blue was added to the media and allowed to incubate for 1.5 hours. Fluorescence of the alamar blue in each well was read at excitation 545 nm, emission 590 nm. Films were removed from wells and cells were stained with Calcein AM and propidium iodide according to manufacturer's protocol, fixed with 4% paraformaldehyde for 1 hour, and rinsed three times with PBS for qualitative analysis of cell death. Cells were imaged with fluorescence imaging (Zeiss – Axio Observer.A1) for viability. Studies were repeated with 0.025 wt% IRDye 800CW in media, cells with media as a positive control, and cells with 70% ethanol in media as a negative control.

To determine if endothelial cells would adhere to the catheters, 19 mm films were cut and affixed to the bottom of suspension 12-well culture plates with 50 µL of 10 mg/mL collagen Type I isolated from rat tails (19). Plates were incubated for 30 minutes to allow for collagen polymerization. Films were seeded with 100 cells/cm² and incubated for one hour. Wells were washed with PBS to remove non-adherent cells and stained with Calcein AM and propidium iodide to aid in visualization of cell binding. The number of adherent cells was analyzed with fluorescence microscopy and compared to positive (collagen plates) and negative (Teflon) controls (20).

2.6 Statistical Analysis

All statistics were performed using Origin® 8.0 analysis of variance (ANOVA) to compare between groups. Tukey's post hoc test was used in conjunction with ANOVA for all sample analysis. Differences were recorded to be statistically significant at $p < 0.05$. All errors are given as standard deviations.

3. Results

3.1 Thermal Analysis Characterization and Polymer Processing

TPU and IRDye 800CW displayed very high onset degradation temperatures with TPU degrading at 283 ± 8 °C and IRDye 800CW degrading at 308 ± 10 °C (Supplemental Fig. 1). Both temperatures were considerably higher than the processing temperature of TPU (195 °C). A custom annular die (Supplemental Fig. 2) was fabricated out of stainless steel to produce hollow tubes. Hospital TPU samples had the smallest average outer diameter (2.56 ± 0.04 mm) compared to the extruded samples (Plain TPU, TPU Composite, Leached TPU

Composite) (Table 1). Furthermore, extruded samples were thicker than Hospital TPU (Table 1, Fig. 2).

Extruded samples appear visually smooth and transparent and were nearly indistinguishable from the Hospital TPU samples (Fig 2A-D). TPU Composite tubes were slightly darker in color than the unmodified polymer tubes suggesting the fluorescent agent does not significantly alter the appearance of the TPU (Fig. 2B, 2C). The curve in the extruded samples was due to the extrusion collection procedure. SEM images of extruded samples displayed irregularly shaped cross sections compared to the circular Hospital TPU (Fig. 2E-2H).

3.2 Surface Analysis and Mechanical Testing

Surface morphology between extruded samples and Hospital TPU appeared similar, consisting of defined grain boundaries throughout the microstructure (Fig. 2I – 2L). TPU Composite tubes contained light precipitates dispersed throughout the polymer surface demonstrating the presence of fluorescent agent (Fig. 2O, 2P). Quantitative roughness measurements (Table 2) obtained from AFM contact mode revealed that Hospital TPU had the smoothest surface while Plain TPU contained roughness values that were statistically significant compared to all other samples ($p < 0.05$). No statistical significance in roughness existed between TPU Composite and Leached TPU Composite tubes compared to Hospital TPU, suggesting the addition of fluorescent agent does not alter roughness morphology. Furthermore, the mixing of the fluorescent dye with TPU acts as a plasticizer, smoothing rough areas during the extrusion process as supported by the increased roughness in Plain TPU samples.

During tensile testing, failure occurred at the clamped ends of all samples. Samples that slipped before failure were not included in data analysis. Hospital TPU had the highest average elastic modulus (1.87 ± 0.19 MPa), while TPU Composite had the lowest elastic modulus (0.17 ± 0.005 MPa) (Table 3, Fig. 3). Though the Hospital TPU elastic modulus and ultimate tensile strength (UTS) were significantly higher compared to the extruded samples, there was no statistical difference within the extruded samples. This suggests the addition of IRDye 800CW does not alter the mechanical properties of the TPU.

3.3 Retention Studies, Fluorescence Imaging, and Photodegradation Analysis

Daily analysis of PBS from TPU Composite tubes showed total loss of $6.35 \pm 5.08\%$ IRDye 800CW from within the polymer matrix over a 23 day period (Fig. 4). Optimal loading level of IRDye 800CW was determined to be 0.025 wt% as higher concentrations (0.075 wt%, 0.125 wt%) resulted in quenching of the fluorescence signal (Supplemental Table 1, Supplemental Fig. 3).

Fluorescent scans of the TPU Composite tubes resulted in a 14-fold increase in SNR compared to the Plain TPU tubes. This contrast enhancement allows clear imaging of the extruded tubes up to depths of 1 cm (Fig. 5). A 50% reduction in signal was observed between the leached and non-leached samples. Non-leached and leached samples were significantly different within each group at every depth ($p < 0.05$) (Fig. 6). Regardless of the

decrease in signal, individual leached samples were clearly imagable at 1 cm with some visible fluorescence at 2 cm.

Photodegradation studies revealed no significant loss of signal over an eight day period (Supplemental Fig 4). Variation in the SNR was observed due to changes in diameter of the extruded samples. Repeated imaging studies revealed no loss in signal when samples were imaged multiple times (data not shown).

3.4 Biocompatibility studies

Typical biocompatibility studies involve placing the material of interest in direct cell contact for a set time period. In our studies, after a 72 hour incubation of HUVECs with IRDye 800CW (0.025 wt%), Plain TPU, and TPU Composite, no statistical difference was seen in cell viability as shown in Table 4. The majority of cells were viable for IRDye 800CW, Plain TPU, and TPU Composite, and confirmation included Calcein AM and Propidium Iodide Staining (Table 4, Fig. 7). Viability values were normalized to the media control values.

In order to be a viable biomaterial, cell adhesion should be minimal in order to avoid excess damage when removing or inserting the PICC. Cells preferentially adhered to Collagen I, a protein found in the native microenvironment of the extracellular matrix. Cells increased substantially in the area due to spreading with extended lamellipodia demonstrating their affinity for the material (Fig. 8 A1). The rounded shape of the cells with no extended protrusions indicated weak adherence to the negative control (Teflon) as well as the Plain TPU and TPU Composite (Fig. 8 A2-4). The number of adhered cells was counted using Image J particle analyzer software (NIH) from 6 images and normalized to Collagen I. Cell adherence to Teflon, Plain TPU and TPU Composite were significantly lower from Collagen I but were not significantly different between each other (Fig. 8B).

4. Discussion

We fabricated fluorescent PICCs as a proof of concept, demonstrating a promising viable alternative to ionizing radiation to visualize catheters. NIR fluorescent dyes are ideal contrast agents due to their excitation in the desired biological window (between ~ 650 nm – 950 nm) where absorption by tissue components is minimized. This property allows enhanced penetration of light into the tissue (13, 16). Though the FDA approved fluorescent dye indocyanine green (ICG) has been used as a contrast agent and more recently for lymph node mapping, it has limited capabilities in functionalization and imaging depth. However, IRDye 800CW is an NIR dye that has a similar chemical structure to ICG, but allows for chemical functionalization and has a 20× enhancement in brightness making it suitable for deeper imaging applications (13). While IRDye 800CW is not FDA approved, it has undergone pre-clinical trials in animals with great success and is poised to enter human clinical trials in the near future (13). For visualization of this contrast agent, many instruments already exist and have regulatory clearance including the Zeiss Pentero and Leica FL800 (13). Therefore, the evaluation and use of IRDye 800CW in our studies is sensible for realistic future clinical implementation. Additionally, medical grade catheters have numerous additives incorporated in their manufacturing protocol such as barium

stabilizers for processing protection, antioxidants to prevent premature degradation, and anti-microbial coatings (21). Therefore, the inclusion of a fluorescent agent within this process is within the scope of standard manufacturing procedures.

With regard to catheter fabrication procedures, thin films were produced prior to extrusion to enhance incorporation of IRDye 800CW within the polymer matrix. Extrusion is a common polymer processing technology used to produce numerous types of catheter designs incorporating various materials for improved performance. An extruder die that forms hollow tubular structures has not previously been machined for tabletop compounders such as the Haake Minilab (~ 5 g hopper) used in this study. Therefore, a novel die design was constructed from Solid Concepts (Supplemental Fig. 2). Due to the resolution of the additive manufacturing process, approximately 2.7 mm outer diameter tubes were the smallest size capable of being manufactured, which is larger than the 0.67 mm outer diameters of standard neonatal PICCs. Extruding a smaller catheter would require a significantly reduced lumen, resulting in higher pressures. The current manufacturing technique could not fabricate a die that could sustain the pressure and still result in a hollow tube. As a result of this limitation, a 2.7 mm outer diameter PICC obtained from Cook® Medical was used as a control (Hospital TPU). Smaller dies are commercially available for larger extrusion systems; therefore we do not see this as a roadblock to future fabrication for such catheters.

From the optical and microscopy images, the fluorescent agent does not significantly change the surface features of the tubes, as the fluorescence enhanced samples remain nearly indistinguishable from the unmodified TPU counterparts. Similar surface features suggest that the functionality of the novel catheter will yield similar results in situ. PICCs must navigate through complex vascular blood vessels to reach their destination, so the ability of a catheter to be easily manipulated is directly related to its stiffness. The catheter tip must be soft enough that it does not cause damage as it progresses through vascular pathways yet strong enough to respond to manipulation (22). From the results, the samples containing the fluorescent agent (TPU Composite and Leached TPU Composite) were slightly softer compared to the Plain TPU; however, no significant difference exists between the elastic modulus and UTS of the three groups. Although a significant difference in elastic modulus and UTS exists between the Hospital TPU and extruded samples, the Hospital TPU might have been processed using a stiffer TPU or contain additives that altered its material properties. Another important property of a PICC is the surface morphology. Significant roughness alterations would change the hydrophobicity of the material, enhancing protein and cell adsorption to the tube ultimately decreasing catheter “ease of removal” (23). Results here indicate that addition of fluorescent agent does not significantly alter the roughness of the fabricated tubes as the composite samples have similar roughness measurements compared to the Hospital TPU samples.

In order for the catheters to be viable, they must be able to be imaged repeatedly as standard PICC lines can be implanted for weeks to months and are imaged on a weekly basis. Repeated imaging of fabricated catheters resulted in no change in fluorescent signal over time. Variation in signal was due to diameter variations of the catheters resulting from limitations of the die used for extrusion. One inherent limitation of IRDye 800CW is its instability in bright light and need for refrigeration. Photodegradation studies of our TPU

Composite at room temperature revealed stability of the fluorescent signal over an eight-day period. These results suggest that the TPU provides a protective coating over the IRDye 800CW preventing it from degrading or photobleaching. An inherent limitation in NIR imaging is the capability to retain adequate resolution with respect to depth. In our experiment, the extruded tube samples are clearly visible up to 1 cm. As this study tested fluorescent capable PICCs for prospective use in neonatal patients, the depth of imaging does not need to be significantly high (3-5 cm) to investigate PICC placement and location. Possible solutions to increase depth resolution could be the use of high wt% IRDye (by testing various ranges of films) and the use of more sensitive fluorescent microscopes, such as the Zeiss Pentero and Leica FL800 might prove advantageous. The TPU Composites display a burst release in loss of dye within the first 5 days during the leaching studies with minor release over the next 18 days. A pre-leach step could be incorporated into the sterilization and packaging process of these PICCs. The ~3.5% loss of the fluorescent agent after 24 hours was due to the physical blending of the fluorescent dye with the TPU. The outermost layer of dye leached out while the dye that remained trapped within the TPU matrix was retained throughout the duration of the study. There was no chemical interaction of the fluorescent dye with TPU as the carboxylate dye contains no reactive groups allowing for conjugation to the TPU. The loss of dye resulted in a 50% reduction in fluorescent signal at 0 and 1 cm between leached and non-leached samples. From viewing the 0 cm images (Fig. 6), it appears the dye on the PICC surface is washed away resulting in the loss of signal. Manufactured PICCs no longer appear translucent, but can be white, suggesting that during processing, materials are coated. Therefore, the manufacturing process might allow for further dye to be retained in the PICC by the formation of a layered coating, which might prevent dye leaching.

In order for these composite catheters to be an alternative biomaterial, they must demonstrate equivalent biocompatibility to the medical grade equivalent. The TPU Composite did not result in a decrease in cell viability compared to media or its individual components (Plain TPU and IRDye 800CW). These results indicate that the combination of TPU and IRDye 800CW had no adverse effects on endothelial cells, the predominant cell type the material will be in contact with in situ. Catheters must traverse blood vessels, remain implanted and be removed with minimal damage occurring to the inner lining of blood vessels. In order for this to occur, minimal adhesion of endothelial cells to the biomaterial should be observed. From our results, less than 10% of cells adhered to the TPU Composite after one hour. Those cells that did adhere, bound very weakly to the material with no cell spreading observed. TPU is a relatively bioinert material commonly used for long term PICC use, the addition of IRDye 800CW did not significantly alter the surface properties or toxicity of the material. These results indicate that minimal damage will occur to endothelial cells when the catheters are implanted or removed.

Conclusion

Preliminary results suggest the incorporation of a NIR fluorescent dye (IRDye 800CW) at 0.025 wt% with medical grade TPU can be successfully extruded and imaged, confirming presence and conservation of dye function. Addition of the fluorescent contrast agent does not significantly affect the surface or mechanical properties of the medical grade TPU.

Furthermore, the TPU provides a protective effect to the IRDye 800CW preventing photobleaching and degradation in bright light and warmer temperatures. Contrast enhanced catheters can be clearly imaged at depths up to 1 cm using the LI-COR Pearl®. This proof of concept study shows that near infrared enhanced catheters may provide a potentially attractive alternative to the use of ionizing radiation for PICC line monitoring. Future work includes long term assessment of IRDye 800CW stability, implantation and monitoring of TPU composites in an animal model and improved fabrication of extruded samples through collaboration with a local catheter manufacturer.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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Fig. 1.
Schematic of the fabrication process for Composite Catheters.

Fig. 2.

Optical images (A-D) and SEM micrographs (E-P) of polymer samples. Hospital TPU (A) appears perfectly round and smooth. The extruded samples (B, C, D) appear smooth and optically transparent similar to the Hospital TPU with the composite samples being nearly indistinguishable from their unmodified counterparts. SEM micrographs consist of cross sectional view (E,F,G,H), top view (I,J,K,L), and roughness profiles (M,N,O,P).

Collectively, the extruded samples have large diameters and thicknesses compared to the Hospital TPU due to the extruder die design (Table 1). Plain TPU (F), TPU Composite (G) and Leached TPU Composite (H) have irregular cross sectional slices due to swelling and sample collection during extrusion. Top view and roughness images between all samples appear similar. Optical image scale bar = 7.5 mm, cross sectional and top view scale bar = 200 μ m, roughness image scale bar = 600 nm.

Fig. 3. Mechanical Properties of Catheters. Elastic modulus (Table 3) was determined by the slope of the linear region between 0 to 10% strain. The UTS (Table 3) was determined to be the point at which the samples fractured. Error bars represent standard deviations for every 100 data points per sample (n=3).

Fig. 4. Retention analysis of IR Dye 800 CW within TPU matrix. $6.35 \pm 5.08\%$ of the IR Dye 800 CW was released from the polymer over 23 days. Inset is the same data with axis adjusted for clarity. The majority of the dye released as a burst within the first five days (5.40%) followed by minimal leaching throughout the duration of the study. Error bars represent standard deviations ($n=8$).

Fig. 5. Fluorescence intensity scans of Plain TPU, TPU Composite, and Leached TPU Composite. Samples were imaged at an excitation wavelength of 778 nm. 0, 1, 2, 3 cm correspond to the imaging depth or the thickness of Superflab covering the samples that the imaging probe penetrated.

Fig. 6.

Contrast Enhancement intensity factor of TPU Composites. The fluorescence intensity decreases as a function of depth, though signal is still observed at 3 cm. All values are statistically different within the non-leached and leached samples. All values are statistically different between the non-leached and leached samples except for at 3 cm. Error bars represent standard deviation (n=4). Asterisk (*) represents statistically significant data ($p<0.05$).

Fig. 7. Biocompatibility of thin films with HUVECs. Cells are stained with Calcein AM (green) to indicate viable cells and propidium iodide (red) to signify dead cells. Incubation of cells with Plain TPU, TPU Composite and 0.025 wt% IR Dye resulted in a non-significant difference in viability compared to the control. No apparent changes in morphology or proliferation were observed due to the polymer composite, polymer or IRDye 800CW.

Fig. 8. Adhesion studies of HUVECs seeded directly on top of substrates. (A1-A4) Typical morphologies of HUVECs stained with Calcein AM (green) on respective substrates. Collagen preferentially adhered to Collagen I with substantial cell spreading. Cells weakly attached to Teflon, Plain TPU, and TPU Composite as evidenced by the round shape with no extended lamellipodia. (B) Normalized numbers of cells adhered to substrates over a 60 minute time period. Cells had a high affinity for collagen I, one of the major proteins found in HUVECs native microenvironment. Minimal adherence occurred for Teflon, Plain TPU, and TPU Composite. Adherences of cells for the three films (Teflon, Plain TPU, TPU Composite) are statistically different from the collagen with no difference observed between the three polymer films.

Table 1
Outer and Inner Diameters and Thickness Measurements

Sample	Outer Diameter (mm)	Inner Diameter (mm)	Thickness (mm)
Hospital TPU	2.56±0.04	1.88±0.01	0.34±0.01
Plain TPU	2.78±0.09	1.28±0.11	0.75±0.01
TPU Composite	2.84±0.11	1.28±0.21	0.78±0.05
Leached TPU Composite	2.84±0.08	1.19±0.03	0.83±0.03

Table 2
Catheter Roughness (Ra) measurements

Sample	AVG Ra (nm)
Hospital TPU	4.86 ± 1.38
Plain TPU	$19.07 \pm 7.36^*$
TPU Composite	7.34 ± 1.78
Leached TPU Composite	6.52 ± 2.42

* Asterisk denotes statistically different ($p < 0.05$) to all other samples, $n = 3$

Table 3
Catheter Mechanical Property Measurements

Sample	AVG Elastic Modulus (MPa)	AVG UTS (MPa)
Hospital TPU	$1.87 \pm 0.19^*$	$88.1 \pm 8.58^*$
Plain TPU	0.19 ± 0.02	56.4 ± 17.6
TPU Composite	0.17 ± 0.005	50.0 ± 10.3
Leached TPU Composite	0.23 ± 0.03	62.22 ± 19.6

* Asterisk * denotes statistically different ($p < 0.05$) to all other samples, $n = 3$

Table 4**Biocompatibility Results**

Sample	Normalized Viability (%)
Control (Media)	100 ± 5.13
IRDye 800CW	91.27 ± 8.54
Plain TPU	94.79 ± 3.26
TPU Composite	92.34 ± 3.93