Skoltech
Skolkovo Institute of Science and Technology

# Thesis Changes Log

**Name of Candidate:** Proshin Pavel

**PhD Program:** Materials Science and Engineering

**Title of Thesis:** Films with pattern-placed drug for use in personalized medicine

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*The thesis document includes the following changes in answer to the external review process.*

**Headers and levels were reconsidered:**
4.1 Introduction to PLACE Technology (Level 2 header)
4.2 Film Fabrication and Parameter Optimization (Level 2 header)
4.2.1 PVA Matrix Optimization (Level 3 header)
4.2.2 Drug Loading Studies (Level 3 header)
4.3 Drug Release Characterization (Level 2 header)
4.3.1 Impact of Laser Microperforation (Level 3 header)
4.3.2 Influence of Porogen Agents (Level 3 header)
4.4 E-beam Sterilization and Film Stability (Level 2 header)
4.4.1 Impact on PLGA Films (Level 3 header)
4.4.2 Impact on Release Properties (Level 3 header)
4.5 Multilayered Film Development (Level 2 header)
5. PLACE Technology: Application and Adhesion Studies

**4.1 Introduction to PLACE Technology**
**Answering the question 1 from review of A.Ermakov next text was changed:**
The maximum flow test for the pump was conducted following the methods described in Chapter~\ref{MM}. Given that our coatings were intended to modify the surface of bone implants and dressing material over relatively large areas (several $dm^2$) while ensuring a high drug load up to milligrams per square centimeter, we selected a 23G needle (330 \textmu m ID).

**4.2.2 Drug loading studies:**
**Answering the question 2 from review of A.Ermakov and the question 4 from review of prof. C.He next text was changed. Since we deal with micron-sized roughness - SEM is enough for screening.:**
The drug-containing mixtures must meet several requirements. Firstly, the maximum drug loading should not significantly increase the viscosity of the matrix, which could lead to gaps in the print. Secondly, the final printed track should be smooth without any microns-sized roughness that could cause wetting defects.
…
Comparison of samples Cef100 and Cef400 without a cover film revealed increased surface microns-sized roughness due to Cefazolin crystallization (Fig.~\ref{fig:Cefx}). In the Cef100 series, samples were formulated with an approximate 1:1 PVA-to-drug ratio, which effectively minimized Cefazolin crystallization due to the high content of amorphous PVA. As the PVA content decreased, however, crystallized regions became prominent, fully covering the Cef400 sample where the PVA concentration was approximately 20\%. Such rough surfaces with microns-sized roughness comparable with film thickness can introduce wetting

defects, resulting in numerous micron- and submicron pores and increased susceptibility to mechanical stress, such as bending or pressure, which may damage the film and accelerate drug release.

**4.2.3.{Base and cover films forming}**
**Answering the question 4 from review of A.Ermakov Figure 4.8 changed (were added error bars, style changed)**

**2.3 {Drug type, dosage and stability issue}**
**Caption changed, answering the question 1 from review of prof. S.Chvalun and the questions 1/3 from review of prof. C.He next text was added (also slides will be supplemented by info about proteins and non-soluble drugs to discuss it):**
Water-soluble drugs are preferred in pharmaceutical development due to their inherent advantages in absorption, bioavailability, and patient safety. A drug's ability to dissolve in water is crucial for effective absorption into the bloodstream, which directly impacts its therapeutic efficacy. Poorly soluble drugs, while common, present challenges that modern formulation technologies have sought to overcome.

One major advantage of water-soluble drugs is higher bioavailability. These drugs dissolve easily in gastrointestinal fluids after oral administration, leading to faster absorption, quicker onset of action, and more predictable therapeutic effects. In contrast, poorly soluble drugs often exhibit erratic absorption, complicating the prediction of their effects. For intravenous administration, water solubility is even more critical, as it ensures the drug can directly enter the bloodstream without causing precipitation or requiring complex solubilization strategies. Poorly soluble drugs administered intravenously risk precipitation, which can lead to complications such as tissue irritation or embolism. Water-soluble drugs also pose a lower risk of tissue accumulation, reducing long-term toxicity. Poorly soluble drugs may precipitate in tissues, forming aggregates that can cause irritation or damage. Studies have shown that poorly soluble drugs are more likely to form deposits in non-target tissues, which increases the risk of adverse effects.

Another advantage of water-soluble drugs is their ease of formulation into various dosage forms, including syrups, injections, and suspensions, facilitating both oral and intravenous routes of administration. Poorly soluble drugs, however, require complex formulation strategies such as nanoparticles or lipid-based systems to enhance solubility, which increases production costs and regulatory hurdles. Despite these issues, poorly soluble drugs remain prevalent, and their solubility challenges lead to variable bioavailability. Inconsistent absorption can result in subtherapeutic levels, requiring higher doses that increase toxicity risks. For example, poorly soluble drugs are often associated with a narrow therapeutic window, where the effective and toxic doses are close together.

Modern approaches like nanoparticles and solid dispersions have been developed to enhance the solubility and bioavailability of these drugs. By reducing particle size or using solubilizing agents, these strategies increase dissolution rates, improving the therapeutic outcomes of poorly soluble drugs and reducing the risks of dose-dependent toxicity.

In my dissertation, I will develop a system specifically designed for water-soluble drugs due to their inherent advantages in pharmaceutical applications. By focusing on soluble drugs, the system can ensure consistent absorption, avoid the complexities associated with poorly soluble drugs, and simplify formulation, making it more effective and reliable for practical use.

**4.3.3.2{Use of PVP as pore-generating agent}**
**Answering the questions 8/9 from review of prof. C.He next text was rewritten, were added clusters dimensions:**
Clusters of PVP, measuring only 0.5 to 2 microns in size, appeared during the long-term accumulation of an image due to the heating of the film by an electron beam. The average area of these clusters was calculated to be approximately 0.25 $\mu m^2$.
**Figure 4.28 was supplemented with a cluster size distribution histogram.**
**Figure 4.30 was supplemented with a cluster size distribution histogram.**

Despite the efforts to achieve a homogenous distribution of PVP clusters, complete uniformity could not be attained. The cryo-section analysis of the film reveals a gradient in the cluster size and distribution, with both parameters increasing from the bottom surface toward the top. This results in a heterojunction-like structure between the two polymers. In such a configuration, the formation of a substantial number of through-pores is unlikely. However, this arrangement is expected to enhance the film's hydrophilicity, leading to increased swelling and the development of water-filled nanopores. Consequently, these factors would improve the film's permeability and accelerate its degradation rate.

**Answering the question 2 from review of prof. S.Chvalun and question 7 from review of prof. C.He Figure 4.31 was added. The SEM image of the edge of the PLGA film containing 10 wt.\% of PVP K17**

**4.2 { Film fabrication and parameter optimization }**
**Answering the question 2 from review of prof. C.He next text was added:**

Several strict requirements were established as the foundation for the binding medium for drug particles: it must be water-based, free of harmful and non-biocompatible components, exhibit low reactivity, be insoluble in organic solvents, and possess sufficient viscosity for extrusion through a thin needle while retaining its extruded form. These stringent criteria narrowed the list of candidates to a single option: polyvinyl alcohol (PVA). PVA is FDA-approved for human use, available in medical-grade formulations, readily dissolves in water to form fluid gels even at low concentrations, and demonstrates inertness when mixed with most drugs. In this chapter, we explore several critical parameters that influence the fidelity and quality of printed PVA water-based solutions. These parameters include printing speed, molecular weight of PVA, degree of hydrolysis, and solution concentration, all of which play key roles in determining the wetting, spreading, and overall behavior of the solution on the substrate.
The molecular weight (MW) of PVA directly influences the viscosity of the solution, which in turn affects its spreading and printability. Higher molecular weight PVA (greater than 200 kDa) increases solution viscosity, allowing for better control of material spread during printing and enabling the formation of structures with minimal PVA content. However, higher molecular weight also means that the PVA will take longer to degrade and be eliminated from the body, which may be undesirable in biomedical applications. Conversely, lower molecular weight PVA requires higher concentrations to achieve the necessary viscosity for printing, complicating the formulation and affecting the properties of the final product.
In this study, a moderate molecular weight PVA in the 50-100 kDa range was selected. This choice strikes a balance between achieving the appropriate viscosity for stable deposition and ensuring that the PVA does not persist in the body for extended periods, making it suitable for biomedical applications such as drug-eluting films.
The degree of hydrolysis of polyvinyl alcohol refers to the extent to which acetate groups in the original polyvinyl acetate (PVAc) have been converted to hydroxyl groups. Fully hydrolyzed PVA typically shows enhanced adhesion to hydrophilic substrates and forms stable films, making it suitable for applications requiring strong bonding to polar materials. However, its high hydrophilicity may lead to excessive swelling, particularly in aqueous environments, which can deform the printed structures and affect release kinetics. Conversely, partially hydrolyzed PVA (such as 85% hydrolyzed) offers a more balanced solution. It maintains good solubility while preventing excessive swelling, thereby promoting stability and precision in drug-loaded films.
The concentration of the PVA solution plays a pivotal role in determining the viscosity, which is closely tied to the molecular weight of the polymer. Higher concentrations of PVA in solution result in increased viscosity, improving control over the printed shape and reducing excessive spreading. However, high concentrations may also cause difficulties in achieving uniform coverage of the substrate and increase the likelihood of clogging during the printing process. In this work, a range of PVA concentrations was tested to find the optimal balance between solution viscosity and printability.
The speed of the printing head directly impacts the wetting and spreading behavior of the PVA solution on the substrate. A slower printing speed allows the solution more time to wet and spread across the surface, leading to uniform coverage and better adhesion. However, excessively slow speeds may cause over-saturation and lead to deformation of the printed structure. On the other hand, faster speeds may reduce the wetting time, causing incomplete spreading and the formation of discontinuities. Optimizing the speed is critical for ensuring a balance between spreading and adhesion, which is essential for achieving consistent and accurate deposition.

In this study, two types of polyvinyl alcohol (PVA) were tested: partially hydrolyzed (PG) with 85% hydrolysis and fully hydrolyzed (FG) variants. A moderate molecular weight range of 70-100 kDa was selected to balance viscosity and spreading behavior. The PG variant had a lower molecular weight of approximately 70 kDa, while the FG variant had a higher molecular weight of about 90-100 kDa. These selections were made to optimize the printing process for stable, high-fidelity drug-eluting films, which were applied to a pre-coated substrate. By adjusting the solution concentration, molecular weight, and degree of hydrolysis, the process was fine-tuned to achieve the desired performance characteristics.
During experiments, it was observed that there was no notable difference in wettability between the 85% hydrolyzed and FG PVA on the substrates used. Based on this finding, 85% hydrolyzed PVA was selected to reduce potential swelling within the films. Additionally, solutions of PG PVA exhibited slightly higher viscosity due to their lower molecular weight, allowing for the use of smaller amounts of PVA additive to achieve the desired consistency and viscosity. This approach minimized the polymer content while maintaining control over the printing fidelity and structural stability in the release profiles.

**Next text was rewritten to improve clarity:**
A PVA matrix was applied to a PP substrate, which had been pre-coated with PLGA films, using a modified 3D printer. The sample models for printing were created as solid blocks (10 × 10 × 0.01 mm) using Fusion360 CAD software and saved as \textit{.stl} files. These models were sliced using PrusaSlicer software with the following settings: a 0.3 mm extrusion width, no solid fill layers at the top or bottom, 50\% infill density, a printing speed of 5-20 mm/s, and a travel speed of 100 mm/s. A 50\% infill snake-like pattern was chosen for the drug-eluting films (DEFs), as it provided a simple and efficient continuous fill without overlapping lines. The resulting g-code was used directly for the printing process without any further modifications.

This process was conducted under standard conditions using a 23G needle (300 \textmu m inner diameter) with a Z-offset of 150 \textmu m (equal to the needle's radius to avoid material buildup). The coated substrates were then dried at 40°C for 10 minutes in a vacuum oven.

**Figure 4.4 changed (second PVA graph added).**

**\subsection{Drug loading studies}**
**Answering the question 4 from review of prof. C.He next text was rewritten to improve clarity, some refs were added:**
Comparison of samples Cef100 and Cef400 without a cover film revealed increased surface roughness due to Cefazolin crystallization. In the Cef100 series, samples were formulated with an approximate 1:1 PVA-to-drug ratio, which effectively minimized Cefazolin crystallization due to the high content of amorphous PVA. As the PVA content decreased, however, crystallized regions became prominent, fully covering the Cef400 sample where the PVA concentration was approximately 20\%. Such rough surfaces can introduce wetting defects, resulting in numerous micron- and submicron pores and increased susceptibility to mechanical stress, such as bending or pressure, which may damage the film and accelerate drug release.

The crystallization of drugs within delivery systems remains a significant challenge in pharmacology, as it can affect the stability and release profiles of the active ingredient. Incorporating PVA or other water-soluble polymers has become a common approach to counteract this issue, as these polymers can reduce crystallization through steric hindrance and specific intermolecular interactions with the drug molecules. By enhancing the amorphous phase, polymers like PVA provide structural support and help maintain a more uniform, stable film.

In addition to incorporating crystallization inhibitors like surfactants, oligomers, or additional polymers, modifications to the drying process can also be beneficial. For instance, printing on a heated bed or applying warm air can accelerate solvent evaporation, thereby reducing the time available for crystallization to occur.

**\section{Drug release from PLACE films with pore-generating agents}**
**Answering the question 3 from review of prof. V.Paunov next text was rewritten to improve clarity, some refs were added:**
To create a film with regulated porosity, diverse porogens can be utilized. Typically, these are polymers soluble in both water and organic solvents. PEG and PVP emerge as prime choices for porogenic additives, both being FDA-approved for medicinal use, emphasizing their biomedical applicability and safety. PEG and PVP were

selected as inert additives in this work due to their biocompatibility and solubility in both water and the initial polymer solution, making them convenient for achieving controlled porosity in the final films. These additives can dissolve and leach out in vivo without adverse effects, facilitating gradual drug release through the pores left behind. Traditional solid porogenic agents, such as salts, sugars, and gelatin nanoparticles, present greater challenges for submicron porosity creation, as they require precise dispersion and stabilization to prevent agglomeration. Furthermore, the use of solid porogens in biological applications can introduce risks of inflammatory responses, osmotic imbalance, or unexpected immune reactions due to residual particles. PEG and PVP offer a simpler, safer alternative for generating consistent porosity without these complications. Consequently, the use of PEG and PVP as porogens enables direct application of the porogen-containing layer onto the drug-loaded layer, eliminating the need for post-processing steps such as leaching or etching. This streamlined process minimizes the risk of drug loss or degradation, ensuring a more efficient production and reliable performance of the final film.

Nevertheless, incorporating hydrophilic porogens such as PEG and PVP into a hydrophobic PLGA matrix can pose challenges due to potential phase separation, impacting film uniformity and performance. The molecular weight (MW) of these additives significantly influences their compatibility with PLGA. Lower MW additives generally exhibit better miscibility with PLGA than their higher MW counterparts. This propensity is attributed to the ease with which smaller molecules permeate and disperse within the polymer matrix.

**Answering the question 5 from review of prof. V.Paunov**
**\section{Cost-Effectiveness and Production Feasibility of Biodegradable PLACE Films for Medical Applications} was added:**

The economic viability and scalability of biocompatible, biodegradable films for drug delivery are promising, driven by the affordability of materials, feasible manufacturing methods, clinical benefits, regulatory compatibility, and long-term market potential.

Material Costs:
The choice of medical-grade, biobased polymers provides an affordable base for these films, particularly as domestic suppliers in Russia can meet the demand, reducing import-related expenses. The films themselves are extremely thin, meaning that the material consumption remains low, even for mass production, further enhancing cost-effectiveness. Solvent-based processes also provide an efficient use of polymer solutions, allowing for precise deposition and minimal waste. Complementary materials, such as polyvinyl alcohol (PVA) and polyvinylpyrrolidone (PVP), are readily available at very low costs, making them ideal for structural support or porogen functions within the films. While the primary solvents used in production are economical, any shift toward more environmentally friendly solvents, like ethyl acetate or ethyl lactate, would increase costs slightly, though it might be justified by regulatory and market demands for sustainable practices.

Manufacturing Feasibility:
The selected synthesis techniques, including 3D printing and direct coating, offer high scalability and versatility. These methods can readily be adapted to both small and large-scale production requirements. For applications that require individual, patient-specific implants, the current manufacturing setup can support a limited production volume, approximately hundreds of devices per year. However, this technology can be scaled up through a roll-to-roll (R2R) production line, which would allow the high-throughput manufacture of standardized films for larger-scale clinical needs, potentially reaching tens of thousands of units per year. This flexibility ensures that the technology can be adapted from small-batch to mass production settings without major overhauls in infrastructure.

Clinical Benefits and Demand:
The controlled-release and biodegradable characteristics of these films offer distinct clinical advantages. Localized drug delivery, enabled by the films' controlled-release mechanisms, can lower systemic side effects and improve patient compliance by providing sustained therapeutic effects at the target site. This is particularly beneficial in applications requiring infection prevention and combination drug therapies, such as wound care, implant coatings, and other localized treatments. By reducing infection rates, these materials could potentially shorten hospital stays, improve recovery times, and minimize the need for repeat interventions, thereby addressing a significant clinical demand in the market.

Regulatory and Safety Considerations:
The use of biocompatible polymers was specifically chosen to facilitate regulatory approval, as these materials reduce the risk of adverse reactions and enhance patient safety. This simplifies the regulatory pathway, given that patient safety and predictable biodegradation are fundamental requirements in medical material approval processes. However, incorporating drugs into the films demands stringent regulatory oversight, as special conditions are needed to ensure drug stability and efficacy. Producing these films in sterile conditions, rather than relying on post-sterilization by radiation, preserves both the physical properties of the film and the therapeutic integrity of the drug. Leveraging the infrastructure of an existing clean room within a prosthetics production facility could streamline compliance with sterilization and regulatory standards while minimizing costs associated with additional facility requirements.

Long-term Potential and Return on Investment (ROI):
These films' potential to improve clinical outcomes has a direct positive impact on ROI. By reducing the incidence of infections and the need for follow-up interventions, these materials could lower the overall healthcare costs associated with implant procedures and wound care. Furthermore, their adaptability for various therapeutic applications, combined with high scalability, suggests a strong long-term potential for both clinical adoption and financial sustainability. The market demand for safe, biodegradable, and effective drug-delivery systems positions this technology favorably for investment and clinical expansion, potentially achieving sustainable revenue through its broad applicability and patient-centered benefits.

**Answering the question 4 from review of prof. D.Gorin \section{Future Directions and Perspectives} was added:**
The developed technology for controlled drug delivery using biodegradable polymer films offers a promising foundation for further innovation and clinical applications. Future work will aim to refine and expand upon the initial findings to optimize performance, enhance sustainability, and explore new biomedical applications. One key focus is the exploration of more stable porogens that offer both reliability and compatibility with biological systems. Investigating alternative polymers, such as poly(2-ethyl-2-oxazoline) or even biodegradable PVA nanoparticles, could allow for improved porosity control, greater material stability, and enhanced biocompatibility, leading to more finely tuned drug release profiles.
Expanding the potential applications of these multilayered (ML) films is another exciting avenue. The technology shows promise for developing advanced wound healing patches that combine prolonged antimicrobial release with tissue regeneration properties. Additionally, the films could be adapted as self-expanding meshes suitable for endoscopic delivery, providing non-invasive, targeted treatment options for hard-to-reach areas in the body.
To align with environmentally conscious practices, there is a strong motivation to develop greener production methods. This would involve replacing commonly used solvents with more bio-friendly options, such as ethyl acetate or ethyl lactate. Such a shift could reduce environmental impact and improve safety in manufacturing, making the process more sustainable and suitable for larger-scale production.
Pilot testing in collaboration with SamSMU will serve as a critical step for process improvements and validation of clinical efficacy. Early-stage trials will help fine-tune technology parameters, optimize drug release kinetics, and address scalability challenges, providing data essential for regulatory approval and wider clinical adoption.
Lastly, expanding the technology to encapsulate non-soluble drugs, peptides, and biologics represents a compelling future direction. By modifying the film structure and porogen composition, this platform could be adapted to deliver a broader range of therapeutics, including those that present solubility challenges, thereby broadening the clinical applications and treatment options available through this innovative material technology.
In summary, these future steps will build on the success of the current work to advance the applicability, scalability, and sustainability of these films, ensuring their continued relevance in diverse medical fields and facilitating their transition from the lab to clinical settings.

**3.2.6**
**Answering the question 1 from review of prof. D.Gorin section {Laser Microperforation Process} was added:**
A Cobolt Tor™ XS 532 nm pulsed laser (50 μJ; 1.9 ns) was employed to microperforate the biopolymer films. The laser light was directed onto the film surface using an 8 × 0.2 objective lens. Each microperforation was created with three sequential laser pulses, operating at a 1 kHz repetition rate. Film positioning under the laser

beam was achieved with a high-precision motorized XY stage (STANDA LTD, Vilnius, Lithuania), offering ±1 μm accuracy.

**4.3.1 Quantification of the vancomycin elution from PLACE films**

**Answering the question 2 from review of prof. D.Gorin next text was rewritten to improve clarity (also some slides will be supplemented to discuss it):**

The analysis highlights that the Peppas-Sahlin and Korsmeyer-Peppas models showed an exceptional fit, with respective values of Robs−pre=0.9893R_{obs-pre} = 0.9893Robs−pre=0.9893, R2=0.9787R^2 = 0.9787R2=0.9787 for Peppas-Sahlin and Robs−pre=0.9892R_{obs-pre} = 0.9892Robs−pre=0.9892, R2=0.9784R^2 = 0.9784R2=0.9784 for Korsmeyer-Peppas. While other models varied in their fit quality, the Higuchi model also showed a relatively good fit. However, it is important to note that the zero-order release model had notably poor goodness-of-fit values, suggesting it may not adequately represent the release dynamics of PLACE films.

The underperformance of the zero-order model implies that the drug release from PLACE films is not controlled solely by a constant-rate process. Instead, factors such as diffusion, polymer relaxation, swelling, and matrix erosion appear to play a substantial role, leading to deviations from ideal zero-order kinetics. The Higuchi model, while effective, assumes diffusion as the main release mechanism, which may limit its applicability in cases where other mechanisms are active.

In contrast, the Korsmeyer-Peppas model's adaptability enables it to account for multiple release mechanisms, including diffusion, swelling, and erosion, making it well-suited for capturing the complex release profiles of PLACE films. The diffusion exponent nnn derived from this model provides further insights into whether the release follows Fickian (diffusion-controlled) or non-Fickian (anomalous) kinetics, aiding in the identification of the predominant release mechanisms. Similarly, the Peppas-Sahlin model offers flexibility, allowing for a more nuanced analysis of release dynamics influenced by both diffusion and polymer relaxation mechanisms.

**Answering the question 3 from review of prof. D.Gorin error bars were added to the graphs where it was possible.**

**Minor typos, compiling and formatting errors have been corrected.**