The advent of drug-eluting stents (DES) has provided the medical community with a technology that is transforming the treatment of coronary artery disease. As the newest treatment modality available to the interventional cardiologist, drug-eluting stents have not only significantly reduced the risk of restenosis, but they are also allowing the interventionalists to treat more complex lesions in patients that would otherwise require more invasive bypass surgery. Development of these drug–device combination products has presented considerable challenges to the device industry because it involves a multi-disciplinary approach that combines conventional device design and manufacturing with the principles of controlled local drug delivery. This review article provides an in-depth discussion of the key elements of drug-eluting stents, focusing on the TAXUS™ paclitaxel-eluting stent as an example of this new class of product. Specific sections will review the drug and polymer matrix components, formulation development and evaluation, pre-clinical studies and clinical trial results.

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Keywords: Restenosis; Paclitaxel; SIBS; Stents; Drug-eluting stents; Controlled drug delivery; Local drug delivery; Taxus; DES

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1. Introduction

The concept of drug-eluting stents (DES), supported by a powerful demonstration of their clinical success, has brought about a paradigm shift in the field of interventional cardiology [1–5]. The DES technology revolutionized this field which had been previously dominated by bare metal stents (BMS). The primary intent behind the development of BMS was to provide mechanical scaffolding to the artery to prevent vessel recoil associated with balloon angioplasty, the predecessor of stent technology for the treatment of occlusive coronary artery disease.

With the introduction of the DES technology, it became crucial to investigate the new components applied to the standard BMS platform, i.e. the active drug and the polymer carrier. The development approach for the DES products now required a ‘combination’ perspective, one based on integrating standard device processes with new disciplines such as, pharmaceutics, pharmacology/toxicology, bioanalytics, pharmacokinetics, pharmacodynamics, and bio-pharmaceutics, all from the standpoint of device-based controlled local drug delivery. The final therapeutic outcome with this system thus became a function of the interactive effects of all elements of the DES, with each other as well as with the biological milieu, as shown in Fig. 1.

2. The DES as a controlled drug delivery system

Conventional drug therapy, administered either intravenously or via an extravascular route (oral, nasal, sublingual, or rectal), typically does not maintain drug concentrations within a desired therapeutic window at the target site for extended periods of time. The short duration of action of such therapy is due to the inability of the conventional formulation to control the temporal delivery of the drug. Additionally, the drug is present not only at the intended site but also in the systemic circulation. For conventional dosage forms, even with the best attempts to maintain plasma concentrations within the therapeutic range, drug concentrations can fall below the minimum effective levels or rise above the maximum safe levels, leading to either under-dosing or toxicity, respectively. Controlled release drug therapy circum-
vents fluctuations inherent to the conventional dosage forms by achieving a continuous release of drug within the therapeutic window for the required time of treatment. Additionally, the placement of such a system at the target location, as in the case of a drug-eluting stent implanted at the lesion site in the vasculature, also provides site-specific drug delivery.

The drug delivery scientist designing, developing, or manufacturing controlled-release products, must establish a thorough understanding of the drug and the polymer carrier. The development of a DES is no exception to this rule, due to the enormous impact of these components on the product performance attributes, namely drug release kinetics, content uniformity, mechanical integrity of the coating, product stability, and ultimately the biological outcome. The drug properties that need an in-depth investigation include, for example, understanding the solubility profile, stability, physicochemical properties, solid and solution state characteristics, drug–polymer interactions, and the therapeutic window for the intended application. The polymer carrier, as a drug delivery coating on the stent, needs to meet several criteria, which include compatibility with the drug, ability to withstand processing, sterilization and storage, adjustable formulation and drug release properties, good mechanical integrity during handling and throughout the clinical deployment procedure in the tortuous vessel anatomy, as well as through its residence in the mechanically and biologically demanding coronary artery. Last, but not the least, it needs to demonstrate vascular compatibility, i.e. have no adverse biological response beyond that of the uncoated BMS. Understanding these drug and polymer attributes is important for the successful development of a new technology and ultimately a successful product.

The subsequent sections discuss these considerations in the context of the Taxus™ paclitaxel-eluting stent system, beginning with the individual components of the system, i.e. the drug and the polymer, followed by formulation development and characterization of the drug–polymer coated stents, pre-clinical evaluation of coated stents in animal models, and finally the clinical investigation of the Taxus™ Express stent system in the management of coronary artery restenosis.

2.1. Paclitaxel

Understanding the complex process of restenosis, which has an iatrogenic etiology and is burdened with multiple pathophysiological, molecular and biochemical pathways, has been an ongoing area of research since the advent of percutaneous coronary interventions. The stent is deployed in coronary arteries at the site of an atherosclerotic lesion, the development of
which involves several highly interrelated processes, including lipid disturbances, platelet activation, endothelial dysfunction, inflammation, oxidative stress, vascular smooth muscle cell activation, altered matrix metabolism, remodeling and genetic factors, shown schematically in Fig. 2 [6].

The compromised pathophysiology of the artery makes it perhaps one of the most challenging biological environments for an interventional procedure. The vascular injury following stenting or other intervention results in a series of events that closely resemble those seen in generalized wound healing [7]. The key pathways that contribute to the formation of neointimal hyperplasia after arterial stenting have been discussed in depth [7–11]. These pathways have been broadly categorized into thrombosis, inflammation, proliferation of the smooth muscle cells (SMC) in the intima and media of the artery, migration of the medial SMC into the intima, and the secretion and organization of the extracellular matrix (ECM). The schematics of the phases of vascular repair after stent-induced arterial injury described by Garasic et al. are shown in Fig. 3 [10]. Given the biochemical, cellular, and molecular complexities involved in the cascade of restenosis, stent-based delivery of a therapeutic entity that could target one or more of the events in the cascade is probably the most attractive, albeit challenging, approach to address the problem of restenosis.

Paclitaxel was selected as the pharmacologic component for the Taxus drug-eluting stent system because of its ability to target the key events in the cascade of restenosis and its physicochemical properties, which make its systemic delivery a nightmare but are very favorable for stent-based delivery. These are discussed in this section as well as in Section 4.

Originally developed as part of a large scale effort headed by the United States National Cancer Institute to investigate chemotherapeutic agents from natural sources, paclitaxel was approved by the FDA in 1992 as an antineoplastic agent to treat metastatic ovarian cancer.
cancer after failure of first-line or subsequent chemotherapy. It is a natural diterpenoid extracted from the bark, roots, and leaves of several Taxus species, including *Taxus brevifolia* and *Taxus media*. The novel mechanism of paclitaxel as a promoter of microtubule assembly responsible for its antitumor activity was first demonstrated by Schiff et al. in 1979 [12]. Unlike other antimicrotubule drugs such as vinca alkaloids, which induce the disassembly of microtubules, paclitaxel promotes the polymerization of tubulin into stable microtubules [13,14].

Although the principal function of microtubules is the formation of the mitotic spindle during cell division, they are also involved in many vital interphase functions including the maintenance of shape, motility, signal transmission, and intracellular transport [13]. Thus, the stabilization of microtubule dynamics by paclitaxel can interrupt many cellular processes including cell division, migration, activation, maintenance of cytoskeletal framework, and intracellular as well as transmembrane protein transport. In the context of restenosis, studies conducted by various investigators have shown that paclitaxel affects development of neointimal hyperplasia in different in vitro and animal models of restenosis [15–18].

From the standpoint of a controlled stent-based delivery to a target vascular lesion, paclitaxel offers a portfolio of physicochemical properties that make it perhaps one of the most appealing agents for this application. It has a molecular formula of C_{47}H_{51}NO_{14}, molecular mass of 853.91, a tetracyclic core (baccatin III) and an ester-linked side chain attached at the carbon 13 position of baccatin III, a melting point range of 209.3 to 216.2 °C, and an octanol/water coefficient range of approximately 600–800 at various buffer and octanol concentrations [19–21]. It has limited solubility in water, with reported values ranging from 0.3 to 30 µg/ml; the wide solubility range is due to the existence of paclitaxel hydrates that have an aqueous solubility lower than the anhydrous form [22]. The chemistry of paclitaxel has been well reviewed and the molecular interactions, crystallographic data, NMR, and mass spectra have been reported in the literature [14,22–27].

### 2.2. Styrene–Isobutylene–Styrene (SIBS) triblock copolymer

A polymer-based drug delivery system was selected for the TAXUS™ DES because of various advantages that such a system offers over the neat deposition of the drug on the metal stent surface. The benefits of a polymer-based drug-eluting stent coating are outlined in Table 1.

The polymer chosen for the Taxus DES system is a soft, hydrophobic, elastomeric triblock copolymer, poly (styrene–b-isobutylene–b-styrene) (SIBS, commercially known as Translute™). The Taxus DES system is the first commercial application of this polymer in a drug delivery platform. Some of the features that make SIBS a desirable carrier for stent-based drug delivery are listed in Table 2. The structure of the SIBS triblock copolymer is shown in Fig. 4.

SIBS compositions with a morphology of a continuous polyisobutylene phase containing a discontinuous polystyrene phase exhibit mechanical properties consistent with those of a classical elastomer [28,29]. As seen from the scanning electron micrograph images of the paclitaxel–SIBS coated stents (Fig. 5a and b), the coating offers a smooth, uniform coverage of the stent and good mechanical integrity through the rigors of

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**Table 1**

Examples of favorable characteristics of a polymer-based stent coating

- Homogeneous coverage of the drug along all stent surfaces
- Retention and protection of the drug on the stent when exposed to routine handling
- Prevention of the loss of drug during processing and stent implantation
- Control of drug release
- Prevention of overdosing from non-uniform drug distribution or immediate burst release of the total drug load
- Local, targeted, short-term, low-dose drug delivery

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**Table 2**

Favorable properties of the SIBS polymer

- Elastomeric properties
- Long-term vascular-compatibility
- Biostability
- Processing compatibility with paclitaxel
- Ability to provide uniform paclitaxel distribution and therefore uniform drug coverage/delivery along the stent
- Ability to retain the drug on the stent through the stent deployment procedure
- Ability to prevent mechanical disruption during implantation
- Ability to control the release of paclitaxel
- Stable long-term shelf life
sterilization and stent expansion. It is non-reactive with the drug throughout the manufacturing process (including sterilization and long-term storage) and provides a stable and protective environment for the drug. Additionally, it has an excellent short and long-term vascular compatibility profile in the porcine coronary model, one of the most demanding animal models, currently considered the industry standard in the evaluations of bare metal and drug-eluting stents. The histology and morphometry data comparisons for the SIBS-coated stents (no paclitaxel) and bare metal stents in the porcine coronary artery model (Fig. 6a and b) indicate that SIBS has a safety profile similar to that of BMS and has no adverse impact on the vessel biology [30].

The biostability of SIBS was examined by extracting the polymer from TAXUS Express paclitaxel-eluting stents explanted from porcine coronary arteries. The polymer exhibited no evidence of molecular degradation throughout the in vivo residence periods from 90 days to one year in porcine coronary arteries [31].

### 3. Paclitaxel–SIBS formulation development and evaluation

The strategy for the formulation development of paclitaxel–SIBS coatings and the pre-clinical dosing evaluation involved a phased approach as outlined below:

- **Assessment of the impact of various formulation parameters on the drug release kinetics and determination of cause–effect relationships**
- **Determination of the physicochemical properties of the lead formulation candidates to understand drug–polymer interactions, drug distribution within the matrix, formulation stability, and factors governing release kinetics**
- **Assessment of biological responses to paclitaxel–SIBS coated stents in animal models of restenosis, leading to the identification of candidates for clinical studies**
- **Clinical trials with lead formulation candidates that demonstrated a safety profile in the porcine coronary model**

The ultimate objective of this process was to determine the window of paclitaxel release kinetics...
that would inhibit the restenotic process without compromising the safety elements offered by the bare metal stents.

The following sub-sections discuss the formulation development and physicochemical characterization of various paclitaxel–SIBS formulations.

3.1. Impact of formulation parameters on the drug release kinetics

Paclitaxel solubility in aqueous environments is very low and has been reported to be in the range of 0.3 to 30 μg/ml [22]. It is also known to exist in hydrate and amorphous forms as well as in the crystalline state [22,26,32]. The hydrate form has been shown to have an aqueous solubility lower than that of the amorphous form. The drug can undergo dimerization in solution [27] and is known to strongly adsorb to various surfaces [33,34]. In addition, paclitaxel is hydrophobic and therefore requires a sufficient volume of medium to maintain non-saturation sink conditions [35]. All of these issues, if ignored, can compromise the interpretation of in vitro drug release studies.

The in vitro release kinetics of various drug–polymer formulations was conducted in phosphate buffered saline (PBS) at 37 °C. The surfactant Tween 20 was added to PBS to increase the aqueous solubility of paclitaxel and prevent drug loss through passive adsorption. Media samples were analyzed at selected time points using an isocratic HPLC method for paclitaxel [36,37].

The early development phase of the Taxus DES system focused on modulating the drug release through formulation changes. Formulation parameters, such as the choice of solvents and the drug-to-polymer ratio, were examined in order to obtain the required mechanical performance and integrity of the drug–polymer coating. The selected solvents, a 2-solvent system in the case of Taxus DES, were evaluated to determine the effect on the rate of drug release. A wide range of release profiles could be obtained by varying the solvent ratios (Fig. 7).

Once the basic formulation compositions, stent coating process, and cause–effect relationships critical
for release modulation of paclitaxel from the SIBS matrix were known, the focus of the next phase was identification and characterization of the formulation candidates and paclitaxel release profiles for investigation in pre-clinical studies. The effect of varying the drug to polymer ratios (% w/w of paclitaxel in the coating) and drug dose densities (1 µg paclitaxel/mm² of the stent surface area to 4 µg paclitaxel/mm² of the stent surface area) on paclitaxel release was examined. Fig. 8 shows drug release from various formulations, one set represents progressively increasing % w/w paclitaxel (8.8%, 25%, and 35% w/w), at a dose density of 1 µg paclitaxel/mm² of the stent surface area, while the other represents three different dose densities (1, 2, and 4 µg paclitaxel/mm² of the stent surface area) for the 25% w/w paclitaxel formulation.

As is seen in Fig. 8, increasing the drug to polymer ratio from 8.8% to 35% w/w at a constant dose density had a notable effect on the cumulative amount of drug released. On the other hand, the effect of dose density for the formulation of 25% w/w paclitaxel, was not as dramatic within the ranges examined in the study.

Based on the release kinetics, these formulations were termed Slow-Release (SR—8.8% w/w paclitaxel), Moderate-Release (MR—25% w/w paclitaxel) and Fast Release (FR—35% w/w paclitaxel) formulations.

In the context of available models describing matrix-based drug delivery, the release rate of a drug dispersed as a solid in an inert matrix has been described by Higuchi [38,39]. This model assumes that the solid drug dissolves first from the surface layer of the device. When this layer becomes exhausted of drug, the next layer begins to be depleted by dissolution of the drug and diffusion to the external solution through the inert carrier matrix, a nanoporous structure in the case of the TAXUS™ stent. In this manner, the interface between the region containing the dissolved drug and the region containing the solid dispersed drug moves into the interior of the matrix as a boundary front. The mechanism of release from and release modulation for paclitaxel–SIBS coatings will be discussed in further detail in Section 3.2, in the context of physicochemical characteristics of the drug–polymer matrix.

3.2. Physicochemical characterization of the paclitaxel/SIBS coating matrix

Several optical and spectroscopic analytical techniques were employed to probe the physical and chemical characteristics of the paclitaxel/SIBS matrix. These techniques included scanning electron microscopy (SEM), transmission electron microscopy (TEM), nuclear magnetic resonance (NMR), differential scanning calorimetry (DSC), X-ray powder diffraction (XRPD), IR spectroscopy including FT-IR, near-IR imaging and Raman spectroscopy, high performance liquid chromatography (HPLC), and atomic force microscopy (AFM).

Paclitaxel–SIBS formulations containing 8.8%, 25%, or 35% paclitaxel (w/w) were evaluated using the above techniques. The samples used for analysis
were prepared as either cast films or coated stents, depending on the requirements of the technique used. The materials and methods are reported in detail elsewhere [40,41].

3.2.1. Scanning electron microscopy

The drug–polymer coatings were examined pre- and post-expansion of the coated stents under aqueous conditions. Fig. 9a and b show representative low and high magnification SEM images of the TAXUS Express2 paclitaxel-eluting coronary stent that underwent ethylene oxide sterilization followed by stent expansion. The smooth conformal coating and the lack of cracking, flaking, or other defects, indicate the ability of the coating to withstand the rigors of handling, sterilization, and expansion.

3.2.2. Transmission electron microscopy

Fig. 10a and b are transmission electron micrographs of the SIBS copolymer and of the drug–polymer matrix with 25% paclitaxel in the SIBS copolymer. Both TEM images show well-ordered cylindrical and lamellar morphologies of the SIBS copolymer. Paclitaxel appears to be dispersed in the SIBS matrix as seen in Fig. 10b.

3.2.3. Nuclear magnetic resonance

Solid-state NMR spectra demonstrated a lack of chemical interaction between paclitaxel and SIBS [41]. The results also indicated that a change occurred from crystalline packing to amorphous packing for paclitaxel, possibly due to physical interaction of paclitaxel with SIBS through aromatic π–π interactions [42,43].

3.2.4. X-ray powder diffraction, Raman microspectroscopy, and FT-IR microspectroscopy

X-ray powder diffraction patterns and Raman microspectroscopy results showed that the paclitaxel active pharmaceutical ingredient (API) as received from the manufacturer is crystalline. However, as a component of both the solvent cast films and the stent coating, it is in an unusually stable amorphous state.
3.2.5. Differential scanning calorimetry

DSC scans for solvent cast films consisting of SIBS-only or of 8.8% or 25% (w/w) paclitaxel/SIBS showed two glass transitions [41]. One transition occurs at subambient temperature, related to polyisobutylene (−63 to −71°C), and the other occurs at a higher temperature (90 to 103°C), and is related to the polystyrene components of SIBS [47]. The small decrease in the \( T_g \) of the polystyrene component with the incorporation of paclitaxel in the polymer could be attributed to the weak physical interactions, via aromatic \( \pi \) electrons, a phenomenon also detected in the solid-state NMR study.

3.2.6. Atomic force microscopy

Atomic force microscopy (AFM) is an exceptional tool that allows characterization of both, the surface as well as the bulk physical morphology of the delivery matrix. Tapping Mode AFM was used to image the surface of coated stents. Fig. 11a, b, and c show the topography images, i.e., real-space projections of the coating surface microstructure, of stents coated with SIBS-only, with 8.8% w/w paclitaxel–SIBS formulation, and the same coating after incubation in the in vitro release medium PBS/Tween 20, respectively. The images of the coating in the release medium were taken in a fluid cell of the AFM with an attachment that enables replacement of the PBSTween 20 with fresh release media to circumvent saturation and eventual recrystallization of the paclitaxel onto the SIBS surface.

As seen in the image in Fig. 11a, the surface of the SIBS-only matrix exhibits a typical block copolymer microphase-separated morphology in these surface scans, often showing branched, wavy worm-like cylindrical and spherical structures. The morphology of the surface of coated stents containing 8.8% paclitaxel (Fig. 11b) shows that the drug appears to exist as discrete particles. Shallow voids several nanometers in depth formed on the stent surface after exposure to PBS–Tween 20 (Fig. 11c).

The subsurface morphology of the slow, moderate and fast drug release profile coatings was also examined using AFM and is shown in Fig. 12. The images show that the frequency and size of the paclitaxel-containing domains increase with increasing paclitaxel content in the matrix.

In summary, the physicochemical characterization techniques described in the preceding sections demonstrate that paclitaxel exists as discrete particles embedded in the SIBS matrix. AFM and TEM confirmed the presence of solid paclitaxel particles dispersed on the surface of the SIBS polymer as well as throughout the thickness of the matrix. The thermal analysis and solid-state nuclear magnetic resonance spectroscopy (NMR) results indicated lack of measurable solubility of paclitaxel in the SIBS matrix as well as lack of chemical interactions between the drug and the polymer.

An examination of the release profiles discussed in the previous section (Section 3.1, Fig. 8) in the light of the above findings indicates that the slow (8.8% paclitaxel) and moderate (25% paclitaxel) release systems follow the model of a matrix drug delivery system having a low paclitaxel loading in the SIBS...
polymer. In these systems, the drug undergoes initial dissolution at the stent surface. As the fluid phase boundary of the release medium penetrates into the subsurface bulk of the matrix, the release occurs via dissolution of the drug and slow diffusion through nanometer sized pores. As the drug loading increases to 35%, as in the fast release formulation, the subsurface drug domains increase in size (see AFM images in Fig. 12), and drug release is controlled primarily by drug dissolution and less by diffusion. The high-loading 35% formulation consequently exhibits a significant initial burst compared to the other formulations.

4. Biological evaluations of paclitaxel and paclitaxel–SIBS coated stents

The mechanism of action of paclitaxel for chemotherapy applications is well-documented in the literature and was discussed briefly in Section 2.1 [12–14]. Various in vitro and in vivo studies with paclitaxel relevant to its use in fibroproliferative conditions such as cancer, arthritis, and transplant-rejection suggest that paclitaxel can impact cell migration [48] and angiogenesis [49,50] in addition to suppressing cell proliferation [13]. At lower concentrations, paclitaxel was shown to elicit a dose-dependent cytostatic effect leading to inhibition of cell proliferation without causing cell death [51]. Paclitaxel had an inhibitory effect on T-cell and B-cell proliferation when examined for transplant-rejection applications [52,53] and inhibited MMP synthesis crucial to arthritis and rheumatism applications [54].

Of the drugs investigated for restenosis, paclitaxel has the advantage of having the most pre-clinical and clinical history, albeit in the oncology area. In contrast to oncology therapy, the management of restenosis involves targeting cell types which are non-malignant and the doses delivered via the Taxus system to curb restenosis are orders of magnitude lower than those used for chemotherapeutic applications. With the differences in the cell types, the drug doses, and the delivery regimen for the two scenarios, it is important to understand the effects of paclitaxel in the context of restenosis.

Studies conducted over the course of the last decade have demonstrated that paclitaxel has various biological effects crucial to the vascular repair process involved in the cascade of restenosis, and were discussed previously in Sub-section 2.1. Paclitaxel has been shown to inhibit proliferation of vascular smooth muscle cells (SMC) in in vitro studies [16,55–57], and to inhibit neointimal thickening in various animal models of restenosis, such as porcine coronary, rabbit iliac, and rat carotid arteries [15–18,58–60]. Axel et al. [16] demonstrated that paclitaxel inhibited smooth muscle cell proliferation without affecting cell numbers or cell viability at concentrations as high as 10 μmol/l. Paclitaxel has also been shown to inhibit smooth muscle cell migration and chemotaxis in cell culture models [15,16,57], to inhibit secretion of extracellular matrix proteins [57,61], to prevent aggregation of platelets [62], and to inhibit neo-vascularization-mediated intima progression [63].
4.1. Effects of paclitaxel on human coronary artery Smooth Muscle Cells (SMC)

We evaluated the in vitro effects of paclitaxel on SMC to determine the dose–response relationship on cell proliferation, effect on cell viability, and the mechanism of action at low paclitaxel concentrations relevant to stent-based delivery for coronary restenosis [64]. At three days, paclitaxel at doses as low as 1 ng/ml, inhibited proliferation of SMC by 60% compared with the vehicle control (Fig. 13a). This inhibition of SMC proliferation was not accompanied by cell apoptosis. At paclitaxel concentrations of 12 ng/ml and higher, maximal inhibition of SMC proliferation was observed (Fig. 13b). There was no change in cell numbers, indicating that paclitaxel at these concentrations arrested cell growth but did not cause cell death or cytotoxicity.

Cell cycle arrest in the mitotic phase, a hallmark of paclitaxel’s mechanism of action associated with chemotherapeutic doses, was not observed. Instead, paclitaxel resulted in primary and post-mitotic G1 arrest within the concentration range evaluated (0.01 to 10,000 ng/ml). The SMC treated with paclitaxel maintained normal cellular and nuclear morphology as seen in Fig. 14, and were viable for at least 21 days.

We also examined the effect of paclitaxel on human coronary artery SMC migration in an in vitro cell culture wound assay model. SMC migration was determined by counting cells that migrated into the wound area. As seen from Fig. 15a, paclitaxel potently inhibited SMC migration compared to the control and was effective at concentrations as low as 2 ng/ml.

In summary, the studies discussed above indicate that paclitaxel targets multiple events involved in the arterial restenosis process and that stent-based delivery of low paclitaxel concentrations to the arterial cellular milieu could be considered cytostatic rather than cytotoxic [64]. Additional studies should provide further insights into novel molecular and biochemical pathways relevant to its pharmacological action as an antirestenotic agent.

4.2. Pre-clinical assessment of vascular response to paclitaxel–SIBS coated stents

The vascular response of stents coated with various formulations of paclitaxel–SIBS (different dose densities, total drug doses, and release profiles) was examined in the porcine coronary artery model, a well-accepted model for this application [65]. Stents were implanted into the left anterior descending (LAD), left circumflex (LCX), or right coronary arteries (RCA) having a target diameter of 2.8–3.5 mm. The vascular response was evaluated for a period ranging from 28 days to 1 year depending on the study objectives.

![Fig. 13. a. Cells incubated with varying concentrations of paclitaxel followed by MTT assay at day 3 or day 5. b. Cells incubated with 2 or 12 ng/ml of paclitaxel (solid symbols) or left untreated (open symbol). Percent live cells determined by trypan blue exclusion at indicated time intervals.](image-url)
The early studies focused on assessing the spectrum of vascular response to a wide dose range of paclitaxel, leading to vessel exposure to drug levels orders of magnitudes apart. Paclitaxel dose densities of 0.6, 1, 2, and 4 μg per mm² of the stent surface area, corresponding to total dosages of 50, 85, 175, and

Fig. 14. Normal interphase nuclei (arrowheads) and typical smooth muscle cell morphology with extended cellular processes (arrow) seen in the absence as well as presence of paclitaxel.

Fig. 15. a. Inhibition of SMC migration by paclitaxel in a culture wound assay model. b. Paclitaxel dose–response relationship for SMC migration inhibition. (In collaboration with Dr. M. Blagosklonny).
345 µg paclitaxel per stent were examined. Additionally, the 2 and 4 µg per mm² formulations contained 35% w/w paclitaxel, resulting in fast release formulations. As seen from Fig. 16, the highest drug exposure from the 4 µg/mm², 345 µg, 35% w/w paclitaxel formulation, which had an in vitro cumulative release approximately 100–150 times higher than the lowest dose, resulted in pronounced vessel relaxation, fibrin accumulation, medial thinning, endothelial cell loss, and possible thrombus formation. These adverse effects were minimized in a dose-dependent manner as the drug exposure of the vessel was reduced in response to lower doses. At paclitaxel dose levels of 2, 1, and 0.6 µg/mm², there was a corresponding decrease in the adverse effects, such that endothelial cell loss, medial thinning, fibrin accumulation, and possible thrombus formation were all progressively lower. The 1 µg/mm² dose was found to achieve an acceptable balance between the above-mentioned safety parameters and efficacy measures, indicated by lumen diameter and reduction of restenosis compared to the BMS, in the porcine coronary artery.

In the subsequent studies, the effects of paclitaxel release rates were examined using the 1 µg/mm² dose density. The dose density and the total drug dose were held constant by varying total coat weights and drug to polymer ratios. The slow, moderate, and fast release formulations, with increasing paclitaxel content (8.8%, 25%, and 35% w/w), at the 1 µg/mm² dose density discussed previously in Section 3.1 were used for these studies. As seen from Fig. 17, there is a direct correlation between paclitaxel dose delivered to the artery and the vascular response. The fast release formulation resulted in noticeable fibrin accumulation while the slow-release formulation resulted in patent lumen, struts covered by stable intima, and re-endothelialization with minimal fibrin. The results shown in Fig. 18 further confirm the vascular compatible effects of the slow-release 1 µg/mm² formulation. The histopathology of this formulation in porcine coronary artery at 28 days using fibrin staining is shown in Fig. 18a and b, while Fig. 18c demonstrates re-endothelialization of the Taxus stent in the

Fig. 16. Vascular response in porcine coronary model to varying paclitaxel exposure levels.
porcine coronary artery at 60 days using scanning electron microscopy [66].

Histopathology of the moderate-release formulation (25% w/w paclitaxel, 1 μg/mm²) was also examined in the porcine coronary model from 28 days up to one year. The results shown in Fig. 19, indicate a patent lumen, thin neointima covering the stent struts, preserved media, and a progressing healing response [30].

4.3. In vivo paclitaxel release

For conventional modes of drug delivery (eg. oral or parenteral), the pharmacokinetics of a drug are

Fig. 17. Vascular response to 1 μg/mm² slow, moderate, and fast release formulations in porcine coronary model.

Fig. 18. a and b. Porcine coronary model histopathology for the 1 μg/mm² slow-release formulation at low and high magnifications, respectively. c. Re-endothelialization via SEM for the 1 μg/mm² slow-release formulation in porcine coronary artery (en face section).
typically determined by analyzing drug concentrations in the extracellular fluid (plasma), and/or urine [67,68]. In such situations, the focus is on systemic drug delivery. Following stent-based drug delivery, however, very little drug may enter the systemic circulation, while the adjacent arterial tissue becomes the site for drug delivery and hence the area of interest with respect to pharmacokinetics. The focus in this case is on local drug delivery, with biological activity of the drug occurring at the target site. Consequently, since there is no appreciable paclitaxel concentration in the blood plasma, systemic pharmacokinetic parameters of $t_{\text{max}}$ and $C_{\text{max}}$ cannot be obtained in the traditional fashion.

Research in the field of local arterial pharmaco-kinetics is in its nascent stage due primarily to the relatively recent evidence of the DES as a viable therapeutic modality. Studies conducted using model agents and mathematical modeling have indicated that drug distribution in the arterial wall is a function of the dynamic interplay between the local physiological transport forces, target tissue architecture and biochemical composition, and drug pharmacodynamics [69,70]. These investigators have also addressed the issue that while the prediction of drug profiles may be possible based on qualitative and semi-quantitative data, the precise relationship between tissue response and local concentration remains elusive [71,72].

The in vivo release of paclitaxel from the Paclitaxel–SIPS coating matrix was determined by quantitatively measuring the amount of residual paclitaxel on stents explanted at selected time points post-deployment. A bilateral rabbit iliac model was used, with one stent each placed in the left and the right iliac artery. Slow- and moderate-release formulations of 8.8% and 25% w/w paclitaxel, respectively, with a 1 $\mu$g/mm$^2$ drug dose density were used in these studies.

The residual amount of paclitaxel in the explanted stent was quantitated by HPLC to calculate the amount of paclitaxel released at each time point. As seen in Fig. 20, the in vitro/in vivo correlation (IVIVC) in this study demonstrated that for both,
the slow and moderate-release formulations, the in vivo release of paclitaxel from stents retrieved from the rabbit iliac artery was closely predicted by that observed in the in vitro release model in PBS/Tween 20 medium. The levels of paclitaxel in the systemic circulation were assessed by an LC/MS/MS method in this and various other studies and were found to be below the levels of quantification (LLOQ 0.03 ng/ml) [30].

In another study, rhodamine-labeled paclitaxel was used to develop a model system for visualizing the drug distribution in local arterial tissue (en face preparation). Rabbit iliac arteries implanted with rhodamine–paclitaxel-eluting stents were explanted at various time points up to 14 days, and were subsequently examined using fluorescence microscopy to qualitatively study local arterial tissue drug distribution. The fluorescence was concentrated at the stent struts at earlier time points (Fig. 21a), but spread away from the struts to the non-scaffolded tissue areas by day 14, indicating transport over time of the rhodamine–paclitaxel molecule into the vessel area between the struts. Mean fluorescence intensity of the stented arterial wall, measured after removal of the stent, increased over time, indicating efficient local tissue uptake of paclitaxel (Fig. 21b).

The findings from the in vivo vascular response in porcine coronary model provided directional guidance for the selection of SIBS-based slow- and moderate-release formulations with 8.8% and 25% w/w paclitaxel, respectively, at the 1 μg/mm² dose density, as candidates for clinical investigation.

5. Clinical investigation of the Taxus DES system

A plethora of pharmacological approaches have been investigated for the management of clinical restenosis via drug-eluting stents, with the intent to target either one or more of the pathways involved in the cascade. Some, such as paclitaxel and sirolimus, have shown clear evidence of clinical success with the application of the right dose and release kinetics. Other agents, such as batimastat, dexamethasone, and tacrolimus, showed promise in pre-clinical models, but could not be translated to success in the clinical setting, while still others have been awaiting successful clinical outcomes. The failure of some clinical trials could be attributed to any one of a number of reasons, such as the selection of an inappropriate therapeutic agent, delivery of a subtherapeutic or a toxic dose, suboptimal drug release kinetics or duration of delivery, bio-incompatible polymer carrier, or a combination of the above.

5.1. Overview of the Taxus clinical trials

The safety and efficacy of the polymer-based paclitaxel-eluting TAXUS stent system (1 μg/mm² total loaded dose) in the management of occlusive coronary artery disease have been investigated in 6 studies: TAXUS I [73,74], TAXUS II [2], TAXUS III [75], TAXUS IV [1,76], TAXUS V de novo [77] and TAXUS VI [3]. All studies, with the exception of Taxus III, were randomized, double-blind, multicenter investigations that compared the paclitaxel-
eluting TAXUS stent with bare metal stents. TAXUS III was an open-label investigation.

Inclusion criteria for these studies included patients with stable or unstable angina or silent/provokable ischemia who required percutaneous coronary intervention. Exclusion criteria generally included recent coronary intervention, previous or planned intravascular brachytherapy in the target vessel, recent MI or stroke, left ventricular ejection fraction <25–30%, renal dysfunction, or contraindications for antiplatelet therapy. Patients received aspirin (75, 80, or 325 mg) and clopidogrel 300 mg before the procedure, and aspirin (>80 or 325 mg/day indefinitely) and clopidogrel (75 mg/day) or ticlopidine (250 mg twice daily) for 6 months after the procedure. Heparin was given before, during, and after the procedure according to each institution’s protocol. Primary endpoints varied by study and included one or more of the following: 30-day major adverse cardiac events (MACE) (TAXUS I and TAXUS III), 6-month in-stent volume obstruction caused by neointimal proliferation (TAXUS II), and 9-month ischemia driven target vessel revascularization (TVR) (TAXUS IV, TAXUS V de novo, and TAXUS VI). MACE was generally defined as cardiac death, myocardial infarction (MI, both Q-wave and non-Q-wave), and TVR. TVR generally included re-intervention (by either coronary artery bypass surgery or percutaneous intervention) of the target lesion (TLR) or re-intervention of the target vessel outside of the target lesion (non-TLR or TVR Remote). TLR–or retreatment rate–is accepted as an important clinical indicator of the performance of drug-eluting stent technology. The following subsections provide a brief outline and outcomes of each trial to date.

5.1.1. TAXUS I

TAXUS I was a randomized, double-blind, multicenter feasibility study designed to assess the safety of Boston Scientific Corporation’s TAXUS slow-release, paclitaxel-eluting NIRx stent versus a NIR control bare metal stent. The trial was conducted at three centers in Germany and enrolled 61 patients. The primary endpoint, the 30-day MACE rate, was zero percent in both the control group and the TAXUS group. At 12 months, the MACE rates were 10% and three percent in the control and TAXUS groups, respectively, demonstrating excellent long-term safety. These excellent results were maintained through the 4-year follow-up with no new MACE in any TAXUS patient between 1 and 4 years. Importantly, no stent thromboses occurred in either treatment group out to 4 years of follow-up.

5.1.2. TAXUS II

TAXUS II was a 536-patient, 38-site, randomized, double-blind, controlled study of the safety and efficacy of the TAXUS paclitaxel-eluting coronary stent system on the NIRx stent platform. In this study, two sequential cohorts (testing the slow- and moderate-release TAXUS formulations) enrolled patients with standard risk, de novo coronary artery lesions. The six-month results showed strong clinical performance as demonstrated by lower MACE and TLR rates in TAXUS patients compared with patients who received a bare metal (NIR) stent. The six-month MACE rates were reduced from 19.8% in control to 8.5% for the slow-release formulation cohort (p=0.0035) and 7.8% for the moderate-release formulation cohort (p=0.0019). At 12 months, the MACE rate in the slow-release cohort remained low at 10.9% compared with the control rate of 21.7% (p=0.0082) and the TLR rate was 4.7% compared with 14.4% for the control (p=0.0035). At 12 months, the moderate-release formulation cohort reported a 9.9% MACE rate (p=0.0048 versus control) with a 3.8% TLR rate (p=0.0010 versus control). Between six and 12 months, no additional TLRS were reported in the slow-release cohort and only one additional TLR was reported in the moderate-release cohort. The binary restenosis rates, defined as 50% or greater vessel re-occlusion as determined by quantitative coronary angiography (QCA), were determined at 6 months for both formulations and compared to the bare metal stent control group. The in-segment (includes the stent length plus 5 mm on either side of the stent) binary restenosis rates were 5.5% and 8.6% for the slow- and moderate-release formulation cohorts, respectively compared with 22.0% for the control group (p<0.0001 and p=0.0010, respectively).

The two-year follow-up for TAXUS II included the largest, long-term QCA and intravascular ultrasound (IVUS) substudy of any drug-eluting stent trial to date, and demonstrated continued safety and efficacy
benefits associated with the TAXUS stent system. Incidences of aneurysms, incomplete apposition (separation of the stent from the vessel wall) and stent thromboses were all low and comparable to control rates. The study’s clinical results indicated a continued significant difference in TLR rates for both formulation cohorts as compared to the control group (5.5% and 3.9% for the slow-release and moderate-release formulation cohorts, respectively as compared with 15.5% for the combined control group, \( p < 0.005 \) for both). QCA and IVUS data demonstrated stable late loss and neointimal hyperplasia formation in the stented vessel out to two years with sustained statistical benefit over the control group.

These excellent results were maintained through the 3-year clinical follow-up with no new TLR or stent thromboses occurring in the TAXUS groups.

5.1.3. TAXUS III

TAXUS III was a single-arm registry examining the feasibility of implanting up to two paclitaxel-eluting stents for the treatment of in-stent restenosis. This group represented patients with recurrent occlusion within a stent. The trial’s main focus was safety, and the primary endpoint was 30-day MACE. The trial enrolled 29 patients, and 28 were treated with the TAXUS paclitaxel-eluting coronary stent system (NIRx stent platform). The trial confirmed safety and reported no stent thromboses, no deaths, and a binary restenosis rate of 16% within the TAXUS stent at 6 months. From 6 months to 3 years, there was only one cardiac death and no stent thromboses.

5.1.4. TAXUS IV

TAXUS IV was a 1314-patient, prospective, randomized, double-blind study designed to assess the safety and efficacy of the slow-release formulation (on the Express stent platform) compared to the control bare metal Express stent for the treatment of de novo coronary lesions. The study’s primary endpoint—target vessel revascularization (TVR) at 9 months—was 4.7% in the TAXUS group versus the control rate of 12.0% (\( p < 0.0001 \)). The study reported a nine-month TLR rate of 3.0% in the TAXUS group compared with 11.3% in the control group (\( p < 0.0001 \)), an in-segment binary restenosis rate of 7.9% in the TAXUS group compared with 26.6% in the control group (\( p < 0.0001 \)), and in-stent binary restenosis rate of 5.5% in the TAXUS group compared with 24.4% in the control group (\( p < 0.0001 \)). The 12-month results continued to support the safety and efficacy, as demonstrated by low rates of MACE (10.6% for TAXUS vs. 19.8% for the control group, \( p < 0.0001 \)), TVR (6.8% for TAXUS vs. 16.7% in the control group, \( p < 0.0001 \)) and TLR (4.2% for TAXUS vs. 14.7% in the control group, \( p < 0.0001 \)).

The benefits for patients who received the TAXUS stent compared to the patients who received a bare metal stent, were maintained at two years, as evident in the 5.6% TLR rate for the TAXUS group compared with 17.4% for the control group (\( p < 0.0001 \)). Thus, the rate of patients living free of TLR events was 94.4% at two years for the TAXUS group, compared to 82.6% for the control group. This benefit was retained at three years as demonstrated by a TLR rate of 6.9% in the TAXUS group versus 18.6% in the control group (\( p < 0.0001 \)).

Results for diabetic patients (including oral and insulin-requiring diabetics) in the TAXUS group showed that benefits were maintained at three years even for this group, notorious for a greater susceptibility and likelihood of restenosis following angioplasty or stenting. The TLR rate for diabetics was 9.8% compared to 23.3% in the control group (\( p = 0.0009 \)). The excellent clinical results in the diabetic subset, which represents approximately 25% of coronary interventions, suggest that this group may stand to benefit substantially from the DES technology.

5.1.5. TAXUS V de novo

TAXUS V de novo was a randomized, double-blind trial studying 1156 patients at 66 sites in the United States, assessing the safety and efficacy of the slow-release formulation (Express\(^2\) stent platform) in reducing restenosis in de novo lesions 10–46 mm in length and 2.25–4.0 mm in diameter. TAXUS V de novo expanded on the TAXUS IV pivotal trial by studying a higher-risk patient population, including patients with small vessels, large vessels and long lesions requiring multiple overlapping stents—the most challenging lesions and highest-risk patients ever studied in a randomized, controlled drug-eluting stent trial in the United States. The primary endpoint of the trial was nine-month TVR, which was significantly lower in the TAXUS group (12.1%) than
in the control group (17.3%) \( (p=0.0184) \). The safety of the TAXUS system in this very complex and challenging group was demonstrated by low overall rates of MACE and stent thrombosis. At nine months, the overall MACE rate in the TAXUS group was 15.0%, compared with 21.2% in the control bare metal stent group \( (p=0.0084) \). The study also reported a TLR rate of 8.6% in the TAXUS group compared with 15.7% in the control group \( (p=0.0003) \). In addition, stent thrombosis rates were identical between TAXUS and control stents (0.7% each for the TAXUS and the control group). These clinical benefits were maintained at 1 year with an overall MACE rate of 18.9% in the TAXUS group compared to 25.9% in the control group \( (p=0.0052) \) and a TLR rate of 11.2% in the TAXUS group versus 19.0% in the control group \( (p=0.0003) \).

The study found significant improvements in the more sensitive, quantitative angiographic measurements (in-segment, in-stent and at the edges) with an in-segment binary restenosis rate of 18.9% in the TAXUS group compared with 33.9% in the control group \( (p<0.0001) \), and an in-stent binary restenosis rate of 13.7% in the TAXUS group compared with 31.9% in the control group \( (p<0.0001) \). Additionally, improvements were seen in in-segment percent diameter stenosis (33.63% in the TAXUS group versus 42.34% in the control group; \( p<0.0001 \)), in-segment minimum lumen diameter (1.81 mm in the TAXUS group versus 1.57 mm in the control group; \( p<0.0001 \)) and in-segment late lumen loss (0.33 mm in the TAXUS group versus 0.60 mm in the control group; \( p<0.0001 \)).

The IVUS observations for the longest lesions ever studied by IVUS in a DES clinical trial, confirmed the consistent performance of the TAXUS system in these high-risk patients and complex lesions. Patients in the TAXUS group exhibited consistent neointimal supression out to nine months across the entire length of the lesions, resulting in stable loss with sustained clinical benefit over the control group. Efficacy and safety results remained consistent with findings from other ongoing TAXUS clinical trials, including TAXUS II, IV, and VI.

The nine-month diabetic sub-population analysis from TAXUS V de novo further supported the efficacy demonstrating significant improvements among diabetic patients receiving the TAXUS system versus those in the control group. The nine-month TLR for the medically treated diabetic sub-population of the TAXUS group was 9.6% compared with 17.5% in the control group \( (p=0.0406) \). An improved in-segment binary restenosis rate of 18.2% was observed in the TAXUS group compared with 38.4% in the control group \( (p<0.0001) \). In addition to the improved restenosis rates, marked improvement in the in-segment late loss was also seen in the diabetic sub-population of the TAXUS group compared to the control group (0.31 \( \pm 0.56 \) mm versus 0.62 \( \pm 0.61 \) mm; \( p<0.0001 \)). Again, these excellent results in diabetics were maintained through 1 year with a TLR rate of 10.9% in the medically treated diabetic subset of the TAXUS group compared to 20.0% in the control group \( (p=0.0245) \).

5.1.6. TAXUS VI

TAXUS VI, an international trial studying 446 patients with complex coronary artery disease at 44 sites, was designed to establish the safety and efficacy of the moderate-release formulation (on the Express stent platform) in the treatment of longer lesions. The trial had a primary endpoint of nine-month TVR. The study’s TVR rate was 9.1% in the TAXUS group versus the control rate of 19.4% at nine-months \( (p=0.0027) \). Additionally, the TAXUS group had a TLR rate of 6.8% (compared with 18.9% in the control group, \( p=0.0001 \)) and an in-segment binary restenosis rate of 12.4% (compared with 35.7% in the control group, \( p<0.0001 \)). The results supported safety as demonstrated by low MACE rates (16.4% MACE rate at nine months in the TAXUS group compared with 22.5% in the control group, \( p=0.1208 \)).

The two-year follow-up data for TAXUS VI demonstrated that the safety and efficacy benefits associated with a moderate-release formulation of the TAXUS Express paclitaxel-eluting stent system were maintained at two years; a continued significant reduction in TLR rate (9.7% for the TAXUS group, as compared with 21.0% for the control group, \( p=0.0013 \)). Stent thromboses remained low and comparable to control rates (0.9% for both the TAXUS group and the control group).

The two-year results for TAXUS VI support long-term safety with increased local exposure of the vessel
to paclitaxel released from the moderate-release formulation compared to the levels released from the slow-release formulation. Even with an in vitro dosing rate 8–10 times greater than the commercialized slow-release formulation, no compromise in safety was observed.

5.2. Cumulative long-term clinical safety and efficacy data

Long-term clinical data is an important measure of a successful new technology, particularly for one as ground-breaking and complex as the DES technology. An analysis of all 3445 patients from TAXUS II, IV, V, and VI trials, was conducted to determine whether the TAXUS system offers sustained safety and efficacy at various follow-up time-points up to three years [78]. Cumulative data from these four trials with different follow-up times (through 1 year for TAXUS V de novo, through 2 years for TAXUS VI, and through 3 years for TAXUS II and TAXUS IV) is shown in Fig. 22a, b, c, and d. As seen from the figures, through three years, the TAXUS stent has a safety profile similar to that of the control stent as gauged by the key safety parameters; stent thrombosis (99.2% for control vs. 98.7% for TAXUS, $p=0.36$), cardiac death (98.0% for control vs. 97.9% for TAXUS, $p=0.81$), and MI (93.6% for control vs. 93.4% for TAXUS, $p=0.96$). The patients receiving TAXUS stents continued to gain from the efficacy benefit as demonstrated by the freedom from TLR offered by the TAXUS stent (80.1% for control vs. 90.6% for Taxus, $p<0.0001$). As seen from Fig. 22c, there is an incremental efficacy differential between the TAXUS group and the Control.

Fig. 22. Key long-term safety and efficacy clinical parameters for the Taxus Express paclitaxel-eluting stent system. Freedom from a. stent thrombosis, b. cardiac death, c. TLR, and d. MI.
group over time (Delta of 9.1% at 9 months vs. 10.5% at 3 years). These data provide clear evidence of the long-term efficacy of TAXUS while maintaining the desirable safety features of the bare metal stent.

6. Conclusions

The Taxus™ Express²™ paclitaxel-eluting coronary stent system was launched in the international markets in 2003 and in the United States in 2004. The enormous success of drug-eluting stents, now hailed as the pioneering technology in the interventional cardiology community, was a culmination of effectively and appropriately linking multi-disciplinary efforts, including pharmacology, polymer science, formulation research, process engineering, analytical characterization, controlled drug delivery, cell biology, pharmacokinetics, pathology, pre-clinical research, and clinical science. The successful clinical outcomes of the Taxus™ paclitaxel-eluting stent system demonstrate the importance of the early technology development process, understanding the requirements of the product on the onset, anticipating the technical risks inherent to the development of a new technology in the context of the requirements, utilizing an effective cross-pollination process to draw from appropriate scientific areas, and building a strong technology foundation that paves the path towards the desired clinical goals.

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