A Microneedles Balloon Catheter for Endovascular Drug Delivery

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Disorders of the inner parts of blood vessels have been significant triggers of cardiovascular diseases (CVDs). Different interventional methods have been employed, from complex surgeries to balloon angioplasty techniques to open the narrowed blood vessels. However, CVDs continue to be the lead cause of death in the world. Delivering a therapeutic agent directly to the inner wall of affected blood vessels can be a transformative step toward a better treatment option. To open the door for such an approach, a catheter delivery system is developed based on a conventional balloon catheter where a fluidic channel and microneedles (MNs) are integrated on top of it. This enables precise and localized delivery of therapeutics directly into vessel walls. Customizable MNs are fabricated using a high-resolution 3D printing technique where MN’s height ranges from 100 to 350 µm. The MNs penetration into a synthetic vascular model is investigated with a computerized tomography scan. Ex vivo tests on rabbit aorta confirm the MN-upgraded balloon catheter’s performance on real tissue. Delivery of fluorescent dye is accomplished by injecting it through the fluidic channel and MNs into the aortic tissue. The dye is observed at up to 180 µm of depth, confirming site-specific endovascular delivery.

1. Introduction

For the last decades, cardiovascular diseases (CVDs) have been the leading cause of death in the world, with more than 17 million deaths per year, according to the World Health Organization.[1,2] The underlying process of CVDs is related mainly to the narrowing of the blood vessels, which is identified as atherosclerosis. Disorders of the inner diameter of the heart arteries lead to angina, heart attack, and other coronary artery diseases. Statistically, atherosclerosis is responsible for more than 75% of CVD deaths.[3]

Historically, bypass surgery, angioplasty, and stents were three significant discoveries that marked the CVD timeline and drastically improved disease treatment. A key breakthrough was achieved by the development of the balloon catheter for dilation of obstructed blood vessels, which is called angioplasty and known as...
percutaneous transluminal angioplasty (PTA).[4] PTA is a minimally invasive intervention that consists of repetitive balloon inflation inside the narrowed artery compressing the fatty layer to the vessel wall. The main drawback of PTA is restenosis, which is the re-narrowing of the treated vessels. A follow-up angiography of patients six months after successful PTA intervention showed restenosis in 33.6% of the patients.[5] Residual restenosis was observed immediately even after two balloon dilation trials in 10% of the patients.[6]

In 1986, 9 years after the balloon catheter development, the world had seen the first coronary stent,[7,8] that is, an implantable metallic tube deployed against the narrowed artery wall providing a permanent blood circulation. In a comparative study, patients treated with bare-metal stent (BMS) had a lower restenosis rate than those treated with balloon angioplasty (31.6% versus 42.1%, respectively).[9] BMS had a lower restenosis rate compared to balloon angioplasty.[10] However, in-stent restenosis and other complications like stent occlusion persist.[11] Cell proliferation and other effects in the contact area between the stent and artery lumen led to the in-stent restenosis.[12] Thus, a drug-eluting stent (DES) technology emerged, where the stent was coated with a therapeutic agent to prevent in-stent restenosis. A long-term (up to 3 years) study of DES versus BMS revealed a lower risk of restenosis, revascularization, and coronary surgery with DES compared to BMS.[13,14] However, it has been shown that the risk of death and myocardial infarction was significantly higher among patients with DES after the first 6 months.[11] The long-term risks associated with DES were attributed to incomplete neointimal coverage, stent thrombosis, and the temporary prescription of antiplatelet treatment.[12,13] The newer generations of DES improved in different aspects such as geometry, stent material, polymer coating, and drug formulation.[15–17] However, compared to BMS, the improvement regarding the risks of stent thrombosis was marginal.[16,17] The fact of having a foreign scaffold material in contact with the arterial tissue over a long time causes vascular inflammation, neointimal formation, and a high risk of thrombosis. For these reasons, technologies like the drug-coated balloon (DCB) gained more relevance recently.[18]

Despite not leaving an implant inside blood vessels, DCBs were designed to prevent restenosis by a single dose transfer of an antiproliferative agent under balloon inflation.[18,19] Paclitaxel is one of the most used drugs in the DCBs in the market.[20,21] Nevertheless, DCBs still have many limitations related to the type of drugs, dose/concentration, and drug washout in the bloodstream.[21–23]

In summary, previous techniques faced two main challenges; the use of a foreign implant that induced late restenosis/thrombosis and a deficiency in providing a sustainable targeted drug delivery. Moreover, PTA with or without the stent are temporary solutions as they are not aimed to target the fatty layer accumulation, which is the leading cause of the disease. It has been demonstrated that human atherosclerosis starts in the deep intima layer of blood vessels, which makes systematic drug delivery more challenging.[23–25] Therefore, by-passing the first layer of the blood vessel lumen will facilitate drug delivery and disease targeting.

To the best of our knowledge, only one research group attempted to use a hollow microneedle (MN) for endovascular drug delivery.[26–28] For this purpose, Mercator Medsystems, Inc. has developed the Bullfrog micro-infusion catheter.[26] The device comprises an MN (900 μm long, 140 μm diameter) sheathed inside the balloon catheter. The system has two ports; one for the balloon inflation using a saline solution and the other for the drug injection. During balloon inflation, the MN is uncovered and pushed into the vessel wall. Then, a therapeutic agent is injected into the adventitial tissue. The micro-infusion catheter showed a significant reduction of luminal stenosis after administering nab-rapamycin in a porcine femoral artery.[27] In another investigation, the Torisel infusion in swine superficial femoral arteries using the Bullfrog catheter showed decreased cell proliferation in the treated segment during the test period (28 days), and the experiments proved the safety and efficacy of the system.[28] The limitations of the Bullfrog device are related to the MN size (900 μm long) and configuration. The size limitation restricts the use of the device to large vessels and delivers only to the outer layer. The deployment configuration of the balloon allows a single MN use, which requires multiple injections, more balloon maneuvers, and longer time. Previously, we have been working extensively on 3D printed MNs for biomedical and biological applications.[29–34] Particularly, we have demonstrated the use of 3D printed MNs to penetrate mouse skin and deliver a fluorescent dye to the targeted level in the tissue.[34]

In this study, we propose 3D printed hollow MNs for endovascular drug delivery. The MNs are integrated on a commercial balloon catheter, and drug delivery is enabled via a fluidic channel connected to the MNs (Figure 1). The MNs are sheathed inside the balloon folds when fully deflated, which protects them from possible damage. In addition, the folded balloon is introduced inside a guiding sheath. The balloon catheter and sheath are guided safely through the blood vessels to

Figure 1. Concept of the MNs balloon catheter delivery system. a) Sheathed MNs between the deflated balloon folds under the lesion area. b) Balloon inflation, MNs penetration, and drug delivery. Insets: 3D views.
the targeted area of the disease (Figure 1a). Then, the balloon is advanced out of the sheath and inflated with a saline solution allowing the MNs to penetrate the vessel wall. Therapeutic agents are then injected directly into the lesion site (Figure 1b). Finally, the balloon is deflated, securing the MNs in its folds, retracted inside the sheath, then retrieved from the body (Video S1, Supporting Information).

Thanks to the 3D printing technology used, the MNs’ dimensions and geometry can be easily tailored to accommodate the patients’ specifications, targeted tissue, penetration depth, etc. Moreover, the 3D printed MNs can be customized to fit on any commercial balloon catheter on the market.

2. Results and Discussion

2.1. Balloon Folding and Delivery Process

Originally, the balloon is manufactured to get wrapped around the catheter shaft when deflated. This is made using special machines for balloon pleating and folding. Especially for our design, the pleating and folding of the balloon can be modified to obtain a symmetrically folded state to sheath the MNs in between. Before assembling the 3D printed MNs and fluidic channel, the deflated state of commercial balloon catheters were modified by folding the balloon accordingly to our design (Video S2, Supporting Information). A guiding sheath is used to guide the folded balloon catheter safely to the vessel’s narrowed area. The sheath protects the vessel wall from any possible risks. When the balloon is fully deflated, the MNs are drawn between its folds. Note that the folds become higher than the MNs when the balloon is deflated. At this point, the balloon catheter can be moved safely in and out of the sheath. (Figure 2 and Video S2, Supporting Information).

Imaging techniques like optical coherence tomography (OCT), intravascular ultrasound (IVUS), and quantitative coronary angiography (QCA) help to quantify, measure, and visualize coronary stenosis. As the narrowing can be located at different sides inside blood vessels, the proposed catheter can be rotated to position the MNs toward the lesion side. Balloon catheters come with radiopaque markers that help to localize the balloon when it is inside the body using an X-ray. Radiopaque markers can be modified to indicate the MNs tip direction. A minus sign marker can be made parallel to the 3D printed fluidic channel from the MNs side. This type of markers can be used as a reference in case of rotation of the device inside the blood vessels. In addition, the MNs can be coated with gold for better contrast under X-ray imaging.

Nevertheless, depending on the lesion, the device rotation might be avoided as the injected drug can diffuse and percolates circumferentially and longitudinally. Gasper et al. used the Bullfrog catheter, which has only one needle for adventitial drug delivery. To visualize the drug distribution, a 4:1 mixture of the drug to contrast agent (IsoVue 370) was injected under fluoroscopy. It has been reported that one injection allowed circumferential and longitudinal distribution of the drug within 10 min after delivery.

2.2. Design and Fabrication of MNs Balloon Catheter

The main design criteria of the 3D printed channel were as follows: integration on a balloon catheter, fluidic connection to the tubing, and having microneedles for endovascular delivery. For the integration on the balloon catheter, the bottom side of the channel was made curved with a radius equal to the balloon catheter. The 3D printed channel was open from one side to house a needle channel with an outer diameter of 320 μm. As for the MNs, the array consisted of only one line of MNs to reduce the channel’s width, hence facilitating the balloon folding and sheathing of the MNs. Gasper et al. used the Bullfrog catheter that has only one needle for adventitial drug injection; circumferential and longitudinal distribution of the drug was observed within 10 min after delivery. Nakagawa et al. explored the intimal thickness of the right coronary artery in the early stage of atherosclerosis for subjects with an average age of 37. The intima thickness varied from 250 to 700 μm. In this work, to investigate the penetration depth into a rabbit aortic tissue, the MNs height was designed to gradually increase from 100 to 350 μm with an increment of 50 μm toward the proximal end of the catheter. To ease the system integration of the MNs, they were fabricated together with a fluidic channel as a single part, as shown in Figure 3. The structure can be tailored based on the application; however, in this study, we used six MNs arranged along a line parallel to the catheter’s axis with a pitch of 600 μm. The MN tips’ inner and outer diameters were 19 and 25 μm, respectively. The structure was made of IP-S photosensitive resin, which combines the following features: ability to print feature size ranging from the submicron to the millimeter scale, smooth surfaces, and low shrinkage effect. The assembly of
the 3D printed MNs on the balloon catheter consisted of two steps. First, the 3D printed channel was connected to a 30G stainless steel blunt tip needle using a Loctite 4011 instant glue (Figure 3c). The other end of the needle was connected to a flexible Tygon tubing (Figure 3c). Attaching the 3D printed channel directly to the Tygon tubing was not practical due to the large wall thickness of the available tubing. Direct attachment of the Tygon tubing would result in a step higher than the MNs shaft length, which would impair the penetration. The 30G needle was used as a link between the Tygon tubing and 3D printed channel. Moreover, the Tygon tube allowed a reliable fluidic connection to the MNs at a deflated and an inflated state of the balloon (Figure 3d).}

2.3. Fluidic and Phantom Vascular Tissue Testing

The balloon catheter’s inflation and deflation should not be altered by adding the MNs and fluidic channel onto the balloon’s surface. Furthermore, there should not be any blockage in the fluidic channel and MNs. Consequently, a fluidic test was performed before and after assembling the 3D printed MNs and fluidic channels on top of the balloon catheter (Video S3, Supporting Information). DI water was injected manually using a 5 mL syringe, confirming the continuous fluid flow, as shown in Figure 4a. A second test consisted of multiple inflation/deflation cycles of the assembly set (3D printed MNs channel and tubing attached to the balloon catheter). Although the designed system is intended for a single-use application, it was tested for 20 cycles, and no malfunction was observed.

The objective of the polydimethylsiloxane (PDMS) phantom vascular tissue experiments was to determine the ability to penetrate using different geometrical dimensions of the MNs and to investigate the MNs penetration depth at different balloon pressures. A customized fluidic channel with 10 MNs was designed and assembled to a balloon catheter (balloon outer...
diameter was 3 mm) (Figure 4b). As the elastic modulus of a rabbit aorta ranged from 0.05 MPa to 0.5 MPa,\cite{38,39} a phantom vascular tissue with a higher modulus of elasticity was selected to test the mechanical stability of the designed MNs. The vascular tissue model was made of PDMS with a 10:1 base to curing agent weight ratio that corresponds to an elasticity modulus of 2.61 MPa.\cite{40} From the results in Figure 4b, with a minimum tip diameter and wall thickness of 26 and 3 μm, respectively, the MNs were able to penetrate a phantom vascular tissue with no mechanical failure. The 3D reconstructed image of the X-ray signal is shown in Figure 4b. The 10 MNs penetrated successfully, and different penetration depths were observed, depending on the MNs heights (Figure 4b). This confirms the possibility to tailor the depth of the delivery location within the tissue. The gap between the MNs' base in the center and the PDMS shell is presumably due to the bed of needles effect.\cite{34,41,42}

The final device (Figure 3d) was used to investigate the MNs penetration depth into the PDMS phantom vascular tissue at different balloon pressures. The penetration depth was measured for each MN from the X-ray images, and the results are shown in Figure 5. The reconstructed images of the MNs penetration into PDMS are shown in Table S2, Supporting Information. The MNs penetration depth can be controlled by selecting the MN height and the balloon inflation pressure (Figure 5). For a 250 μm thick PDMS phantom tissue, the MN penetration depth can be precisely controlled from about 35 μm to 240 μm by selecting the right combination of MN height and balloon inflation pressure.

2.4. Ex Vivo MNs Penetration and Delivery into Rabbit Aorta

To assess the MNs penetration into the aortic tissue upon balloon inflation, a histological analysis was performed. Due to minor misalignment of the histological cut the 100 μm high MN (the first MN from left to right) cannot be observed, as shown in Figure 6b. Before the sectioning, the aortic tissue went through a fixation step in formalin, dehydration/clearing process. It was then embedded in a paraffin wax block to finally reach the sectioning step with a microtome. Even though we tried to align the cutting axis with the MNs, a small deviation from it is sufficient to prevent capturing of all 6 MNs penetration sites in one

Figure 4. a) Fluidic test of the fluidic channel and MNs. DI water injection through MNs after the first and second assembly. b) Photograph of the MNs penetration into PDMS and Cross-section view of the X-ray image reconstruction of the MNs penetration into the phantom vascular tissue. Scale bars: 1 mm.

Figure 5. MNs penetration depth into PDMS phantom vascular tissue for different balloon pressures. a) 350 μm long MN. b) 300 μm long MN. c) 250 μm long MN. d) 200 μm long MN. e) 150 μm long MN. f) 100 μm long MN.
section. As shown in Figure 6b, the MNs penetration sites can be clearly seen. With a MN height equal to or higher than 300 μm, the MNs can reach the adventitia of the rabbit’s aortic tissue (the last two spots from left to right (Figure 6b). In comparison, MNs with heights between 250 and 150 μm reached the rabbit aorta’s media layer (the first three spots from left to right (Figure 6b).

Interestingly, the histological image showed no damage or rupture of the tissue, especially in the surroundings of the penetration sites. The sectioned tissue’s fluorescent image showed the rhodamine B dye spreading (Figure 6c). In particular, a higher dye concentration was observed in the MNs penetration sites compared to the surrounding tissue. Based on an image analysis using ImageJ software to quantify the red dye distribution. Arrows indicate the MNs penetration sites from the lumen side. Scale bar: 500 μm.

In the second test, the balloon catheter with MNs was inflated inside a rabbit aorta, and fluorescein isothiocyanate (FITC) dye was injected into the tissue (Figure 7a). After opening the aortic tissue, the MNs punctures were visible on the lumen surface, and the FITC dye injected was apparent as well (Figure 7b).

After each test, the delivery system was examined, and all MN tips were inspected; no damage was observed, confirming the mechanical stability of the MNs (Figure 7c). The fluorescent dye signal was observed from the surface level of the aortic tissue to about 180 μm of depth, and it was spread out around the penetration hole, indicating the delivery inside of the tissue (Figure 7d). The depth of the fluorescent dye signal varied linearly with the MNs height. The penetration depth of the MNs was affected by the high elasticity of the aortic tissue. This is a well-known effect, especially for skin penetration, where most of the MN displacement created a tissue indentation, and only a fraction (10–30%) of the MN height penetrates into the tissue.[41,42] Reed et al. showed a similar result of a sharp silicon microprobe penetration into a rabbit iliac artery. Although the 80 μm high microprobe was sharp, it deformed the internal elastic lamina and did not penetrate through. Penetration was successful with a 140 μm high sharp probe at a pressure of 67 kPa.[43,44] Moreover, the balloon catheter type and size directly affected the MNs penetration depth. Intravascular imaging techniques such as intravascular ultrasound and optical coherence tomography provide accurate measurements of the vessel size and lumen morphology.[45] An investigation of the required balloon to vessel...
lumen ratio would guide the balloon size selection for a precise MN penetration with no vascular complications.

3. Conclusion

We presented a novel concept of 3D printed MNs on a balloon catheter for endovascular drug delivery. The proposed design allows hollow MNs to penetrate and deliver into the targeted area in the vessel wall. The functionality tests of the device confirmed a robust assembly after performing more than 20 cycles of inflation/deflation of the balloon catheter. A fluidic test demonstrated fluid flow through the MNs fed by a fluidic channel along the catheter. A phantom vascular tissue was fabricated to investigate the MNs capability to penetrate the tissue.
for different MNs dimensions. Using an X-ray imaging technique, we confirmed the MNs penetration into the phantom vessel with a minimum MNs' tip diameter and wall thickness of 25 and 3 μm, respectively. Then, the drug delivery feature of the delivery system was assessed with an ex vivo test on a rabbit aorta, utilizing balloon inflation as the mechanism to drive the MNs into the tissue. Fluorescent images showed the penetration depth depends on the MNs' length. Confocal images of the fluorescent dye injected through the MNs into the aortic tissue showed dye signal from the surface level up to 180 μm of depth inside the tissue depending on the MNs' length. Histological analysis of the aortic tissue confirmed the dye distribution and non-invasiveness of the MNs. No mechanical damage was observed during and after the penetration experiments, ensuring the mechanical stability of the MNs. These results show that MN-upgraded balloon catheter is capable of localized and targeted endovascular drug delivery into artery walls. The fabrication process ensures a highly customizable solution that can be tailored to the patient-specific requirements.

4. Experimental Section

3D Printed MNs: A detailed view of the MNs design with all dimensions is provided in Figure S1, Supporting Information. 3D printing was carried out using the Nanoscribe Photonic Professional GT laser lithography system (Nanoscribe GmbH, Germany). A summary of the printing settings is presented in Table S1, Supporting Information. A focused laser beam induced IP-S (Nanoscribe GmbH, Germany) polymerization at 780 nm wavelength, ~150 mW average power, and 100 mm s⁻¹ scan speed (Figure 3a). Subsequently, the 3D printed structure was submerged in a fresh solution of mr-DEV 600 (Microresist Technology GmbH, Germany) for 15 min to clear unpolymerized resist, followed by an additional 15 min in the developing solution under vacuum. Afterward, the sample was submerged in isopropanol (IPA) for 15 min under vacuum and dried with a nitrogen gas stream. Finally, the sample was inspected under an optical microscope (Figure 3b). In the case of residual photoresist inside of the MNs, an additional cleaning cycle was performed.

Balloon Folding: The balloon catheter was deflated and pressed from both sides to make it flat. Then, the balloon was folded symmetrically to the catheter axis and clamped using a mini vise (Figure S3, Supporting Information). Afterward, the clamped balloon and vise were left in the oven for 1 h at 90 °C. The heating temperature and time were optimized based on different trial tests to get a V-shape folded balloon when deflated.

MN Penetration into Phantom Vascular Tissue: A customized fluidic channel with 10 MNs was designed and assembled to a balloon catheter (balloon outer diameter was 3 mm) (Figure S4a,b, Supporting Information). The first 5 MNs (on the left side in Figure S4a, Supporting Information) had a tip diameter and wall thickness of 30 and 5 μm, respectively, while the second 5 MNs (on the right side in Figure S4a, Supporting Information) had a tip diameter and wall thickness of 25 and 3 μm, respectively. The MNs height was gradually increasing from 100 μm to 350 μm, with an increment of 50 μm toward the center. For a better X-ray contrast, the MNs were coated with 100 nm of gold. The PDMS (Sylgard 184 Silicone Elastomer, Dow Corning Corp., MI) cylinder was fabricated by mold casting and had a length, outer diameter, and thickness of 50 mm, 3 mm, and 250 μm (Figure S4c, Supporting Information). Fabrication details are presented in the Supporting Information. Then, the balloon catheter with MNs was inflated inside the cylinder pushing the MNs into the PDMS (Figure S4d, Supporting Information). Finally, a non-destructive method was applied to inspect the 3D printed MNs penetration inside the PDMS shell using a computerized tomography (CT) scanner (CoreTom, XRE-Tescan). For X-ray imaging, the catheter shaft was attached to a sample holder and secured inside the CT scanner. The PDMS shell and the inflated balloon catheter were mapped longitudinally with a resolution of 2 μm.

Ex Vivo Animal Study: To study the MN-upgraded balloon catheter’s potential for drug delivery into aortic tissue, a fresh rabbit abdominal aorta was collected from a white male New Zealand rabbit (3–4 kg weight). The aortic tissue was washed and preserved in a saline solution for the ex vivo experiment within 12 h after euthanasia. A syringe loaded with FITC (Sigma Aldrich, USA) dye was connected to the flexible Tygon tube, which was connected to the channel with the MNs. The deflated balloon catheter was inserted inside the rabbit aorta and inflated (Figure S5, Supporting Information). Successively, FITC was manually injected into the vessel wall. The balloon catheter was then deflated and retrieved. Next, the aortic tissue was cut transversally (about 6 mm long) and opened with scissors longitudinally. Finally, the opened aortic tissue was fixed on a glass slide and imaged with a 10x objective lens on the Leica SP8 inverted confocal microscope (Leica, Germany).

Similarly to the first test, the deflated balloon catheter was guided inside the rabbit aorta, inflated, and a fluorescent dye rhodamine B (Acros Organics, USA) was injected through the MNs into the aortic tissue. The microneedles’ penetration spots were localized with a tissue marking dye that was used as a reference for the tissue sectioning step. Then, the aortic tissue was cut transversally (about 6 mm long). The collected aorta was fixed in 10% neutral buffered formalin for 24 h. Next, the aortic tissue was placed in a cassette and processed through a graded series of alcohols and xylenes. The tissue was then embedded in a paraffin wax block. The formalin-fixed paraffin-embedded (FFPE) sample was sectioned with a microtome at 4 μm thickness (Sakura Finetek, USA). The sectioning was parallel to the MNs penetration line. The sections were mounted on slides and stained with hematoxylin and eosin (H&E) (Merck, Darmstadt, Germany). The cross-sectional area was imaged using an optical microscope with a 10x objective lens (Olympus BX43, Tokyo, Japan).

The animal experiment protocol was approved by the Unit of Biomedical Ethics at King Abdullah University, Faculty of Medicine (Reference No 579-20) and the Animal Care and Use Committee at King Fahd Medical Research Center (ACUC-20-06-15).

Supporting Information
Supporting Information is available from the Wiley Online Library or from the author.

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Conflict of Interest
The authors declare no conflict of interest.

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Research data are not shared.
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