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Review

Biological aspects of radiation and drug-eluting stents for the prevention of restenosis

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Abstract

Based on recent advances, this article aims to review the biological basis for the use of either radiation or drug-eluting stents for the prevention of restenosis, and to elucidate the complementary role that they may play in the future. Vascular restenosis is a multifactorial process primarily driven by the remodeling of the arterial wall, as well as by the hyperproliferation of smooth muscle cells (SMC). These pathophysiological features are the target of therapeutic strategies aimed at inhibiting constrictive remodeling as well as inhibiting SMC proliferation. The success of radiation as well as anti-proliferative drugs such as paclitaxel and sirolimus lies in the primary and/or multifactorial inhibition of cell proliferation. Radiation has the additional feature of preventing constrictive remodeling while sirolimus has the potential property of being anti-inflammatory, which may be a desirable feature. The effects of radiation are not reliant on any uptake and "metabolism" by the target cells, as in the case with drugs, and thus radiation potentially may be more effective as a result of its more-direct action. However, radiation does have some significant drawbacks compared to drug-eluting stents, including a much delayed re-endothelialization resulting in the need for prolonged anti-platelet therapy. Based on recent clinical data, drug-eluting stents have been shown to markedly reduce the likelihood of restenosis, which actually favors this approach for the prevention of restenosis. From a biological perspective, drug-eluting stents and radiation have certain differences, which are reviewed in this article.

Keywords: Restenosis; Stents; Angioplasty; Smooth muscle; Remodeling

1. Introduction

More than 1.5 million percutaneous interventions are performed worldwide each year for coronary disease [1]. The recurrence of obstructive lesions, or restenosis, has been the major complication of percutaneous transluminal coronary angioplasty (PTCA) since its introduction by Gruentzig in the mid 1970s [2–6]. This process has been considered to occur as a result of three separate mechanisms: (1) elastic vascular recoil, (2) neointimal hyperplasia and matrix deposition and (3) constrictive remodeling.

The rationale for delivering ionizing radiation to the sites of coronary angioplasty and stenting, to prevent restenosis, emerged from the understanding that neointimal hyperplasia represented a proliferative response to PTCA and stenting. Radiation represented a potentially effective means of dealing with that response [7–10]. Trials of intra-luminal irradiation, either using a radioisotopic stent or intra-luminal brachytherapy, have revealed impressive results, with up to fourfold decreases in restenosis reported [11]. Several studies

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have been performed to test the efficacy of gamma-radiation emitters (SCRIPPS [12] and GAMMA 1 [13]) and beta-emitters (Beta-wrist [14] and START [15]) for the treatment of in-stent restenosis. All of these have shown positive benefits from the use of radiation. The situation is different for the treatment of newly diagnosed stenosis with radioactive stents or intra-luminal brachytherapy. The studies have either revealed aneurysmatic alterations of vessels [16], edge effects (stenosis or restenosis at the proximal and/or distal end of an irradiated segment) [17,18], or simply have failed to show any prevention of restenosis [19,20]. It appears that intra-luminal irradiation is a promising tool for the treatment of in-stent restenosis, while the irradiation of newly diagnosed stenosis fails to show a positive benefit. However, there are two important complications related to the use of brachytherapy—an edge restenosis (candy-wrapper effect) and the risk of late thrombosis. Edge restenosis was first noted with the use of radioactive stents, and it is considered to be the result of the fall-off in the radiation dose at the edges (ends) of the stent. It has been proposed that this may exert a proliferative stimulus—as described in vitro [21]—on the smooth muscle cells (SMC) of the vessel wall resulting in a neointima at the site of the stent edges after these lower doses of irradiation.

Besides the advantage of successfully preventing the recoil mechanism, the implantation of a stent offers the opportunity to use it as a vehicle for local drug delivery. Several compounds such as sirolimus, tacrolimus, everolimus, paclitaxel, QuaDS-QP-2, actinomycin D, heparin and dexamethasone have been tested for stent coating, primarily with the aim of the inhibition of SMC proliferation. Among these compounds most experience has been gathered for sirolimus and paclitaxel, and additional studies for related compounds are published or in preparation. The ‘‘First in man’’ experience of sirolimus-coated stents was reported by Sousa et al. [22,23] from Brazil and most recently this group reported the lack of restenosis in a 2-year follow up after implantation of sirolimus-coated stents in human coronary arteries with newly diagnosed stenosis. Subsequent multicenter randomized trials (e.g. the RAVEL trial) essentially confirmed the results of the initial feasibility study with again overwhelming success in the prevention of restenosis after implantation of a sirolimus-coated stent [24,25]. These studies have been criticized for the favourable patient population enrolled. Subsequent multicenter trials, the SIRIUS trial and E-SIRIUS trial, were recently reported showing a clear benefit for the treatment of complex coronary lesions/single-long-atherosclerotic coronary lesions by sirolimus-eluting stents. Some subgroups had less favourable outcomes but even in these patients a profound benefit in comparison to controls was observed [26,27]. Comparably promising results have been obtained from the use of paclitaxel-coated stents, an antiproliferative compound which has also been shown to be efficacious for the treatment of in-stent restenosis (ASPECT, Taxus I–III) [28–31].

Apart from the clinical benefits of most types of drug-eluting stents in the prevention of restenosis at least for newly diagnosed lesions, the economic value of stenting versus brachytherapy appears to be rather unpredictable at the present stage. While the costs of drug-eluting stents are determined by the stent on the one hand versus the decrease in re-interventions [32,33], the use of brachytherapy will at least require additional devices and the availability of a radiation therapist. These costs have to be viewed in face of the decrease in the number of re-interventions. The judgement of the economic aspects is currently left to additional studies. Nevertheless, given the recently reported efficacy of most drug-eluting stents for the prevention of restenosis, it seems timely to review the biological basis of intraluminal irradiation and drug-coated stents and try to elucidate the complementary role that they may play in the future.

2. Radiation—intravascular brachytherapy

Numerous studies have consistently demonstrated remarkable suppression of neointima formation using radiation from a variety of isotopes delivered by an endoluminal approach. At least three groups have documented similar results in the pig coronary artery model of restenosis after overstretch balloon injury, using the γ-emitter ^{192}Ir at roughly comparable doses. Wiedermann et al. [34] found suppression of neointimal formation 4 weeks after angioplasty when 20 Gy was given at a radial depth of 1.5 mm just before arterial injury. Similarly, the group at Emory demonstrated a marked suppression of neointima formation using ^{192}Ir with a dose–response effect in vessels irradiated with 3.5, 7, and 14 Gy at a radial depth of 2 mm. A continued benefit was seen at 6 months in arteries irradiated with 14 Gy [35,36]. One important difference between the results reported from the Columbia group and the Emory group is the effect of radiation dose on outcome, i.e. the dose–response relationship. The Columbia group described beneficial effects of 15 and 20 Gy, given at 1.5 mm from the center of the source, but the results with 10 Gy showed greater neotintimal proliferation than in controls. In contrast, the Emory group saw a beneficial effect even with the lowest dose of 3.5 Gy (at 2 mm). Further studies by other authors have shown greater neointimal proliferation at low doses (e.g. 5 Gy) compared to controls, suggesting that balloon or stent injury combined with such low doses is a poor combination. The partly contradictory results of the radiation studies mentioned above may be based on two general problems with such studies, the comparability of animal models and the influence of radiation dose, dose fractionation and penetration depth of the radiation. However, although additional studies are required to resolve these questions, the message taken from studies available so far is that radiation at a certain minimal dose is capable of preventing neointimal proliferation.

Because of concerns about prolonged treatment times and radiation safety problems associated with penetrating gam-

ma-ray emitters like ^{192}Ir, a number of investigators have studied the potential use of β-emitting isotopes (^{90}Y, ^{90}Sr/Y, ^{188}Re and ^{186}Re) in the prevention of restenosis. Verin et al. [37] reported the use of a flexible ^{90}Y coil deployed at the end of a guidewire using a balloon catheter centering device, after balloon injury in the carotid and iliac arteries of hypercholesterolemic rabbits. They demonstrated a reduction in BrdU-positive cells in the intima and media of arteries receiving 6, 12 or 18 Gy compared to controls; however at 6 weeks only the 18 Gy dose was effective in reducing neointima formation. The Emory group examined the use of stainless-steel encapsulated seeds containing ^{90}Sr/Y [38]. The results of this study indicated that after doses of 7 and 14 Gy (again at 2 mm depth), healing at 2-weeks post-angioplasty in the coronary artery was similar to that observed using ^{192}Ir. Again, the comparability of the results of radiation studies is hampered by the use of different models (species, hypercholestrolemia) and/or different radionuclides that actually reflects the complex problem of restenosis and the multiple mechanisms to be targeted. Further studies are required to evaluate the effectiveness of radiation approaches especially regarding the radiation penetration depth—because this determines the vessel wall layers to be treated—and the risk factors for arteriosclerosis and restenosis.

However, scanning electron microscopy and Indium-labelled platelet studies have shown incomplete healing of vessels with doses of $\geq$ 15 Gy at 1 and 3 months follow-up [39]. Inadequate endothelial recovery of an irradiated artery after angioplasty renders its luminal surface prothrombotic, and in the setting of an appropriate physiologic stimulus, results in thrombotic occlusion. The problem of late thrombosis observed in clinical trials certainly is compatible with the delayed healing observed in the animal studies.

Myointimal proliferation leading to variable degrees of vessel occlusion can also represent a late consequence of therapeutic radiation exposure even in the absence of balloon injury. The effect may occur many years after exposure [40]. A chance observation in a study designed to investigate the time sequence of changes in the spinal cord of pigs after irradiation has, however, provided some insight into possible mechanisms. Irradiation was carried out with a single dose of 27.5 Gy of ^{60}Co gamma-rays to a 10 cm length of the cervical spinal cord in 4-month-old female large white pigs [41]. The observed major vessel changes were in the main ventral artery within the lining of the spinal cord. At the earliest time point, 6 weeks after irradiation, there was qualitative evidence for a reduction in the number of endothelial cells lining the wall of this blood vessel. This observation is consistent with findings for the microcirculation in simple model systems [42], where a decline in endothelial cell number has been reported in the first 4–8 months after irradiation. From 10 to 12 weeks after exposure there had been the development of a clearly defined sub-endothelial space, this contained hyaline material (Fig. 1A) and the infiltration of mononuclear cells from the blood. White blood cell adherence to the endothelium and an excess number of mononuclear cells in the lumen of the vessel were also noted indicating chemo-attraction into the irradiated area (Fig. 1B). By 12–14 weeks after irradiation the lumen of the vessel was reduced to a varying degree and the sub-endothelial space was filled by what appeared to be loose connective tissue (Fig. 1C). At slightly later times (16–18 weeks) the sub-endothelial space was filled with dense tissue resulting in a varying degree of vessel occlusion (Fig. 1D). The internal

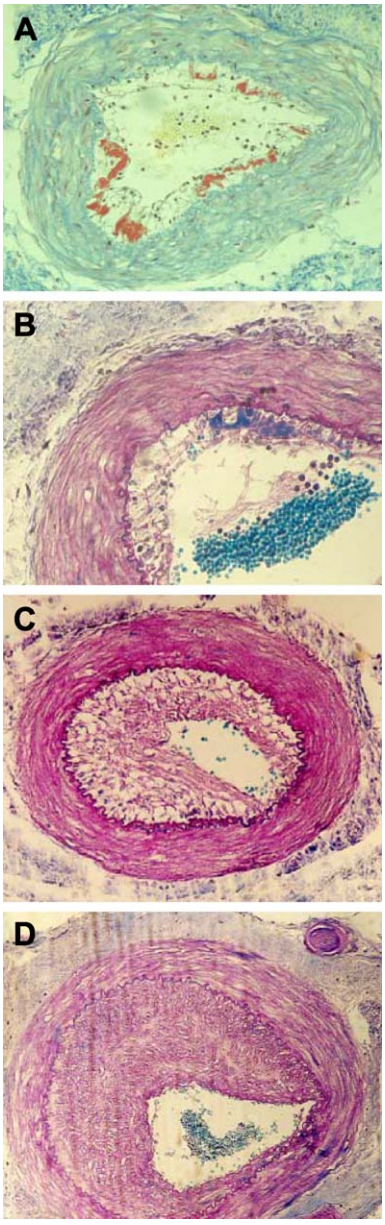

Fig. 1. Histological appearance of the inferior spinal artery of the spinal cord of pigs after the local irradiation of the cervical spinal cord with a single dose of 27.5 Gy of Co-60 gamma-rays: panel A: 10 weeks, shows white blood cells accumulating in the lumen of the vessel and in the subendothelial space. The subendothelial space also shows an accumulation of hyline material (MSB stain), panel B: higher powered view of an adjacent section to show the subendothelial changes (Luxol fast blue, PAS), panel C: 14 weeks, shows almost complete occlusion of the vessel lumen with loose connective tissue (Luxol fast blue, PAS) and panel D: 16 weeks, almost complete occlusion of the lumen of the blood vessel with dense connective tissue, myointimal proliferation (Luxol fast blue, PAS).

elastic lamina remained intact. A vasculitis in small vessels adjacent to the locally irradiated vessel recently has been reported [43]. This vasculitis was apparently unrelated to dose. An inflammatory reaction was a consistent finding and the possible role of the up-regulation of cytokines was discussed. Comparable changes have also been reported in microcirculation networks; for example, in the renal glomeruli of the pig after irradiation [44].

Cellular radiation responses involve both DNA-damage-dependent and DNA-damage-independent signalling pathways. As outlined in Fig. 2, DNA-damage, mainly DNA-double strand breaks, is able to activate the protein kinases ATM and DNA-PK, both located in the cell nucleus. ATM and DNA-PK will then phosphorylate p53 in specific serine residues, which results in the stabilisation and activation of the tumor suppressor protein p53 [45]. Once p53 is activated, it acts as a transcription factor and induces the expression of the p21-protein, a product of the WAF/CIP1 gene [46,47]. This protein accumulates in the cell nucleus and inactivates the activity of cyclin dependent kinase (cdk2)/cyclin E complexes. This leads to a block in cell cycle progression and arrests cells in G1-phase to allow time for the repair of DNA-damage. Cells with accurately repaired DNA can re-enter the cell cycle and progress through S-phase into mitosis. Thus, DNA-damage repair is necessary for cell survival and undisturbed proliferation following radiation exposure. When DNA double-strand breaks are incorrectly repaired during G1-arrest then the cell might encounter severe problems in distributing DNA/chromosomes in the following mitosis that may result in mitotic/reproductive cell death. The latent period for expression of this injury can be very variable, ranging from short times post-irradiation in rapidly dividing cells to very long times in near-quiescent cell populations in vivo.

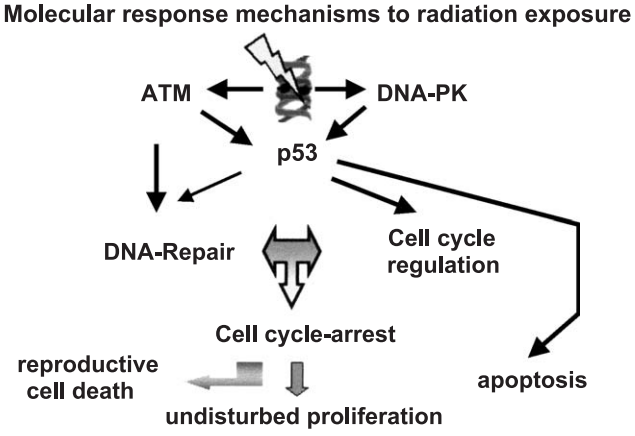

Fig. 2. Radiation-induced DNA-damage results in the induction of a variety of molecular signals leading to damage recognition, cell cycle arrest and DNA repair. The interaction of this network of molecular events determines the cellular fate, i.e. cell death or cell survival. DNA double-strand breaks mainly are responsible for the activation of specific proteins, which are involved in damage sensing like the ATM protein, or in DNA repair like DNA-Protein Kinase (DNA-PK). Both proteins, however, activate/stabilize the tumor suppressor protein p53, which is a major control protein in regulating cell cycle arrest and transactivation of DNA repair-relevant genes as well as genes controlling apoptotic pathways. As a result of successful DNA-repair irradiated cells will survive the radiation insult and re-enter cell cycle for undisturbed proliferation. In the case of non-successful DNA-repair, cells can either enter directly the apoptotic pathway mainly under the control of p53 or go through a limited number of cell cycles, which will result in the so-called mitotic or reproductive cell death due to severe problems in distributing the incorrect-repaired DNA/chromosomes during mitosis.

Recently, it has been shown that the upregulation of the tumor suppressor gene PTEN inhibits the proliferation, migration and survival of vascular SMC by blocking the AKT and FAK-dependent signalling cascades [48]. Expression of PTEN has been shown to be upregulated by radiation through a mechanism involving the activation of the early growth response-1 gene, Egr-1 [49]. Egr-1 activity can be increased several fold by radiation doses greater than 4 Gy in a variety of cell types [50,51]. Therefore, it seems very likely that DNA-damage-independent induction of PTEN may contribute to the inhibitory effect of radiation on vascular SMC proliferation and migration.

A substantial literature has accumulated on the effects of ionizing radiation on the cellular components of blood vessels. For endothelium, a variety of molecular phenomena have been examined ranging from the function of lipoprotein receptors to adhesion molecule expression. For the most part it can be stated that endothelium is much more sensitive to the late effects of irradiation than are the other cellular residents of the arterial wall. The importance of the radiosensitivity of arterial endothelium in a setting of acute angioplasty is probably negligible insofar as the actual site is concerned, since perhaps >90% of the endothelium is denuded by the procedure. However, endothelium from branch arteries or in the vasa vasorum could play a significant role in postangioplasty healing and might easily be influenced by endovascular irradiation particularly from a penetrating gamma-emitter or external irradiation.

3. Drug-eluting stents

3.1. Drug-dose responses in vitro

The influence of various kinds of compounds for the inhibition of SMC proliferation to prevent restenosis has been tested. HMG-CoA reductase inhibitors have been studied extensively both in vitro [52,53] as well as clinically [54–57]. Other compounds studied include corticosteroids [58], calcium antagonists [59] or angiotensin converting enzyme inhibitors [60,61]. A general problem using the systemic application of single compounds for antiproliferative purposes is that concentrations needed in vitro for the required effects cannot be easily achieved in vivo due to critical side effects. This has led to the current situation in that no compound tested so far has made a major contribution to the prevention of restenosis [62].

Sirolimus has been studied for its immunosuppressant and antiproliferative properties on a variety of cell lines over several years. This compound mimics a starvation-like signal as distinct from amino acid and glucose deprivation [63]. It acts during both co-stimulatory activation and cytokine-driven pathways via a unique mechanism: inhibition of a multifunctional serine–threonine kinase, the mammalian target of rapamycin (=sirolimus) (mTOR) [64]. Sirolimus forms a complex with FKBP12, which inhibits mTOR activation resulting in sustained p27 kip1 levels and inhibition of pRb phosphorylation as well as impaired DNA synthesis by inhibition of p70S6K kinase [65–69]. Regarding vascular biology, special interest is linked to the proliferation of SMC and fibroblasts. Primary fibroblast cultures have been exposed to sirolimus at concentrations of 0.1–100 ng/ml with or without platelet derived growth factor (PDGF) or basic fibroblast growth factor (bFGF). Interestingly, none of the concentrations of sirolimus used induced cytotoxic reactions, but an inhibition of PDGF—as well as bFGF-induced proliferation was observed after all doses of the compound used. For PDGF the most marked effect (60% inhibition of proliferation) was observed with 30 ng/ml while for bFGF the effects were less pronounced (37% inhibition) and was optimal at 10 ng/ml [70]. A similar inhibition of growth factor effects was found for sirolimus (10 ng/ml) using rat hepatocytes [71]. For vascular SMC, concentrations as low as 1 ng/ml have been shown to exert antiproliferative effects in vitro [65]. The inhibition of growth factor effects was much greater than that of other immunosuppressants such as cyclosporin A or FK506 [72], which exemplifies the special role of sirolimus for inhibition of cell proliferation. This is presumably caused by its mechanism of inhibiting mTOR. Sun et.al. [73] have shown that the effect of sirolimus in vitro is not only restricted to SMC proliferation but sirolimus also inhibits the bFGF-driven migration of SMC. This effect is dependent on p27. This again indicates the involvement of the mTOR pathway. The inhibition of cell migration has been studied on both human and rat SMC, and it was found to be significantly reduced by sirolimus at doses of 2 ng/ml [66].

The other compound of pivotal interest for stent coating is paclitaxel, which has been shown to exert antiproliferative effects on SMC at concentrations several orders of magnitude lower than those used for the treatment of cancer. The compound acts by inducing cellular microtubules to form stable chains with resulting G0/G1 and G2/M arrest [74]. Concentrations higher than 10 nmol/l show antiproliferative effects on bovine coronary SMC [75], while studies on human arterial SMC showed a 50% reduction in cell proliferation at concentrations as low as 2 nmol/l in both monocultures and cocultures with human arterial endothelial cells [76]. Remarkably, the antiproliferative effect can be considerably enhanced in a supra-additive fashion by the combined use of paclitaxel with cyclosporine A, via the activation of the protein kinase C pathway [75]. Also, paclitaxel is a chemotherapeutic compound clearly capable of inducing apoptosis. Studies on DNA fragmentation revealed that the dose–responses for the induction of apoptosis are at least partially related to the antiproliferative effects [75], which is not the case for the dose responses using sirolimus.

3.2. Drug-dose response and latencies in vivo

Besides the advantage of successfully preventing the recoil mechanism, the implantation of a stent offers the opportunity to use it as a vehicle for drug delivery. To allow a controlled drug release, a variety of biomaterials/polymers such as copoly(ester-amide)elastomers [77] and phosphorylcholine have been tested especially with view to drug release fromulas, a reduction in platelet and protein adhesion, and the capability of an endothelialization of the stent [78–81]. Progress with such polymers has founded the era of drug-coated stents.

The use of sirolimus-coated stents or stents coated with related drugs shows promise for eliminating the problem of in-stent restenosis. Results obtained from animal models as well as clinical studies have been so overwhelming that leading cardiologists in the field talk about a ‘‘turning point in cardiology’’ [82]. In the meantime several studies have shown the consistent finding of largely impaired neointima formation in both animal models [83,84] and in patients for a variety of arteries, such as femoral and coronary arteries [22,23,85,86]. A drug load of 185 μg sirolimus per stent successfully reduced neointima formation in a porcine injury model [84]. From the data available so far, sirolimus-coated stents may abrogate the problem of in-stent restenosis and may challenge the clinical outcome of coronary-artery bypass grafting under certain conditions. There are developments such as a sirolimus-coated stent that utilizes a non-erodable methacrylate copolymer matrix with a 30% drug-to-polymer ratio, by weight. A thin coating of 5–10 μm is applied to a Bx Velocity stent (Cordis, Johnson & Johnson) with a total quantity of 185 μg (140 μg/cm^2). This is delivered in a slow release formula over a period of 4 weeks. Despite local delivery of the drug, peak systemic concentrations occur 1 h after implantation of the sirolimus-coated stent. This amount is about 10% of those levels applied for systemic immunosuppressive purposes. The decline below the detectable level of 0.4 ng/ml occurred within 3 days [87,88].

For paclitaxel, several polymer coatings such as pLA/pLC, polymer sleeves and CSG, with drug loadings between 0.2 and 200 μg in slow and fast release formulations, have been used [88]. Paclitaxel loadings of 0.2–187 μg per stent have been tested in a porcine model of restenosis with a resulting 84% reduction in neointima formation at a drug load of 187 μg per stent [89]. Although an effective inhibition of neointimal formation has been shown by these approaches, there is also a dose-dependent cytotoxic effect, which results in impaired wound healing, persistent intimal fibrin deposition, intra-intimal haemorrhage as well as

increased intimal and adventitial inflammation as demonstrated in animal studies [90].

4. Summary and conclusions

Drug-eluting stents have been shown to markedly reduce the likelihood of restenosis when applied to relatively short lesions in previously untreated coronary vessels, and recent studies have also revealed the success of drug-eluting stents for the treatment of more complex coronary lesions and the treatment of in-stent restenosis [22–31]. Presuming that there is a sustained benefit from these devices, it is likely that widespread adoption of this technology will reduce the frequency of in-stent restenosis, the main target of intravascular brachytherapy at this time. If the application of drug-eluting stents continues to prove to be an effective and safe treatment for in-stent restenosis, then it is likely that this therapy will be adopted in lieu of intra-vascular brachytherapy in the light of simpler logistics, and vascular brachytherapy will be reserved for very limited number of niche applications. However, irradiation is still useful for peripheral vascular sites where drug-coated stents may have less utility.

Nevertheless, vascular brachytherapy has some advantages compared to drug eluting stents. Because the radiation source train may be repositioned within the vessel to treat longer lesions or lesions in multiple vessels it may have cost advantages compared with drug-eluting stents. In addition, it is likely that a radiation catheter may be able to be used at sites where it is impossible to deliver a stent, i.e. within a previously deployed stent or in some tortuous, highly angulated, small vessels or at very distal arterial locations. There may also be situations where a stent is undesirable because it may ''jail'' a side-branch or will not allow normal artery bending. The advantage of radiation in those lesions, situated at sites of arterial bifurcations or at highly flexible arterial locations such as the popliteal artery, is that the antiproliferative treatment could be delivered without placing a stent. From a mechanistic point of view, the effect of radiation is independent of its uptake and ''metabolism'' by the target cells and may be potentially more effective as a result of its more-direct action. All of the above mentioned considerations imply that radiation treatment is more flexible compared to drug-eluting stents, allowing the antirestenotic therapy to be delivered independently from the stenting, for any desired segment length and anatomic location. In addition, the radiation dose and dose distribution can be varied and adjusted as desired and may even be repeated.

Radiation does have some significant drawbacks compared to drug-eluting stents, including a much delayed re-endothelialization resulting in the need for prolonged antiplatelet therapy (Clopidogrel and Aspirin) particularly in the stented patient. On the other hand, late stent-thrombosis may also be an issue for drug-eluting stents under certain conditions. However, the potential for late radiation injury would appear to be more likely than with the drug-eluting stent.

The biology of radiation effects is different from the effects produced by sirolimus or paclitaxel. Although radiation has the advantage of inducing positive or adaptive remodeling [91], this feature may be less important regarding in-stent restenosis, since in this situation an outward remodeling of the vessel wall is limited by the stent itself or may even result in incomplete apposition. The typical effect of radiation is DNA damage in various cell types including endothelial cells, smooth muscle cells and fibroblasts, depending on the radiation penetration depth into the vessel wall.

From the biological standpoint, the question as to which is the best approach can be answered only with regard to several biological and clinical issues such as recoil, vessel remodeling, cell proliferation, apoptosis and local inflammation (see Table 1). Stenting itself has the clear advantage of resulting in an effective prevention of recoil. It is also eminently successful in the case of plaque instability and intraluminal thrombus. The success of radiation as well as antiproliferative drugs such as paclitaxel and sirolimus or related compounds lies in the inhibition of cell proliferation. Nonetheless, the success of sirolimus raises the question about its potential biological advantage over other approaches. From the data available so far it may be that the combination of antiproliferative plus potential anti-inflammatory effects allows the stent to be ''more quietly'' integrated into the vessel wall than when radiation is used. Further studies will be needed to elaborate on this hypothesis.

The mechanisms of action of brachytherapy and drug coated stents are not completely understood. In general, both radiation and the most effective drug coatings (Sirolimus, Paclitaxel) are anti-proliferative. Sirolimus has the

Table 1
Comparison of biological features of radiation and drug-coated stents

Feature	Radiation	Coated stent
Recoil	no acute effect	total prevention
Vasomotor	acute loss of vasodilatory effects	blocked
Remodeling	prevents constrictive remodeling	constrictive remodeling or incomplete apposition
Cell proliferation	G0/G1 and G2 arrest p53 → p21 (WAF1/CIP1) delay of cell cycle progression	depending on drug: *paclitaxel*: primarily G2/M (micro-tubules) but also G1 arrest
	Treatment of various vascular layers depending on radiation penetration depth	*sirolimus*: G1/S delay (p27, pRb, p70S6K kinase)
Apoptosis	not in therapeutic doses	paclitaxel: depending on dose sirolimus: probably not
Inflammation	present	paclitaxel: present sirolimus: potential inhibition

additional potential property of being anti-inflammatory, which may be an additional desirable feature. On the other hand the effect of radiation on inflammation in the vessel wall has not been evaluated. The largest deficiency that exists in the preclinical evaluation of both drug-eluting stents and vascular brachytherapy is the fact that most of these studies have been carried out in non-atherosclerotic vessels.

It seems likely that radiation will continue to find application in certain subsets of patients with in-stent restenosis. Economic considerations may promote its continued use for other indications. Given the above, continued investigation of critical clinical issues in preclinical models and long-term follow-up of irradiated patients and in patients receiving drug-eluting stents seems warranted.

Hence there remain several unanswered questions regarding the use of radiation or drugs as the preferred agent in the "antiproliferative" treatment of restenosis. Points requiring clarification include:

(1) The role of inflammatory responses and anti-inflammatory drugs in the treated and adjacent vessels.
(2) The effect of radiation or drugs on the progression of atherosclerotic changes in vessels.
(3) Studies of any late effects after drug treatments in comparison to those known after irradiation treatments.
(4) The influence of prior irradiation on the subsequent response to drug treatment.

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