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Tissue Uptake, Distribution, and Healing Response After Delivery of Paclitaxel via Second-Generation Iopromide-Based Balloon Coating

A Comparison With the First-Generation Technology in the Iliofemoral Porcine Model

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Objectives This study sought to evaluate vascular drug uptake, distribution and response of second-generation paclitaxel coated balloon (PCB) (Cotavance, MEDRAD Interventional, Indianola, Pennsylvania) and compare it with first-generation technology, containing identical excipient and drug concentration.

Background Original PCB technologies displayed a heterogeneous deposition of crystalline paclitaxel-iopromide inside the balloon folds, whereas second-generation PCBs consisted of more homogeneous, circumferential coatings.

Methods Paclitaxel tissue uptake was assessed in 20 iliofemoral arteries of a domestic swine. Vascular healing response was assessed in the familial hypercholesterolemic model of iliofemoral in-stent restenosis. Three weeks after bare-metal stent implantation, vascular segments were randomly revascularized with first-generation PCBs (n = 6), second-generation PCBs (n = 6), or plain balloon angioplasty (PBA) (n = 6). At 28 days, angiographic and histological evaluation was performed in all treated segments.

Results One-hour paclitaxel tissue uptake was 42% higher in the second-generation PCBs ($p = 0.03$) and resulted in more homogeneous segment-to-segment distribution compared with first-generation PCBs. Both angiography (percentage of diameter stenosis: second-generation 11.5 ± 11% vs. first-generation 21.9 ± 11% vs. PBA 46.5 ± 10%; $p < 0.01$) and histology (percentage of area stenosis: second-generation 50.5 ± 7% vs. first-generation 54.8 ± 18% vs. PBA 78.2 ± 9%; $p < 0.01$) showed a decrease in neointimal proliferation in both PCB groups. Histological variance of the percentage of area stenosis was lower in second-generation compared with first-generation PCBs (51.7 vs. 328.3; $p = 0.05$). The presence of peristrut fibrin deposits (0.5 vs. 2.4; $p < 0.01$) and medial smooth muscle cell loss (0 vs. 1.7; $p < 0.01$) were lower in the second-generation compared with first-generation PCBs.

Conclusions In the experimental setting, second-generation PCB showed a comparable efficacy profile and more favorable vascular healing response when compared to first-generation PCB. The clinical implications of these findings require further investigation. (J Am Coll Cardiol Intv 2013; 6:883–90) © 2013 by the American College of Cardiology Foundation

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Paclitaxel-coated balloon (PCB) technologies have been clinically introduced as an alternative therapy to drug-eluting stents and plain balloon angioplasty (PBA) for treatment of coronary in-stent restenosis (ISR) (1,2) and de novo peripheral artery disease (3,4). After these promising results, further use of PCBs has been extended to other applications such as de novo coronary lesions (5–9). First-generation coatings, although clinically effective, used manual coating techniques and deposited the drug preferentially within the balloon folds, resulting in nonhomogeneous drug surface distribution. In the past several years, advancements in coating techniques have yielded more reproducible circumferential coatings with higher dosing precision and uniform drug-excipient distribution along the balloon surface. In this series of studies, we compared the pharmacokinetic profile of the original Paccocath technology (first-generation PCB) (Paccocath Technology, Bayer Pharma AG/MEDRAD Inc., Indianola, Pennsylvania) with second-generation PCB in the iliofemoral territory of a healthy swine. In addition, we evaluated the vascular healing response of the same devices in the iliofemoral territory in the familial hypercholesterolemic swine (FHS) model of ISR.

Abbreviations and Acronyms

%AS = percentage of area stenosis
%DS = percentage of diameter stenosis
EEL = external elastic lamina
FHS = familial hypercholesterolemic swine
IEL = internal elastic lamina
ISR = in-stent restenosis
MLD = minimal lumen diameter
PBA = plain balloon angioplasty
PCB = paclitaxel-coated balloon
QVA = quantitative vascular angiography
RVD = reference vessel diameter

Methods

Device description. The first-generation PCB used in this study (Paccocath technology, Bayer Pharma AG/MEDRAD Inc.) was coated using a proprietary dipping process containing a paclitaxel-iopromide formulation (3 μg/mm^2) deposited preferentially in the folds of the balloon. The second-generation PCB used in this study (Cotavance technology, Bayer Pharma AG/MEDRAD Inc.) used a similar paclitaxel-iopromide formulation and concentration. Precise microsyringe circumferential deposition was used, allowing a higher degree of coating uniformity and precision of dosing (data on file at MEDRAD). All balloons and self-expandable nitinol stents used for model creation were 5 to 7 × 20 mm.

Experimental design and procedures. Figure 1 describes the study designs and groups. All experiments were approved by the Institutional Animal Care and Use Committee. Animals received standard of care outlined in accordance with the USDA Animal Welfare Act and the Guide for Care and Use of Laboratory Animals (9). Each animal received dual antiplatelet therapy 1 day before the procedure and continued until termination. General anesthesia was induced with xylazine and tiletamine HCl/zolazepam HCl and maintained with inhaled 1% to 3% isoflurane. Carotid artery access was obtained via cutdown. Before catheterization, heparin (5,000 to 10,000 U) was administered to maintain activated clotting time >250 s. Nitroglycerin was administered intra-arterially to prevent and relieve vasospasm.

Tissue transfer study. A total of 20 iliofemoral arteries (5 domestic swine) were included in the paclitaxel tissue transfer study. Test devices (12 first-generation vs. 8 second-generation) were inflated based on quantitative vascular angiography (QVA)–derived vessel reference diameters to achieve a balloon-to-artery ratio of 1.2:1.0 for 30 s. One hour after the procedure, animals were euthanized, and the treated vessels were harvested for paclitaxel analysis. Tissues were cut into 3 sections (proximal, medial, and distal), weighed, and then homogenized in a phosphate-buffered solution. The homogenate was labeled with an internal standard, extracted, and evaporated to dryness. The reconstituted sample was analyzed by reverse-phase high-performance liquid chromatography with tandem mass spectrometry detection. The assay range for paclitaxel in porcine tissue was 0.3 to 500 ng/g using a nominal sample size of 100 mg of tissue.

Vascular healing response study. IN-STENT RESTENOSIS DEVELOPMENT. The FHS model (10–13) has been used and validated by our laboratory for the evaluation of device efficacy in the iliofemoral territory in de novo (14) and ISR (15) settings. In this study, a total of 5 FHS were included in the vascular healing response study. The protocol for the development of ISR in the peripheral vasculature model of FHS has been described previously (16), and a detailed description of the method is provided in the Online Appendix. A total of 20 iliofemoral arteries (4 in each animal) were screened using QVA to select the appropriate segment for treatment. Two sites were excluded due to inappropriate vessel diameters. All vessels were injured with an oversized balloon followed by self-expandable bare-metal stent implantation (day 0). After 21 days, all stented segments were randomly treated with first-generation PCB (n = 6), second-generation PCB (n = 6), or PBA (n = 6). Four weeks later (day 49), terminal angiography was performed, and arterial tissue was sent for histological evaluation.

ANGIOGRAPHIC ANALYSIS. QVA was performed at all time points using QAngio-XA Software version 7.1.14.0 (Medis, Medical Imaging Systems, Leiden, the Netherlands). The following parameters were measured using the guiding catheter as a standard for calibration: minimal lumen diameter (MLD) within the treated segments, reference vessel diameter (RVD) measured at the proximal and distal portions of the treated sites, and the balloon and stent-to-artery ratio. The percentage of diameter stenosis (%DS) pre-procedure (day 21) and follow-up (day 49) were calculated as: (1 − [MLD/RVD]) × 100% and the late loss was calculated as (MLD at follow-up − MLD at post-inflation).

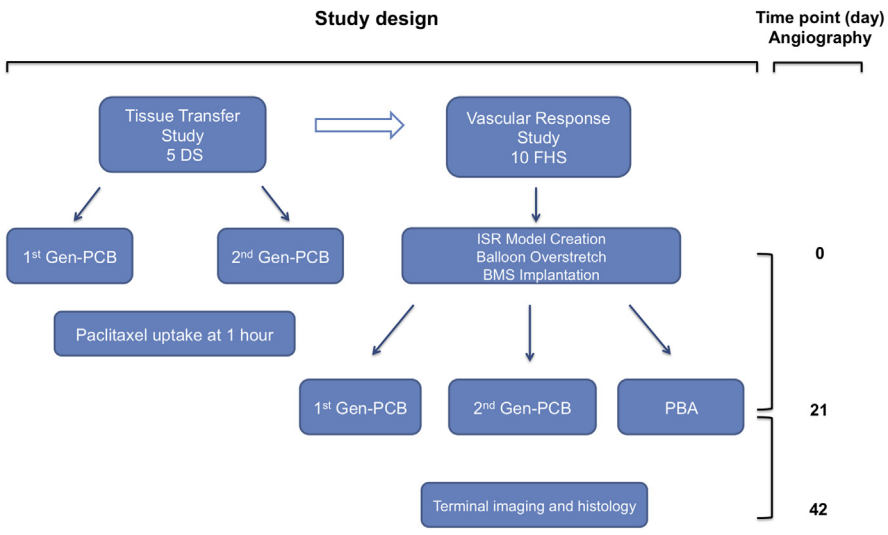

Figure 1. Flowchart of the Study Designs

BMS = bare-metal stent(s); DS = domestic swine; FHS = familial hypercholesterolemic swine; Gen = generation; ISR = in-stent restenosis; PBA = plain balloon angioplasty; PCB = paclitaxel-coated balloon.

HISTOLOGICAL ANALYSIS. The histological analysis was conducted by an independent pathology laboratory (Alizée Pathology, LLC, Thurmont, Maryland). After terminal imaging, animals were euthanized, and all treated vessels harvested and immersed in 10% neutral buffered formalin. All vessels were first embedded in methylmethacrylate and then cut in 40- to 50-μm sections obtained from the proximal, mid, and distal portions of each stented segment. These sections were stained with hematoxylin and eosin and elastic trichrome. The cross-sectional areas (external elastic lamina [EEL], internal elastic lamina [IEL], and lumen area) of each section were measured and used to calculate vessel layer areas with the following formulas: media = EEL − IEL; neointima = IEL − lumen; the percentage of area stenosis (%AS) = (1 − [lumen area/IEL area]) × 100. The criteria of Schwartz et al. (17) and Kornowski et al. (18) were used to evaluate the amount of injury and inflammation. Detailed description of qualitative analysis is provided in the Online Appendix.

Statistical analysis. Normally distributed parametric data are expressed as average and SD or variance, while skewed as median and interquartile range. For continuous and ordinal data, the Levene equal variance and Shapiro-Wilk normality tests were initially performed. When equal variance and normality were observed, 1-way analysis of variance with Holm-Sidak post-analysis of variance or Student *t* test were used to test for differences in variables between balloon types. When either the equal variance test or normality test failed, the Kruskal-Wallis (with Dunn's method for post hoc) or Mann-Whitney *U* test was used. A value of $p \leq 0.05$ was considered statistically significant. An F test of equality of variances was used to assess neointimal homogeneity.

Results

Paclitaxel tissue uptake study. Arterial tissue was harvested 1 h after PCB dilation. The pharmacokinetic profile of all tested groups based on concentration is shown in Figure 2. The baseline overall vessel size was comparable in both groups (data not shown). The 1-h total paclitaxel tissue uptake was 42% higher in the second-generation PCB compared with the first-generation PCB ($p = 0.03$). Segment-to-segment analysis revealed that delivery of paclitaxel from the second-generation PCB resulted in a higher paclitaxel uptake in the proximal and distal portions of the vessel compared with the first-generation PCB (Fig. 2), resulting in more uniform distribution.

Vascular healing response to paclitaxel transfer. ANGIOGRAPHIC ANALYSIS. The angiographic analysis is summarized in Table 1. All angiographic variables were comparable before initial injury. At the time of PCB treatment (21 days), the mean baseline %DS was similar in all groups. The mean final balloon inflation diameters were also similar, leading to a similar angiographic acute gain post-vessel treatment. At last follow-up, the %DS was significantly lower in both PCB groups compared with PBA (second-generation PCB: 75% reduction and first-generation PCB: 52% reduction). Similarly, there was a statistically significant 3- to 4-fold reduction in angiographic late lumen loss in

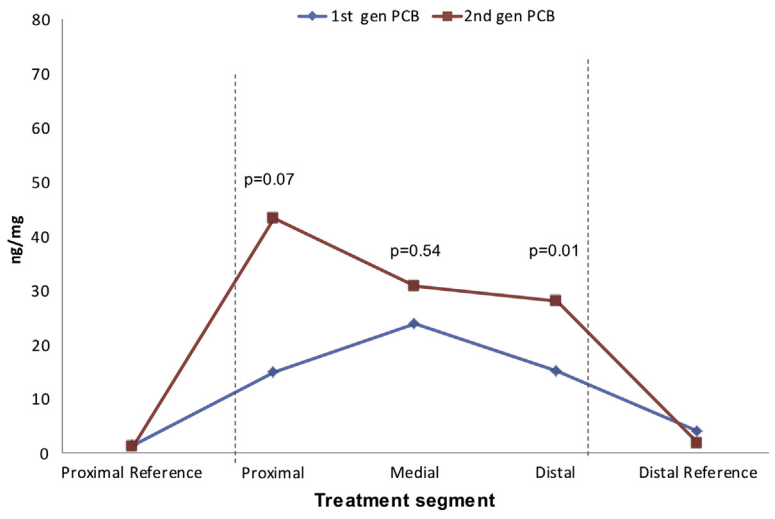

Figure 2. One-Hour Tissue Transfer Study

Paclitaxel acute uptake is higher and distribution more uniform along stented segments after drug delivery via second-generation PCB. Abbreviations as in Figure 1.

both PCB groups compared with PBA. There were no statistical differences in any of the angiographic endpoints between first-generation and second-generation PCBs.

HISTOLOGICAL ANALYSIS. A summary of the histological findings is shown in Table 2. All variables representing neointimal hyperplasia such as neointimal thickness, neointimal area, and %AS were comparable between both PCB groups and significantly lower than PBA (Fig. 3). Neointimal homogeneity defined as the variance of %AS was significantly higher in the first-generation PCB compared with the second-generation PCB (328.3 vs. 51.7; $p = 0.05$). Similarly, the analysis of %AS among sections showed Gaussian-like distribution in the second-generation PCB ($p = 0.59$) and skewed in first-generation PCB ($p = 0.03$) (Fig. 4). Significant restenosis (%AS >75) did not occur in any of the second-generation PCBs, whereas it was found in 1 first-generation PCB stent and in 3 in the PBA control group. There was a tendency toward positive vessel remodeling in the second-generation PCB expressed by higher EEL and IEL areas.

The healing and biocompatibility profile of all tested devices is presented in Figure 5. The peristrut inflammation scores were lower (<1.0) in both PCB groups compared with PBA (>2.0). Neointimal maturity score was highest in the second-generation PCB and PBA groups compared with the first-generation PCB. In the medial layer, smooth muscle cell loss was not observed in the second-generation PCB and PBA groups, whereas evidence of acellularity was present in first-generation PCB. Finally, peristrut fibrin deposits were more commonly seen in the first-generation PCB compared with the second-generation PCB and PBA control groups. Representative pictures of stent cross sections of all 3 groups are presented in Figure 6.

Discussion

First-generation PCB coatings used manual coating techniques and deposited the drug preferentially within the balloon folds, contributing in part to nonhomogeneous drug surface distribution. Early experimental studies suggested this early-generation coating, although clinically effective, displayed inconsistent results in drug delivery and healing response (19–22). In the past several years, advancements in coating techniques resulted in more reproducible circumferential coatings with higher dosing precision and uniform drug-excipient distribution along the balloon surface. In this series of studies, we compared the pharmacokinetic profile of the original Paccocath technology (first-generation PCB) with the second-generation PCB in the iliofemoral territory of healthy swine. In addition, we evaluated the vascular healing response of the same devices in the same vascular territory in the FHS model of ISR.

The use of healthy arteries for evaluating uptake and retention of local drug delivery agents is a well-established model (20,22,23). Although limited by the absence of underlying disease, the anatomic and physiological resemblance to humans has proved this model to be effective in estimating the pharmacokinetic behavior of different drug-eluting devices (20,22,24,25). On the other hand, predicting clinical efficacy in nondiseased animal models has proved to be challenging (23). In this study, we used the FHS model of iliofemoral ISR to assess the vascular response to both PCB coating technologies (16). Previous studies have shown the

Table 1. Summary of Quantitative Vascular Analysis in All Treated Vessels

	Second-Generation PCB (n = 6)	First-Generation PCB (n = 6)	PBA (n = 6)	p Value
Injury (day 0)				
RVD	4.28 ± 0.4	4.24 ± 0.3	4.12 ± 0.7	0.9
BAR	1.23 ± 0.02	1.26 ± 0.02	1.24 ± 0.05	0.54
Post-stent MLD	4.96 ± 0.5	4.8 ± 0.1	4.6 ± 0.2	0.3
Pre-treatment (day 21)				
RVD	4.18 ± 0.5	4.15 ± 0.4	4.04 ± 0.8	0.91
MLD	3.42 ± 0.5	3.28 ± 0.5	2.97 ± 0.5	0.28
%DS	17.9 ± 10.2	20.1 ± 15.6	25.9 ± 9.6	0.51
After treatment				
RVD	3.85 ± 0.4	3.80 ± 0.4	3.71 ± 0.6	0.88
MLD	3.53 ± 0.8	3.74 ± 0.3	3.46 ± 0.5	0.69
Balloon diameter	4.68 ± 0.5	4.51 ± 0.4	4.53 ± 0.5	0.77
Acute gain	0.5 ± 0.11	0.46 ± 0.6	0.50 ± 0.4	0.36
%DS	7.30 ± 23.3	0.95 ± 11.0	6.18 ± 8.7	0.76
Follow-up (day 49)				
RVD	4.21 ± 0.3	4.33 ± 0.4	4.22 ± 0.7	0.89
MLD	3.72 ± 0.5*	3.39 ± 0.6*	2.24 ± 0.5	<0.01
%DS	11.5 ± 11.0*	21.9 ± 11.1*	46.5 ± 10.9	<0.01
Late loss	−0.41 ± 0.9*	0.35 ± 0.6*	1.22 ± 0.6	<0.01

Values are mean ± SD. *p < 0.05 versus plain balloon angioplasty.
BAR = balloon-to-artery ratio; %DS = percentage of diameter stenosis; MLD = minimal lumen diameter; PCB = paclitaxel-coated balloon; PBA = plain balloon angioplasty; RVD = reference vessel diameter.

utility of this model in the evaluation of efficacy of drug-delivery devices in the novo (14) and ISR (15) settings. These studies have shown that the combination of the intrinsic metabolic defect of the FHS and vessel wall injury is sufficient to demonstrate differences in efficacy after the use of different PCB technologies compared with PBA (17).

The tissue uptake study revealed that the second-generation PCB had slightly higher paclitaxel concentration and a more uniform segment-to-segment distribution along the artery compared with the first-generation PCB. In the vascular healing response study, the efficacy of paclitaxel in reducing neointimal proliferation was comparable in both PCB groups and significantly reduced compared with PBA controls. However, the variance of %AS along the length of stented segment was higher in the first-generation PCB compared with the second-generation PCB (within a 30% to 60% range of %AS) (Fig. 4). Our results support the notion that more uniform and reproducible balloon coating results in more homogeneous drug distribution along the treated site. This is an important finding because the variation in tissue concentration of paclitaxel appears to influence the overall healing response after PCB treatment. In addition, second-generation PCB resulted in a more favorable biological response, showing lower fibrin deposition and neointimal and medial smooth muscle cell loss and the presence of more mature neointimal layers. Our data also support previous studies reporting the potential deleterious vascular effects of paclitaxel delivered outside of its therapeutic window (15).

Table 2. Histomorphometric Analysis of All Stents Explanted at Terminal Follow-up

	Second-Generation PCB (n = 6)	First-Generation PCB (n = 6)	PBA (n = 6)	p Value
EEL area, mm^2	27.8 (25.0–30.0)*	22.7 (21.0–25.0)	24.3 (21.0–28.0)	0.04
IEL area, mm^2	25.0 ± 1.7*†	19.6 ± 1.6	18.9 ± 1.6	0.02
Lumen area, mm^2	12.4 ± 1.9*†	8.8 ± 3.7†	4.0 ± 1.4	<0.01
Medial area, mm^2	2.4 (1.7–2.7)	3.36 (2.5–3.9)	4.79 (2.0–8.0)	0.37
Neointimal area, mm^2	12.8 ± 2.2	10.7 ± 3.5	14.9 ± 2.8	0.07
Neointimal thickness, μm	854.4 ± 161.0†	864.3 ± 394.0†	1,354.8 ± 284.0	0.01
Area of stenosis, %	50.5 ± 7.0%†	54.8 ± 18.0%†	78.2 ± 10.0%	0.01

Values are mean (range) or mean ± SD. *p < 0.05 vs. first-generation PCB. †p < 0.05 vs. PBA.
EEL = external elastic lamina; IEL = internal elastic lamina; other abbreviations as in Table 1.

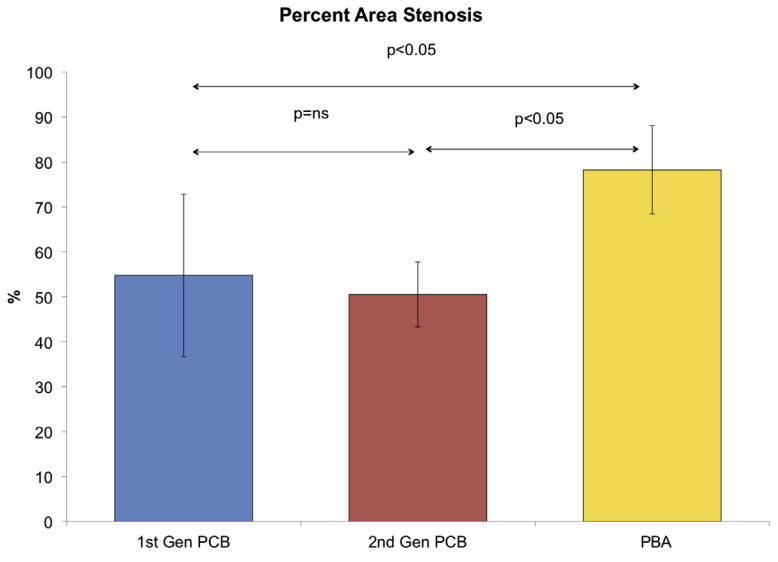

Figure 3. Percentage of Area Stenosis on Histological Analysis

The efficacy of paclitaxel in reducing neointimal proliferation was comparable in both PCB groups and significantly reduced compared with PBA controls. Abbreviations as in Figure 1.

To date, only 1 study directly compared the vascular effects of 2 different PCBs (26). This study, however, differed significantly with regard to the excipient and balloon technology used (iopromide vs. roughened surface). Although new PCB developments are under investigation (27,28), the influence on vascular response and pharmacokinetics has not been compared with those of the original Paccocath formulation. Therefore, to the best of our knowledge, our study is the first to provide a head-to-head comparison of first- and second-generation PCB technologies. In addition, because both PCBs used in this study shared identical coating formulations and drug concentrations, the importance of the coating methods and techniques was distinguished and validated by our findings.

The clinical implications of these findings are important. First, as PCB technologies evolve and the tissue delivery and distribution of paclitaxel become more consistent, the introduction of this technology into clinical practice becomes more appealing. In addition, because these new-generation coatings appear to induce lower degrees of delayed healing, there is a potential to expand the use of this technology to a broader range of applications (6,8). This is particularly important in the setting

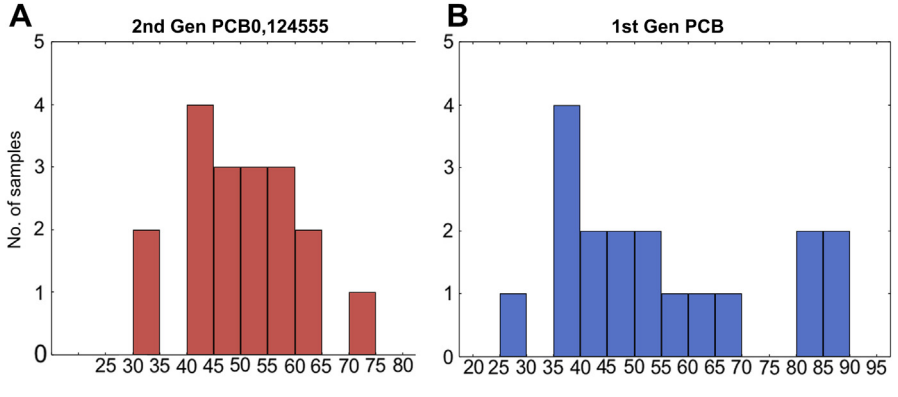

Figure 4. Histograms Representing Neointimal Distribution According to the Percentage of Area Stenosis in Stented Cross Sections

Normal and within a range of 30% to 60% area stenosis, neointimal distribution was observed in second- **(A)** and skewed in first-generation PCBs **(B)** (Shapiro-Wilk, $p = 0.03$ and $p = 0.59$).

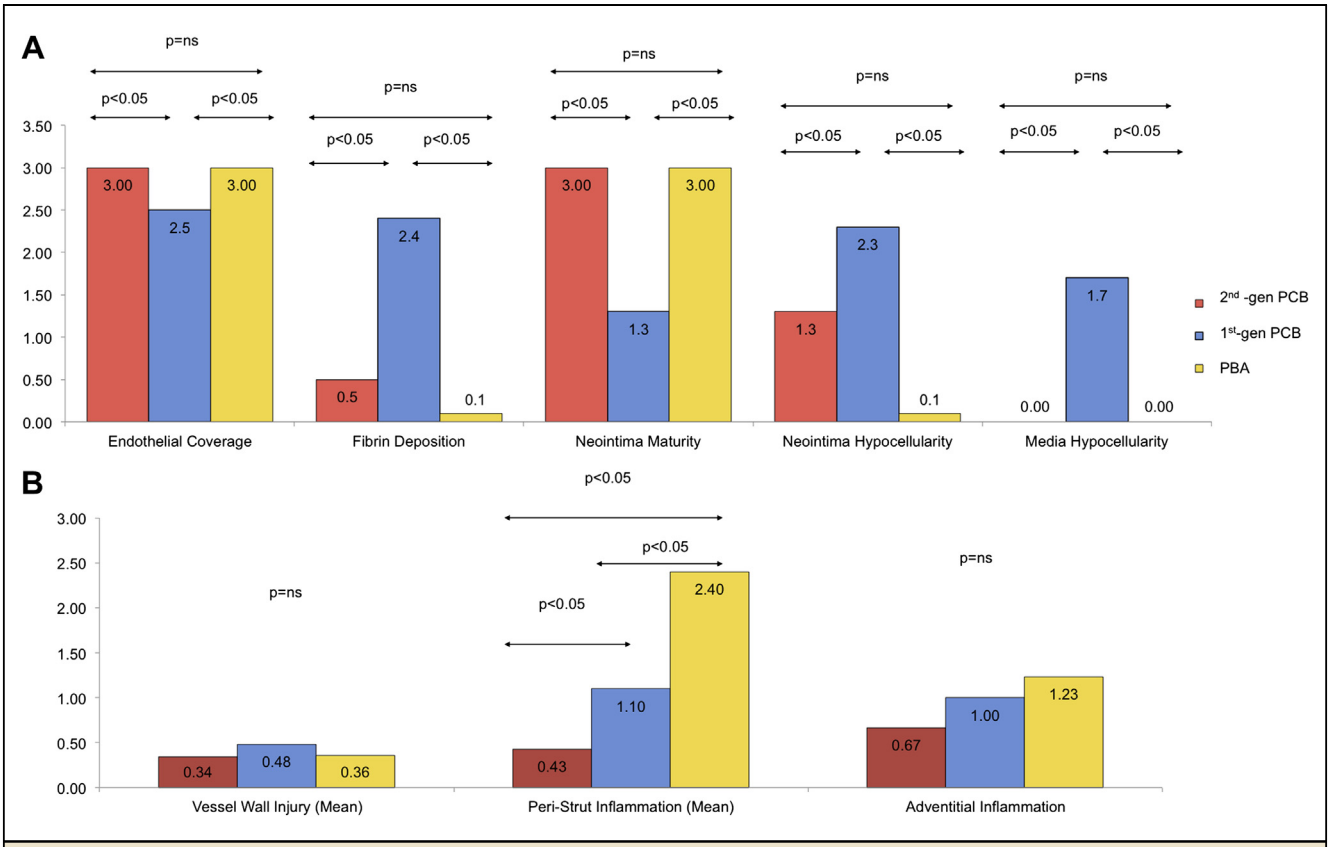

Figure 5. Healing and Biocompatibility Profile

Qualitative histopathological analysis representing healing **(A)** and biocompatibility **(B)**. Second-generation paclitaxel-coated balloon resulted in a more favorable biological response showing lower fibrin deposition and neointimal and medial smooth muscle cell loss and the presence of mature neointimal layers. Abbreviations as in Figure 1.

of adjunctive bare-metal stent use in which the synergistic use of PCB in the coronary territory remains controversial (8,29).

Study limitations. First, although a disease animal model was used, the lack of an atherosclerotic plaque component neglects the impact of tissue characteristics on drug uptake and retention. Second, although long-term paclitaxel retention was not tested in this study, the retention up to 6 months has already been reported for both PCB groups (30).

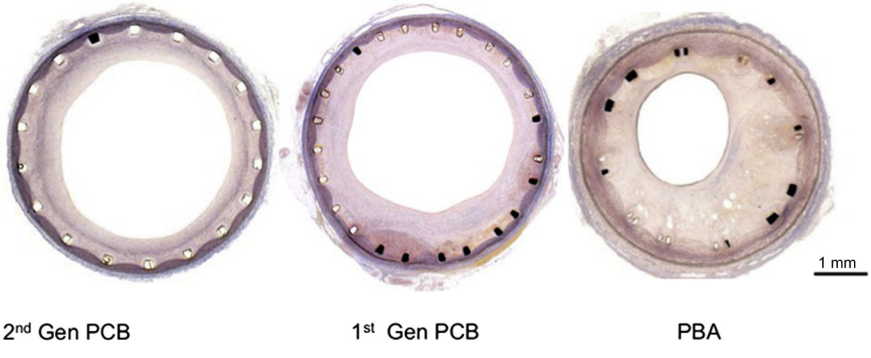

Figure 6. Representations of Vessel Sections of All Groups

Abbreviations as in Figure 1.

Conclusions

In the experimental setting, second-generation PCB technologies displayed a similar degree of efficacy compared with the original first-generation PCB formulation. However, paclitaxel delivery was more uniform throughout the vessel length and resulted in more favorable vascular tissue profile. The clinical implications of these findings deserve further investigation in human clinical trials.

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REFERENCES

1. Scheller B, Hehrlein C, Bocksch W, et al. Treatment of coronary in-stent restenosis with a paclitaxel-coated balloon catheter. N Engl J Med 2006;355:2113–24.
2. Unverdorben M, Vallbracht C, Cremers B, et al. Paclitaxel-coated balloon catheter versus paclitaxel-coated stent for the treatment of coronary in-stent restenosis. Circulation 2009;119:2986–94.
3. Tepe G, Zeller T, Albrecht T, et al. Local delivery of paclitaxel to inhibit restenosis during angioplasty of the leg. N Engl J Med 2008;358:689–99.
4. Werk M, Langner S, Reinkensmeier B, et al. Inhibition of restenosis in femoropopliteal arteries: paclitaxel-coated versus uncoated balloon: femoral paclitaxel randomized pilot trial. Circulation 2008;118:1358–65.
5. Stella PR, Belkacemi A, Dubois C, et al. A multicenter randomized comparison of drug-eluting balloon plus bare-metal stent versus bare-metal stent versus drug-eluting stent in bifurcation lesions treated with a single-stenting technique: six-month angiographic and 12-month clinical results of the drug-eluting balloon in bifurcations trial. Catheter Cardiovasc Interv 2012;80:1138–46.
6. Cortese B, Micheli A, Picchi A, et al. Paclitaxel-coated balloon versus drug-eluting stent during PCI of small coronary vessels, a prospective randomised clinical trial. The PICCOLETO study. Heart 2010;96:1291–6.
7. Wohrle J, Zadura M, Mobius-Winkler S, et al. SeQuentPlease World Wide Registry: clinical results of SeQuent please paclitaxel-coated balloon angioplasty in a large-scale, prospective registry study. J Am Coll Cardiol 2012;60:1733–8.
8. Hamm C. Paclitaxel-eluting PTCA-balloon in combination with the Cloroflex Blue stent vs. the sirolimus coated Cypher stent in the treatment of advanced coronary artery disease. American Heart Association Scientific Sessions 2009; November 14, 2009; Orlando, FL, 2009.
9. Latib A, Colombo A, Castriota F, et al. A randomized multicenter study comparing a paclitaxel drug-eluting balloon with a paclitaxel-eluting stent in small coronary vessels: the BELLO (Balloon Elution and Late Loss Optimization) study. Journal of the American College of Cardiology 2012;60:2473–80.
10. Hasler-Rapacz J, Ellegren H, Fridolfsson AK, et al. Identification of a mutation in the low density lipoprotein receptor gene associated with recessive familial hypercholesterolemia in swine. Am J Med Genet 1998;76:379–86.
11. Prescott MF, McBride CH, Hasler-Rapacz J, Von Linden J, Rapacz J. Development of complex atherosclerotic lesions in pigs with inherited hyper-LDL cholesterolemia bearing mutant alleles for apolipoprotein B. Am J Pathol 1991;139:139–47.
12. Granada JF, Kaluza GL, Wilensky RL, Biedermann BC, Schwartz RS, Falk E. Porcine models of coronary atherosclerosis and vulnerable plaque for imaging and interventional research. EuroIntervention 2009;5:140–8.
13. Tellez A, Krueger CG, Seifert P, et al. Coronary bare metal stent implantation in homozygous LDL receptor deficient swine induces a neointimal formation pattern similar to humans. Atherosclerosis 2010;213:518–24.
14. Granada JF, Milewski K, Zhao H, et al. Vascular response to zotarolimus-coated balloons in injured superficial femoral arteries of the familial hypercholesterolemic Swine. Circ Cardiovasc Interv 2011;4:447–55.
15. Milewski K, Zurakowski A, Pajak J, et al. Effects of local intracoronary paclitaxel delivery using the Remedy transport catheter on neointimal hyperplasia after stent implantation in a porcine model. Cardiovasc Revasc Med 2011;12:82–9.
16. Milewski K, Afari ME, Tellez A, et al. Evaluation of efficacy and dose response of different paclitaxel-coated balloon formulations in a novel swine model of iliofemoral in-stent restenosis. J Am Coll Cardiol Intv 2012;5:1081–8.
17. Schwartz RS, Huber KC, Murphy JG, et al. Restenosis and the proportional neointimal response to coronary artery injury: results in a porcine model. J Am Coll Cardiol 1992;19:267–74.
18. Kornowski R, Hong MK, Tio FO, Bramwell O, Wu H, Leon MB. In-stent restenosis: contributions of inflammatory responses and arterial injury to neointimal hyperplasia. J Am Coll Cardiol 1998;31:224–30.
19. Gray WA, Granada JF. Drug-coated balloons for the prevention of vascular restenosis. Circulation 2010;121:2672–80.
20. Posa A, Hemetsberger R, Petnehazy O, et al. Attainment of local drug delivery with paclitaxel-eluting balloon in porcine coronary arteries. Coron Artery Dis 2008;19:243–7.
21. Schnorr B, Kelsch B, Cremers B, Clever YP, Speck U, Scheller B. Paclitaxel-coated balloons–survey of preclinical data. Minerva Cardioangiol 2010;58:567–82.
22. Cremers B, Toner JL, Schwartz LB, et al. Inhibition of neointimal hyperplasia with a novel zotarolimus coated balloon catheter. Clin Res Cardiol 2012;101:469–76.
23. Schwartz RS, Edelman E, Virmani R, et al. Drug-eluting stents in preclinical studies: updated consensus recommendations for preclinical evaluation. Circulation Cardiovascular interventions 2008;1:143–53.
24. Scheller B, Speck U, Abramjuk C, Bernhardt U, Bohm M, Nickenig G. Paclitaxel balloon coating, a novel method for prevention and therapy of restenosis. Circulation 2004;110:810–4.
25. Miller KM. A drug-eluting stent case study: TAXUS Express2 – from development to approval. New England Chapter Parental Drug Association Workshop, 2004.
26. Cremers B, Biedermann M, Mahnkopf D, Bohm M, Scheller B. Comparison of two different paclitaxel-coated balloon catheters in the porcine coronary restenosis model. Clin Res Cardiol 2009;98:325–30.
27. Posa A, Nyolczas N, Hemetsberger R, et al. Optimization of drug-eluting balloon use for safety and efficacy: evaluation of the 2nd generation paclitaxel-eluting DIOR-balloon in porcine coronary arteries. Catheter Cardiovasc Interv 2010;76:395–403.
28. Stella PR, Belkacemi A, Waksman R, et al. The Valentines Trial: results of the first one week worldwide multicentre enrolment trial, evaluating the real world usage of the second generation DIOR paclitaxel drug-eluting balloon for in-stent restenosis treatment. EuroIntervention 2011;7:705–10.
29. Cortese B, Sgueglia GA, Granada JF. Paclitaxel coated balloons, the time for awareness has come. Int J Cardiol 2013;20:1–2.
30. Speck U, Cremers B, Kelsch B, et al. Do pharmacokinetics explain persistent restenosis inhibition by a single dose of paclitaxel? Circ Cardiovasc Interv 2012;5:392–400.

Key Words: in-stent restenosis model ■ paclitaxel-coated balloon ■ second-generation.

APPENDIX

For supplemental material, please see the online version of this article.