Management Committee Meeting of COST Action CA16217 "European network of multidisciplinary research to improve the urinary stents" REPORT

Sofia, Bulgaria
31st January-1st-2nd February, 2019

Follow-up of MoU objectives: progress report of working groups. Scientific planning.-Scientific strategy (MoU objectives, GP Goals, WG tasks and deliverables).
WG1. D. Rako (Leader WG1). Duje commented that there have been delays in the tasks of this group, due to the fact that the leader of the Group has been changed three times. The tasks have been distributed in three main sections: ureteral metal stents (D. Rako); polymeric ureteral stents (F. Soria) and urethral stents (P. De Graaf). In this way the work is speeded up.
WG2. F. Clavica (Vice-leader WG2) was explaining the changes that WG2 members have made to the MoU to improve the objectives and make them more realistic. In addition to proposing a Workshop for September in Bern (Switzerland) within the activities of this WG2. New deadlines were also proposed for the scientific papers corresponding to this WG2. WG3. N. Buchholz (Vice-leader WG3), commented on the preparations being made to begin the work of WG3. Commenting on the need to request more support to bring together a multidisciplinary group due to the workload of this group. As in WG1, the objective to advance the tasks could be to distribute the items of the WG in different sub-groups.
WG4. A. Barros (leader WG4) presented the objectives of this WG. Currently this WG have more than 40 participants on the intranet. We are working on the joint creation of a database of biomaterials (WG4&WG5). During, February 2nd in Sofia, will be held the First Workshop of this WG. The need for support to coordinate the tasks by the Vice-leader of this group was commented on. The AC said that it would speak with the Vice-leader of this WG, due to the great task that corresponds to this WG.
WG5. G. Ciardelli (leader WG5) presented a brief review of the state of the art in this WG. Discussing the aims and milestones of this WG. The leader WG5 also proposed to hold a TS in conjunction with the WG4 due to the points they have in common in 2020. The need for support to coordinate the tasks by the Vice-leader of this group was commented on.
WG6. There was no presentation or comments in this WG, because the resignation of WG Leaer (Ingelin Claussen). Right now, we are not WG6 leader. As there was no presence of the leader or Vice-leader of this WG. None of the Vice-leaders considered it appropriate to present themselves as candidates. Mainly because it is considered that the profile should be of a urologist. The AC is committed to finding a candidate in the area of urology who can take over this leadership.

"Short Term Scientific Missions (STSM): review of completed reports and new applications"
AC. Coordinator D. Carugo (UK) the current status of applications for STSM. The estimation for this 2GP is 10 STSM. With a total budget of 23.500 €. To the current date, we have financed 8 STSMs. The average costs as you can see is less than 2000€/STSM. And we foresee that we are left with funds for 3-4 more STMS during this period. Very important remmeber, as you have to justify all the costs before April 31. Prof Valentina Cauda, Ms Elena Dragoni, Dr Petra de Graaf, Ms Marina Bandeira, Dr Joana Silva, Ms Maryam Mosayebi, Dr Zoran Markovic, Ms Sara Villarroya Castillo, Ms Federica Teodoro, Mr Lorenzo Lucherini, Ms Chaitra Venkatesh.

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Science Communication Manager. Update. N. Azevedo (Portugal) (Science Communication manager). presented the ENIUS website (www.enius.org). After many changes and misunderstandings. We have the website up and running. A "Participate" tab has been added. To include activities such as Training Schools, Workshops, etc. There are still many changes to be made, especially updating the names of leaders, and placing more updated information to attract more participants. Right now we are working to advertise the update, the new activities and the new MC members.

As part of the dissemination activities of our network during this 2GP. It is necessary to highlight those that we carried out during the National Congress of the Spanish Association of Urology, in the Courses that are carried out in my center of the European Association of Urology. A contribution, thanks to the invitation of Dr. Barros and Prof. Lima in the Symposium on Research & Innovations in Urology. Lisbon. And Prof. Kallidonis, during a lecture at the World Endourology Congress in Paris, also made an interesting diffusion of ENIUS. All these activities were at 0€ cost to the network. Since it was about invitations to participate with the expenses paid by the organizers of the events. AC encourage all MC Core Group members as far as possible to spread ENIUS.

We have had a very interesting request for DISSEMINATION MEETINGS to Prof. Missirlis from Greece. Prof. Missirlis has applied for Funds to attend the 4th international symposium on Nanoengineering for Mechanobiology (N4M) in Genova (Italy) next March 24-27, 2019. The MC and the Science Officer of the Action should approve. It was sent for a e-vote on 7 November (e-Voted). On 15 November his request was approved by the MC.

Implemention of Cost polices on: Promotion of Gender balance. Gender Balance Coordinator. Valentina Cauda from Italy.. Currently, only 34% of MC members are women and 27.3% of Core Group members. About Training activities (TS & Workshops); 42% women (Trainers) and 48% women (Trainees). **ECI**. AC showed a summary table of this GP2. There are 44 ECIs involved in ENIUS activities in this GP2. Regarding Trainers, we have 18% in Oxford; 44% in Caceres and on next Workshop we will have 13 ECI.

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SHORT TERM SCIENTIFIC MISSION (STSM) – SCIENTIFIC REPORT

The STSM applicant submits this report for approval to the STSM coordinator

Action number: CA16217 – European network of multidisciplinary research to improve the urinary stents
STSM title: Advanced nanomaterial coatings for antimicrobial drug eluting urinary stents
STSM start and end date: 02/05/2018 to 16/05/2018
Grantee name: Chaitra Venkatesh

PURPOSE OF THE STSM/

(max.500 words)

The STSM addresses scientific object 3 of the ENIUS Action, which aims to assess the "opportunities for improved stents related to the evaluation of new biomaterials, nanotechnology applications, new coating, drug-eluting stents and biodegradable stent materials". This STSM will contribute to addressing the prime objective of the ENIUS COST Action by forging links between different multidisciplinary groups and exchanging expertise towards identifying and assessing opportunities to improve urinary stent design. This is a collaborative study between the Politecnico di Torino (Italy) and the Athlone Institute of Technology (AIT) (Ireland). This work will be carried out in conjunction with Teleflex, a global provider of urology products and urinary stents. Urinary stents are one of the most commonly used medical devices, however there is a continued struggle to control associated bacterial infections. Modern concerns about antibiotic resistance and the evolution of numerous new resistant bacterial strains accentuate this problem. The potential for urinary stents to provide controlled drug release offers an important medical advance both in the management of microbial challenges and in the treatment of medical conditions and disease of the urinary system.

The aim of this STSM is to contribute to the development of innovative antibacterial and drug-eluting stent coatings using porous zinc oxide (ZnO) nanomaterials developed by Politecnico di Torino. This ZnO has documented strong antimicrobial properties and networks of nanopores which present ideal characteristics for drug-eluting functions. The home and host institutions in collaboration together, have deposited this functional coating using novel approaches on a polymer formulation used by Teleflex in the fabrication of their ureteral stents and the performance parameters will be characterised and optimised.

The application of nano-structured materials with drug-eluting and antimicrobial features for their use in

future urinary stent coatings will be evaluated. Furthermore, the STSM targeted the capacity-building objectives 7, 8 and 9, dedicated to training young researchers, stimulating the rise and growth of novel scientific idea in the field, and linking the scientific research with an industrial company.

The Action aims at formulating biocompatible, antibacterial and drug-eluting coatings for novel ureteral stents fabrication. This is achieved by coating a novel formulation of polymeric ureteral stents with the innovative porous ZnO thin films. Porous ZnO is prepared under mild conditions according to synthetic process developed at Politecnico di Torino, and is therefore compatible with a wide range of substrates including polymers. In this STSM, the deposition of porous ZnO coatings on flat polymeric substrates was performed. The coated substrates were fully analysed and compared with respect to the raw polymer substrates, by using various characterization techniques. The morphology and chemical composition were investigated by Field Emission Scanning Electron Microscopy (FESEM) and Energy Dispersive Spectroscopy (EDS), respectively. The structural features of the materials were assessed by X-ray Diffraction (XRD), while Optical Contact Angle (OCA) measurements were performed to investigate the surface wetting properties.

DESCRIPTION OF WORK CARRIED OUT DURING THE STSM

Polyethylene, Polypropylene and Pellathane with 54% Barium sulphate and 10% isoplast are the three different polymer substrates prepared at Athlone Institute of Technology. Polyurethane with 35% Barium Sulphate and 10% isoplast was procured from Teleflex. For comparison, Polyethylene and Polypropylene from Athlone Institute of Technology were also prepared as films. The three polymers were processed by the method of extrusion at 180 °C and were then drawn into films.

During the STSM, porous zinc oxide coatings (ZnO) were prepared according to a specific synthesis process developed by Politecnico Di Torino. Highly porous, flower-like ZnO powders were first prepared by a sol-gel method and then mixed with different solvents (acetic acid, water and ethanol) to get a ZnO-based paste. Two different types of paste were prepared: one solution with ethanol and the other with zinc acetate dehydrate in ethanol. All three polymers were coated with solution one and checked for adhesion. Similarly, the polymers were coated with solution two. Parameters such as temperatures were modified depending on the nature of the polymer for adhesion. However, we did not observe any improvement in the adhesion of the ZnO films by using solution two. Hence, the experiments were continued using only solution one.

The surface wettability properties were investigated using optical contact angle measurements by Goniometry. All the polymers showed a hydrophobic behaviour. In order to observe if there was any improvement in the adhesion of the ZnO coating due to an improved hydrophilic behaviour, the polymers were plasma treated before coating for 1 minute in Ar gas. This allowed to change the polymer wettability from hydrophobic to hydrophilic. However, there was no strong improvement in the adhesion. Scanning electron microscopy was conducted both on the polymers with ZnO coating and without coating for

morphological analysis.

Two new batches of ZnO powders were synthesised by sol-gel method. Some polymers were plasma treated before the deposition of the paste. The paste was coated on the polymer as above by treating at 70 °C for 30 minutes. In order to have better adhesion, the polymers were also coated by the method of hot embossing with the help of silicon and glass plates. In this method, the silicon was first coated with ZnO paste and treated at 70 °C for 30 minutes. The polymer was then placed atop the coated silicon and a weight was placed on the top of the sample, for good coat transfer. All this was performed on a hot plate at 90 °C for 10 minutes.

Once the paste deposition method by hot embossing for good adhesion was optimized, the polymer samples with ZnO coating and without coating by similar method were prepared for mechanical and thermal characterisations. These mechanical and thermal characterisation will be carried out in Athlone Institute of Technology.

X-ray Diffraction was conducted on the sample of pristine polypropylene, polypropylene ZnO-coated samples, and, as a reference, polypropylene treated sample in absence of ZnO to analyse the structural features. Scanning electron microscopy was conducted on the coated polymers and on the coated silicon for the morphology and chemical compositions.

DESCRIPTION OF THE MAIN RESULTS OBTAINED

The processing of the polymers by the method of twin screw extrusion and drawing them into films was done successfully at the home institute Athlone Institute of Technology. During the STSM at Politecnico di Torino, the zinc oxide (ZnO) coating was prepared successfully according to synthesis process developed by Politecnico di Torino. The ZnO paste deposition on the polyethylene and pellathane did not have good adhesion. However, the results of paste deposition on polypropylene resulted into good ZnO coatings by the method of hot embossing.

The scanning electron microscopy images of polypropylene showed that polymer melt and the ZnO coating was partially embedded inside the polymer. The X-Ray Diffraction showed the peaks of the treated polypropylene had some more peaks when compared to the untreated polypropylene. The coated polypropylene showed diffraction peaks belonging to wurtzite ZnO.

Many samples of polypropylene coated with ZnO were prepared successfully by hot embossing for further characterisations. Moreover, mechanical and thermal characterisations will be carried out on the samples at Athlone Institute of Technology.

From completion of STSM, the STSM applicant received training on solution preparations and coating techniques. The knowledge obtained will aid in the collaboration of work between Athlone Institute of

Technology and the Politecnico di Torino.

FUTURE COLLABORATIONS (if applicable)

(max.500 words)

The work produced during the STSM will form part of a joint publication by Politecnico di Torino and Athlone Institute of Technology.

One of the main focuses of this STSM was to establish a new connection with Politecnico di Torino and Athone Institute of Technology. Through this newfound connection, there is an ongoing collaboration between the two institutes.

Athlone Institute of Technology and the Politecnico di Torino are interested in future STSM student exchanges and are actively looking for mechanisms to fund collaborative projects. One of such STSM applicant from Politecnico di Torino will be visiting AIT to work on urinary stents.

SHORT TERM SCIENTIFIC MISSION (STSM) – SCIENTIFIC REPORT

Action number: CA16217
STSM title: Urinary stents made of biodegradable and drug-eluting organic/inorganic composite.

STSM start and end date: 29/03/2019 to 21/04/2019
Grantee name: Elena Dragoni

PURPOSE OF THE STSM

The main purpose of this STSM was to develop the method for the synthesis of innovative antibacterial and drug-eluting stent coatings using porous zinc oxide (ZnO) nanomaterials developed by Politecnico di Torino (in a flower-like morphology) and linear polymers obtained in Maria Curie Sklodowska University in Lublin. The antibacterial properties of ZnO and its regular porous structure make possible to use this material as a drug delivery carrier. The aim of the first month of the STSM was to prepare linear and slightly crosslinked polymers of 2-hydroxyethyl methacrylate (HEMA) and its copolymers with acrylic acids and glycidyl methacrylate which gives the broad possibility of the polymer chemical post-modification and also fully characterize them with the following techniques: Thermogravimetry and Differential Scanning Calorimetry (TGA and DSC), fourier-transform infraRed Spectroscopy (FT-IR). Materials were prepared using different percentages of ZnO powders.

The completed STSM is strongly aligned with the key aims of the ENIUS Action.

DESCRIPTION OF WORK CARRIED OUT DURING THE STSM

I started my STSM learning the process to synthesize polyHEMA and then I run some trials to find and improve the correct method to process composites of polyHEMA with the desired amount of ZnO.
To synthesize this polymer, I used the radical polymerization method.

The first used method of polymerization was dispersion polymerization. The reagents used for the synthesis of the linear polymer were:

- 5g of 2-hydroxyethyl methacrylate (HEMA), liquid reagent.
- 2% (0,1g) of initiator 2,2-methylpropionitrile (AIBN) 98%, solid reagent.

I mixed together the two reagents and then I added 50 mL of toluene as solvent to create the solution.

Before starting the polymerization, it is better to put the solution under a flow of nitrogen, in order to remove the oxygen, which is an inhibitor of the polymerization reaction.

To start the radical polymerization, the initiator have to be activated. Increasing the temperature, the double bonds present in the structure of AIBN gets broken in an homolytic way and two activated radicals are created.

In order to induce this homolytic breakup and carried out the polymerization, the solution has been put under magnetic stirring at the stable temperature of 70°C for about 6 hours. After the polymerization, the solvent was evaporated under reduced pressure and the polyHEMA was obtained as a white powder.

Then I tried to find the appropriate method to prepare the composites of obtained polymer and the powder of zinc oxide. The carried out test containing the melting of polymer and the method with the use of dissolved polymer did not give a composites with desired properties.

Another approach to the preparation of polymer-ZnO composites was the direct bulk polymerization of HEMA monomer with ZnO. I started to prepare the composites of polyHEMA and ZnO. The first method consisted in filling the rectangular moulds (3x2x0.3cm between two plates of glass) with the solution made by the liquid monomer, the initiator and powders of ZnO, and put them into the oven at the stable temperature of 70°C for 5 hours.

I met different problems using this approach; the most relevant consisted in the difficulty to remove the sample of composite from the glass plates, maybe because during the reaction of polymerization, OH groups of the monomer intereacted with OH groups of the glass, making them strictly bound to glass parts of the mould. Nevertheless, two positive things can be obtained using this method; the first is that the composites appeared to have 2 layers because of the sedimentation of the powders at the beginning of the polymerization, when the reagents are still liquid and the second one, that putting the sample in water, it absorbs water thank to the swelling propriety of polyHEMA.

Another problem was caused by the excessive polymerization that made the sample to have cracks on the surface, in particular on the side with polyHEMA only.

The second method I used, the one that later I chose to prepare 3 classes of materials, consisted in the pre-polymerization before the preparation of the forms, in order to fill the forms with a viscous solution and to avoid an aggressive and excessive polymerization into the oven. Considering that the reaction started

before, the can stay into the oven only for 4 hours, at the same temperature of 70°C.

With this method, ZnO is well dispersed inside the polymer and the final composite appears really homogeneous, but the sample it could be removed easier from the plates, and moreover the surface does not show cracks.

After the preparation of the composites, they have been characterized through TGA and FTIR analysis.

DESCRIPTION OF THE MAIN RESULTS OBTAINED

Concerning the FT-IR analysis made to the polyHEMA, following are shown the results of the polymerization.

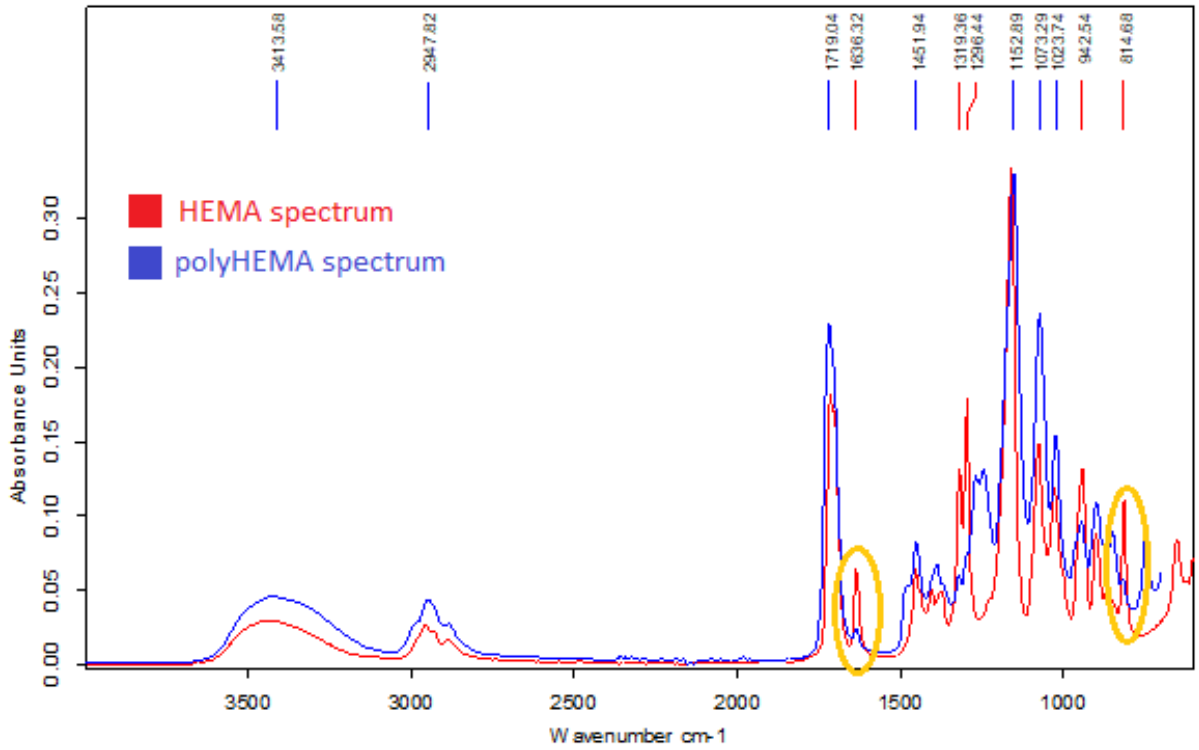


Figure 1 FT-IR analysis of polyHEMA compared to that of HEMA monomer.

Concerning the composites, firstly, to run some trials of the two methods of synthesis, I prepared composites with different concentrations of ZnO, and of each was made the characterization by TGA.

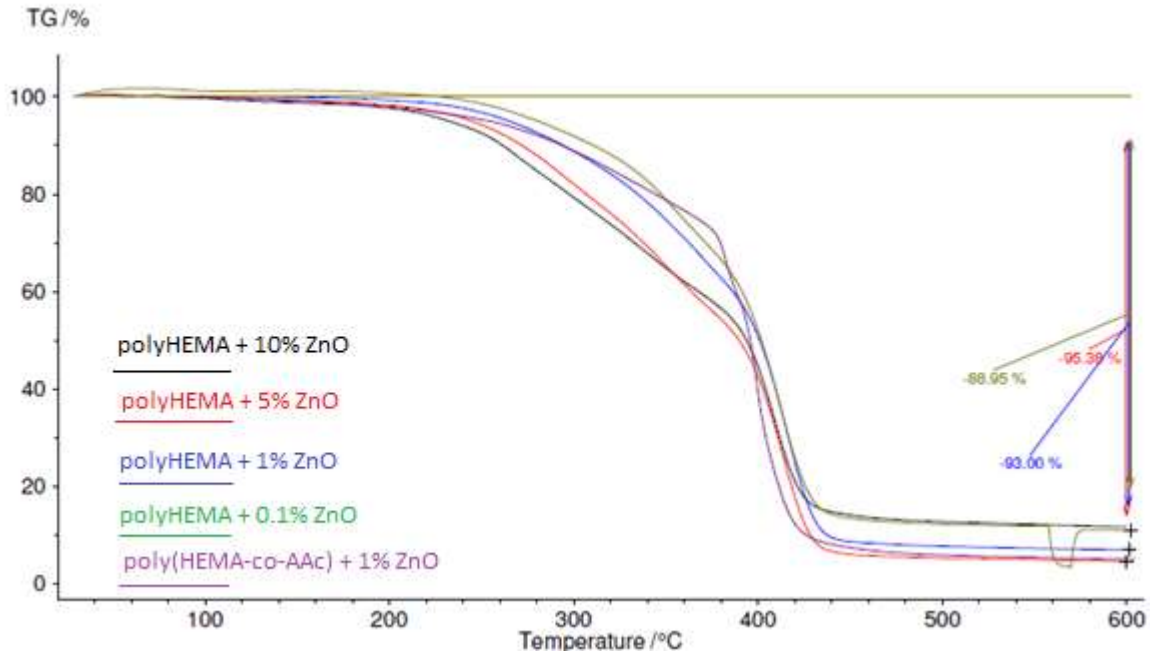


Figure 2 TGA analysis made the first samples of composites.

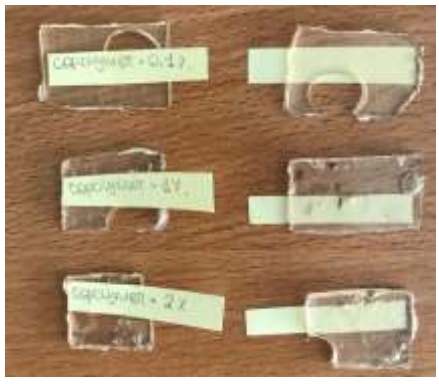
Once I decided to apply the second method of synthesis, I prepare the following classes of composites, each with different concentrations of ZnO:

Group 1 - Polymer

- PolyHEMA (4mL) + 0.1 % ZnO.
- PolyHEMA + 1 % ZnO.
- PolyHEMA + 2 % ZnO.

Group 2 - Copolymer

- Poly(HEMA-co-AAc) + 0.1 % ZnO.
- Poly(HEMA-co-AAc) + 1 % ZnO.
- Poly(HEMA-co-AAc) + 2 % ZnO.



Group 3 – Copolymer crosslinked

- Poly(HEMA-co-AAc) + 5% crosslinker (Ethylene Glycol Dimethacrylate) + 0.1 % ZnO.
- Poly(HEMA-co-AAc) + 5% crosslinker (Ethylene Glycol Dimethacrylate) + 1 % ZnO.
- Poly(HEMA-co-AAc) + 5% crosslinker (Ethylene Glycol Dimethacrylate) + 2 % Zn

The only difference between the two spectra is noticed over the characteristic wavenumber of the carbons double bond, where the HEMA spectrum show a higher peak. This result proves that the polymerization occurred correctly.

Concerning the analysis made to the different composites prepared, the FT-IR characterization was made to compare each other the samples belonging to the same group, having different concentration of ZnO.

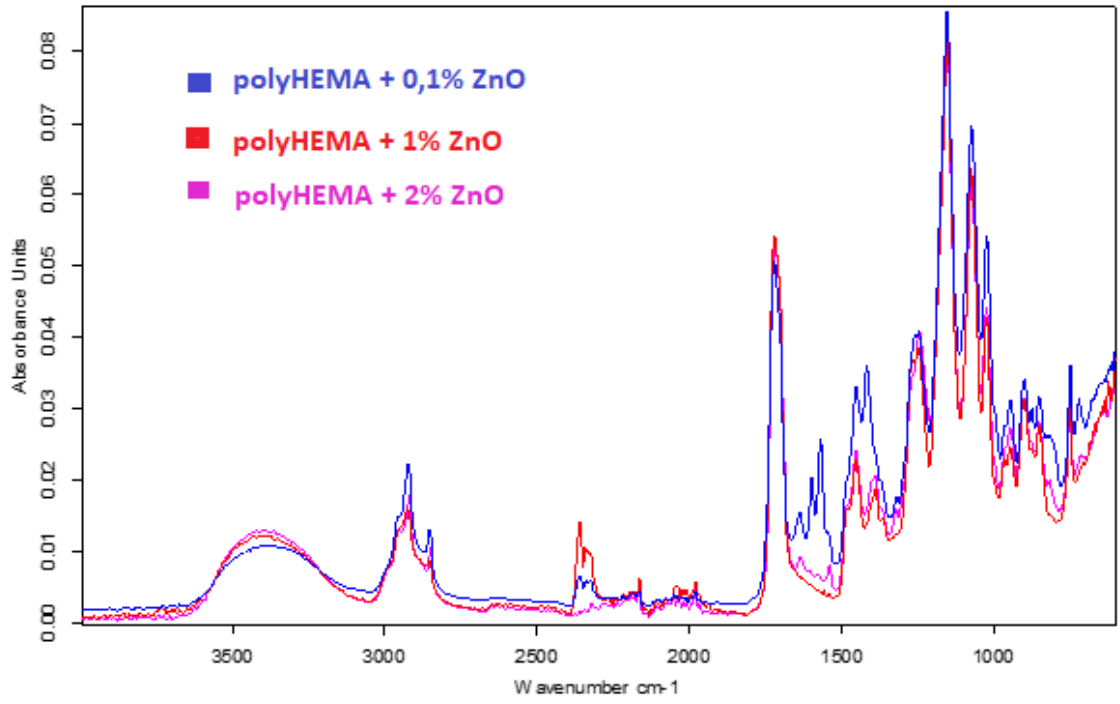


Figure 3 Comparison of FT-IR spectra of polyHEMA's samples with different percentages of ZnO.

The spectra of the other 2 groups of materials (copolymer and copolymer with crosslinker) are similar because the structure of the polymers is almost the same.

About the TGA analysis that were made, the curves of the three types of composites with 0.1% of ZnO are shown below.

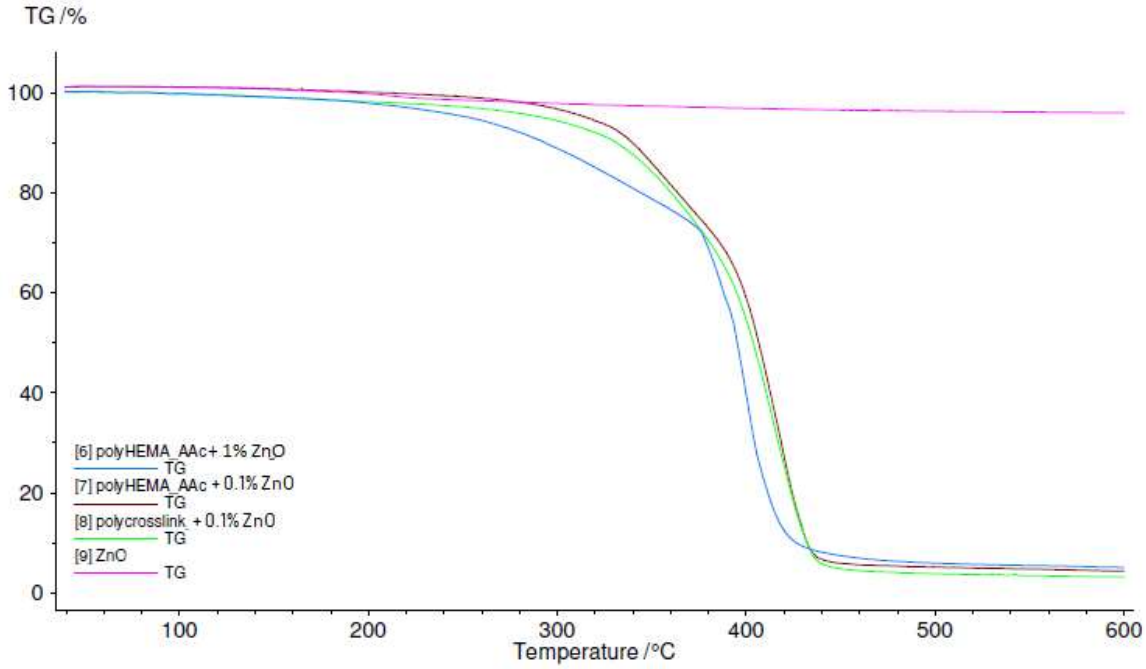


Figure 4 TGA analysis 0.1% ZnO

FUTURE COLLABORATIONS

In the next STSM it is planned to prepare the composites of Group 1 and Group 2 using the optimized methods of bulk polymerization , which were previously mentioned. Then, I will charge each sample with Ibuprofen by absorption to produce a drug delivery composite and study the application as drug eluting stent.

The same procedure of synthesis will be repeated in order to absorb another drug, which is the Diclofenac Sodium Salt.

After the uptake of drugs, in Lublin I will do the characterization of the samples with TGA analysis and then I will take them to Turin to analyse the release of drugs and then the degradation of the materials.

SHORT TERM SCIENTIFIC MISSION (STSM)
SCIENTIFIC REPORT

This report is submitted for approval by the STSM applicant to the STSM coordinator

Action number: CA16217
STSM title: Future research lines in urinary stents: how to improve vascularization of a cellular urethral graft
STSM start and end date: 18/03/2019 to 22/03/2019
Grantee name: Petra de Graaf

PURPOSE OF THE STSM:

At the University of Oulu, in Dr. Skovorokdin group tools are developed to study the vasculature in kidneys during development. These tools can help performing future steps in the research of Dr. De Graaf. We can use the Chick Chorioallantoic Membrane (CAM) assay to test our current and future hydrogels. Angiogenic properties of different compositions can be quantified. In addition, the CAM assay can answer the question whether networks formed in the hydrogel will connect to the chorioallantoic membrane vessels. Lastly, if we can detect chick blood in the 3D structures in the hydrogel we can confirm the perfusability of the formed networks. Furthermore Dr. Skovorokdin has developed time lapse technologies to follow kidney development in 4D (3D in time), which may be adjusted for applications in the urethral tissue engineering project.

Personal milestones are:
1. Get familiar with the CAM assays.
2. Learn more about the possibilities of 4D imaging to use this technology to follow the 3D network formation in hydrogels.
3. Learn from angiogenesis during development to optimize vascular network formation in the tissue engineered graft
4. Discuss potential next steps in the collaboration, including position papers, collaborative experiments and consortium building for future grant applications, either in the European H2020 program but also outside this program.

DESCRIPTION OF WORK CARRIED OUT DURING THE STSMS

During the STSM Dr. Skovorokdin showed chicken embryo intercardial injection of labeled human endothelial cells, which we followed during the stay for 3 days. The labeled cells were incorporated in the extra-embryonic vascular network (M1). We microscopically analyzed mouse embryonic explanted organs (kidney and heart) in costum made bioreactors to see if the outgrowing endothelial cells will connect to the human endothelial cells (HUVECs) in the chamber surrounding the explanted organ (M3). At the moment this connection is not always established, and we discussed this. It is unclear whether the problem with is the origin (umbilical cord) or the species difference. In the Regenerative Medicine Center Utrecht (RMCU) endothelial

cells from different origin are characterized and analyzed, which lead to new potential leads in research. Petra de Graaf was introduced to the 4D imaging microscopes (M2), this is highly suitable for 4D analysis of the hydrogels, however, this STSM was too short to set up a hydrogel with vascular network, so this will be for future collaboration.
Lastly, Petra de Graaf presented her work to the research group and was introduced to the stem cell core facility and to several coworkers including Professor Seppo Vainio, Prateek Singh, Laura Dönges and Dr. Irina Raykhel (M4). The fruitfull discussions on endothelial networks, endothelial antibody specificity, the use EVO-DEVO for organogenesis of the urethra and ureter, biofabrication for organ engineering and advanced imaging. This will help us to further develop our Enius WG6 (Future developments).

DESCRIPTION OF THE MAIN RESULTS OBTAINED

Over the course of 3 days we could detect labeled human endothelial cells in the extra-embryonic vascular network of the chicken embryo. 6 chicken embryo's were injected, of which 4 survived. Life embryo analysis showed incorporation of the human cells in a perfused network. Embryo's including extra-embryonic vascular network were fixed and stained after I left with both human and chicken specific endothelial markers to analyse the junctions.
The most important result I obtained was being able to gain hands on experience with the chicken embryo's, join the discussions in the lab and being introduced to the coworkers of Dr. Ilya Skovorokdin, especially Professor Seppo Vainio and Dr. Irina Raykhel.

FUTURE COLLABORATIONS (if applicable)

We agreed to exchange knowledge and expertise in the future. After the visit in Oulu a postdoc form Dr. Skovorokdin's lab, Dr. Irina Raykhel, visited the RMCU center in Utrecht.
Petra de Graaf will send hydrogels and scaffolds to test in the CAM assay, when this analysis is finished we will prepare a manuscript based on these results and (try and) co-publish together.
Petra de Graaf is planning writing a MSCA-ITN application for a training network (H20202 framework, deadline January 2020), where the University of Oulu will be a member of the network, as well as a spin off of the University of Oulu (FinnAdvance in person of Prateek Singh, biofab solutions to biomedical problems). Other collaborative granting opportunities are monitored, we will search more active after the summer due to running projects and obligations.

SHORT TERM SCIENTIFIC MISSION (STSM)
SCIENTIFIC REPORT

This report is submitted for approval by the STSM applicant to the STSM coordinator

Action number: 16217
STSM title: Physico-chemical characterization of gamma irradiated polyurethane composites with carbon quantum dots
STSM start and end date: 11/02/2019 to 08/03/2019
Grantee name: Zoran Markovic

PURPOSE OF THE STSM:

(max.200 words)
Design of new types of antibacterial surfaces is of great importance due to increased formation of antibiotic-resistant bacteria strains on many surfaces both in healthcare institutions and industries (i.e. food industry or pharmaceutical industry). Formation of these surfaces is a very big challenge because many parameters such as surface roughness, wettability, surface resistance, surface morphology or surface charge can affect more or less bacteria adhesion or later bacteria eradication effectively. Apart from surface properties, size and shape as well as charge of bacteria strains play a very significant role in bacteria adhesion to various surfaces. One of the possible strategies to reduce or prevent bacterial infections is to synthesize photoactive coatings or nanocomposites. Different photosensitizers produce reactive oxygen species (ROS) and eliminate bacteria more or less efficiently
In the past two years our focus was development of antibacterial composites based on polyurethane and polydimethylsiloxane (PDMS). In the planned study, we performed characterization of the gamma assisted modification of polyurethane based nanocomposites doped by hydrophobic carbon quantum dots (hCQD-PU). We examined the effect of different parameters, such as the dose of gamma irradiation applied to the nanocomposite (1, 10 and 200 kGy) on chemical and morphological properties.

DESCRIPTION OF WORK CARRIED OUT DURING THE STSMS

(max.500 words)

Transparent polyurethane (PU) was dipped in the solution of carbon quantum dots (CQD) with a concentration of 1 mg/ml. The encapsulation of dots into the PU was done by the swelling method at ambient temperature during 48 h. Samples were then exposed to the source of gamma rays at the doses of 1,10 and 200 kGy.
During STMS, we have performed following characterizations:
1.Fourier transformed infrared (FTIR) spectroscopy and X-photoelectron spectroscopy
2.The contact angle measurements of the gamma-irradiated hCQD-PU nanocomposites
3.Surface morphology of the gamma-irradiated hCQD-PU nanocomposites was recorded by atomic force microscope – AFM.
4.Photoluminescence (PL) measurements of the gamma-irradiated hCQD-PU nanocomposites was measured at excitation wavelengths between 320 and 480 nm.
5. Electron paramagnetic spectroscopy (EPR) was used to determine the singlet oxygen and hidroxyl radicals generation of different hCQD-PU nanocomposites
6. Photoluminescence (PL) measurements was used for determination of hidroxyl radicals generation of

different hCQD-PU nanocomposites

DESCRIPTION OF THE MAIN RESULTS OBTAINED

We have obtained following results

1. FTIR spectroscopy was used to assess the presence of different functional groups. In the FTIR spectra of all the observed gamma irradiated hCQD-PU nanocomposites we detected O–H vibration mode, mode of C-H stretching vibrations, modes of C=O bonds, C=C bonds C-O-C and C–O bonds. XPS revealed tha the percentage of sp^2 bonds and sp^3 bonds increased (decreased) with the increase of the dose of gamma irradiation.
2.Contact angle was decreasing with the increase of the dose of gamma irradiation. The contact angle decreased from 96.5° (hCQD-PU-1kGy) to 72.6° (hCQD-PU-200kGy).
3.AFM was used to analyze the morphology and surface roughness of samples irradiated with different doses of gamma irradiation. From AFM images we can easily observe the difference in the surface morphology of different samples. The surface roughness significantly increased with the increase of the dose of gamma irradiation. Surface rougness was in the range from 20.9 to 61.7 degrees.
4.The highest intensities of emission bands were detected at the excitation wavelength of 360 nm for both hCQD-PU-1kGy and hCQD-PU-10kGy samples, while for the hCQD-PU-200kGy sample the highest intensity was measured at 400 nm excitation wavelength. All samples have broad green emission luminescence centered between 432 and 538 nm, for different excitation wavelength.
5. The samples of gamma-irradiated hCQD-PU nanocomposites were recorded with EPR in the presence of 2,2,6,6-tetramethylpiperidine (TEMP) which is used as a spin trap agent. The molecules of TEMP selectively react with singlet oxygen ($^1O_2$) and form the stable product 2,2,6,6-tetramethylpiperidine-1-oxyl (TEMPO), which shows the characteristic signal. The EPR spectra of TEMP in the dark does not show any signal. From the spectra, we concluded that the sample hCQD-PU-10kGy has the highest production of singlet oxygen, almost five times stronger than the other two samples. The samples hCQD-PU-200kGy and hCQD-PU-1kGy are generating less singlet oxygen.
6.Determination of hidroxyl radicals using BMPO spin trap and EPR was unsuccesful. Therefore for the determination of production of hidroxyl radicals we decided to use photoluminescent method. The analysis of •OH radical's formation on the samples under visible light irradiation was performed by fluorescence technique with using terephthalic acid, which readily reacted with •OH radicals to produce highly fluorescent product, 2-hydroxyterephthalic acid. We have discovered that sample hCQD-PU-200kGy produces largest amount of hydroxyl radicals.

FUTURE COLLABORATIONS (if applicable)

I am very grateful to my host Dr Zdeno Spitalsky and his colleagues for their warm hospitality and to COST Action 16217 for giving me this opportunity to visit Polymer Institute of Slovak academy of Sciences in Bratislava, Slovakia. This STSM was highly beneficial as it enabled efficient knowledge transfer and it is my sincere hope that this visit will further strengthen bilateral collaboration.
What remains is to continue with a joint work on the optimization parameters of produced polymer nanocomposites doped with modified carbon quantum dots with a view to using them as a material for urinary stents. It is planned to present the obtained results in a joint paper prepared for a peer-reviewed journal and through conference paper(s). This STSM experience as well as the obtained results and planned future work could also serve as a a starting point for future research project proposals involving Polymer Institute, Slovak Academy of Sciences in Bratislava, Slovakia and Vinča Institute of Nuclear Science, University in Belgrade, Serbia.

SHORT TERM SCIENTIFIC MISSION (STSM)
SCIENTIFIC REPORT

This report is submitted for approval by the STSM applicant to the STSM coordinator

Action number: CA16217
STSM title: Stent-on-a-chip:studying encrustation of ureteric stents using microfluidics
STSM start and end date: 01/11/2018 to 29/01/2019
Grantee name: Dario Carugo

PURPOSE OF THE STSM:

Ureters are conduits that transport urine from kidney into the bladder; in healthy individuals the propagation of urine is driven by muscular contractions of the ureter. However, ureteric obstructions may occur under certain clinical conditions, including occlusions generated by kidney stones or external compression (i.e., due to a tumour). High levels of ureteric obstruction are normally associated with acute pain and severe increase in the renal pressure. In this clinical situations, ureteric stents are generally inserted into the ureter to restore urine drainage. They consist of flexible tubes containing multiple side holes (fig. 1) allowing for urine to bypass the occlusion.

Figure 1: Ureteric stent (left), detail of the side holes (right)

Despite the extensive clinical experience, complications related to stenting are still frequent, with significant impact on a patient's quality of life, efficacy of treatment, and cost of patient care. Formation of encrustation over the stent surface is regarded as one major determinant of stent failure. However, a clear understanding of the physical parameters responsible for encrustation has not been achieved yet. In the present study, we employed a microfluidic-based model of the stented ureter to reveal the formation of

encrusting deposits over a ureteric stent surface. We specifically focused on the effect that the flow dynamics (and specifically wall shear stress) has on the initiation and progression of encrustation.
Computational simulations were performed to investigate the effect of ureteric obstructions on the flow dynamics within the stented ureter.

DESCRIPTION OF WORK CARRIED OUT DURING THE STSMS

1. DESIGN

The model presented in this study is a simplified representation of the stented and occluded ureter, allowing for high-throughput screening of different stent designs or clinical conditions.
In order to replicate different modalities of ureteric obstruction, five different designs were generated in this study:

- Unobstructed and stented ureter; i.e. control design (design 1)
- Partial obstruction placed between side holes (design 2)
- Total obstruction placed between side holes (design 3)
- Partial obstruction placed on a side hole (design 4)
- Total obstruction placed on a side hole (design 5)

The design was carried out with software Autodesk® Inventor® Professional 2019 (academic version) (Autodesk Inventor Pro).
A 2D sketch (fig. 2.a) was initially generated, which was subsequently extruded in order to obtain a 3D structure (fig. 2.b).

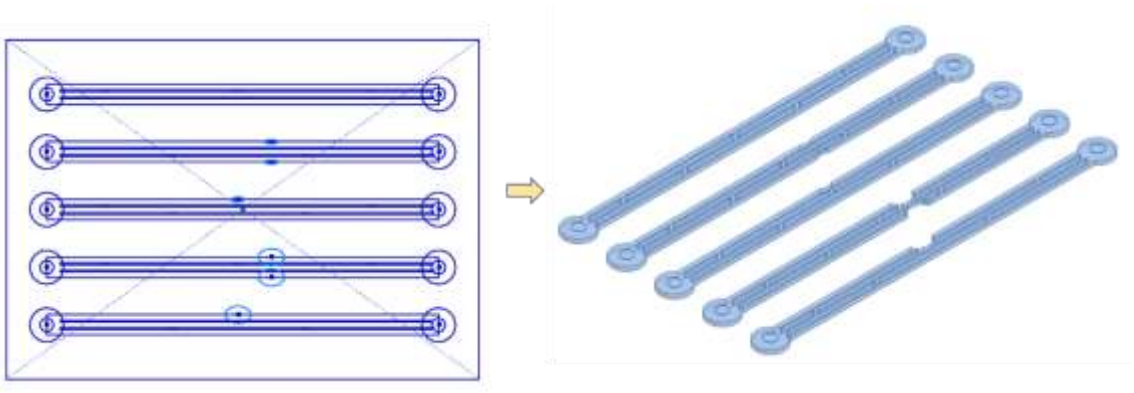


Figure 2: a: 2D sketch (top view) of the device, b: 3D representation of the different designs investigated

2. SIMULATIONS

Numerical simulations were set up using ANSYS® Workbench Software. The program sets up the simulation in different stages (fig. 3): importing the geometry, meshing, simulating the flow field, post-processing.

- The geometry can be created in ANSYS® Design Modeler or imported in the appropriate format file from a Computer Aided Design (CAD) software.
  In this work, the geometry was created in Autodesk® Inventor® Professional 2019 and imported in ANSYS® Workbench 19.1.

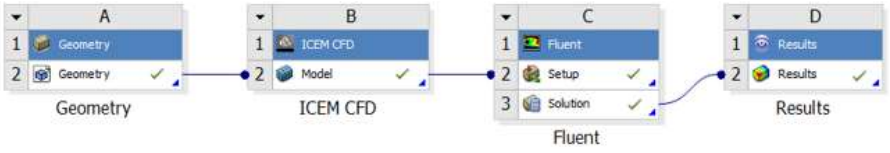


Figure 3: Outline of the simulation setting up process

- Material: water liquid
- Boundary conditions (fig. 4)
  - Inlets: volumetric flow rate of 1mL/min normal to the boundary
  - Outlet: outflow weighting equal to 1
  - All other surfaces: stationary wall

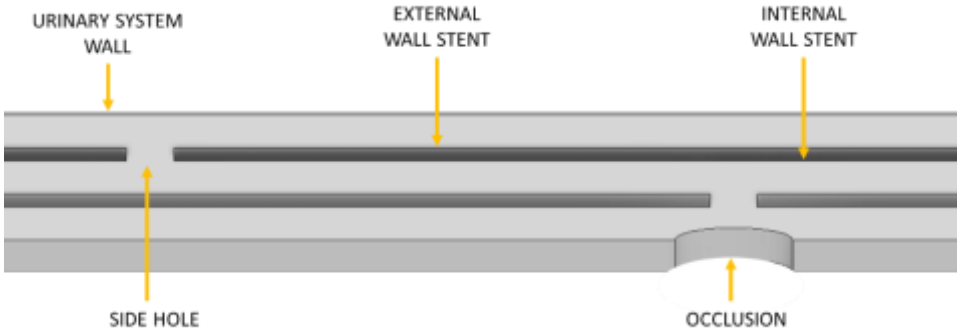


Figure 4:Boundary conditions

- Run and compute the solution

In an initial phase of the project, it was necessary to identify an optimal mesh element size that represented the best compromise between solution accuracy and computational time. This was achieved through a 'mesh dependence' study, where five different mesh element sizes were tested: 1 mm, 0.5 mm, 0.3 mm, 0.1 mm and 0.05 mm.

To analyse the data and find the best mesh size, data were imported into ANSYS® CFD-post. The velocity magnitude was calculated along a specific line. Successively, the velocity was plotted (fig. 5).

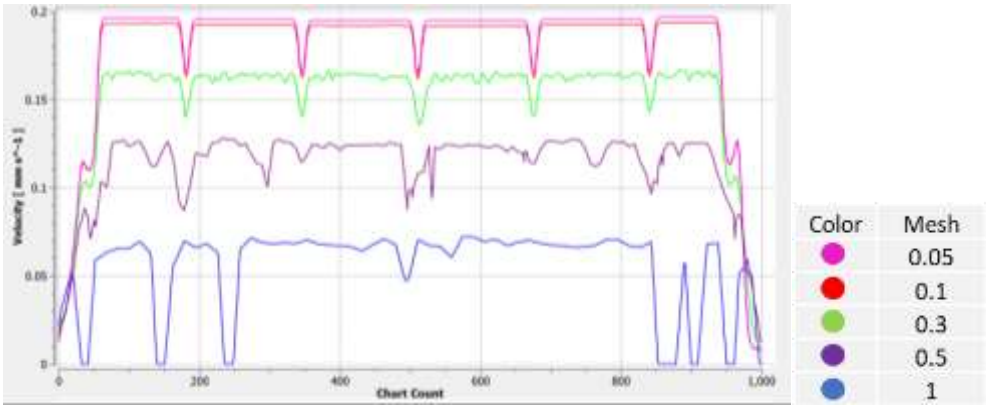


Figure 5: Results of the mesh dependence study

The mesh dependence study could also be performed by evaluating different parameters at different position.

The simulations with a mesh size of 1 mm, 0.5 mm and 0.3 mm presented fluctuations in the velocity magnitude profile. These significantly reduced in the mesh sizes of 0.1 mm and 0.05 mm. These two simulations led to very similar values but required a very different computational time. If the first simulation (mesh size 0.1 mm) required 30 minutes, the second simulation (mesh size 0.05) required 2 hours.

For this reason, in reduce computational time and still obtain a very accurate solution, it was chosen to use the mesh of 0.1 mm. It is able to mesh all the different model designs (fig. 6) with a good result, as the finest mesh of 0.05 mm.

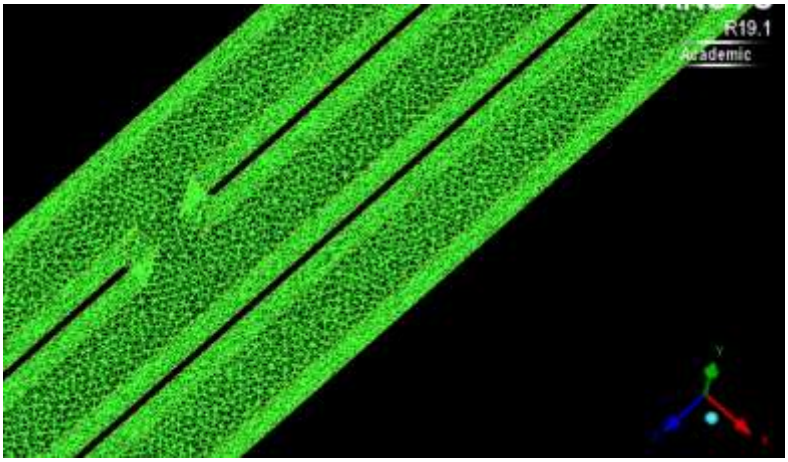


Figure 6: Mesh elements

DESCRIPTION OF THE MAIN RESULTS OBTAINED

Earlier experimental and theoretical studies have postulated that there is a correlation between WSS and deposition of encrusting particles over the stent surface [1]. They defined “inactive” side holes of the stent the ones with a low mean WSS (0.020 Pa). On the contrary side holes with a mean WSS ≥ 0.020 Pa will be considered as “active” side holes. The same threshold value was used in this work.

Figures 7 and 8 show the WSS over the bottom surface of the 5 designs; WSS values ranged between approximately 0.000 Pa and 0.430 Pa.

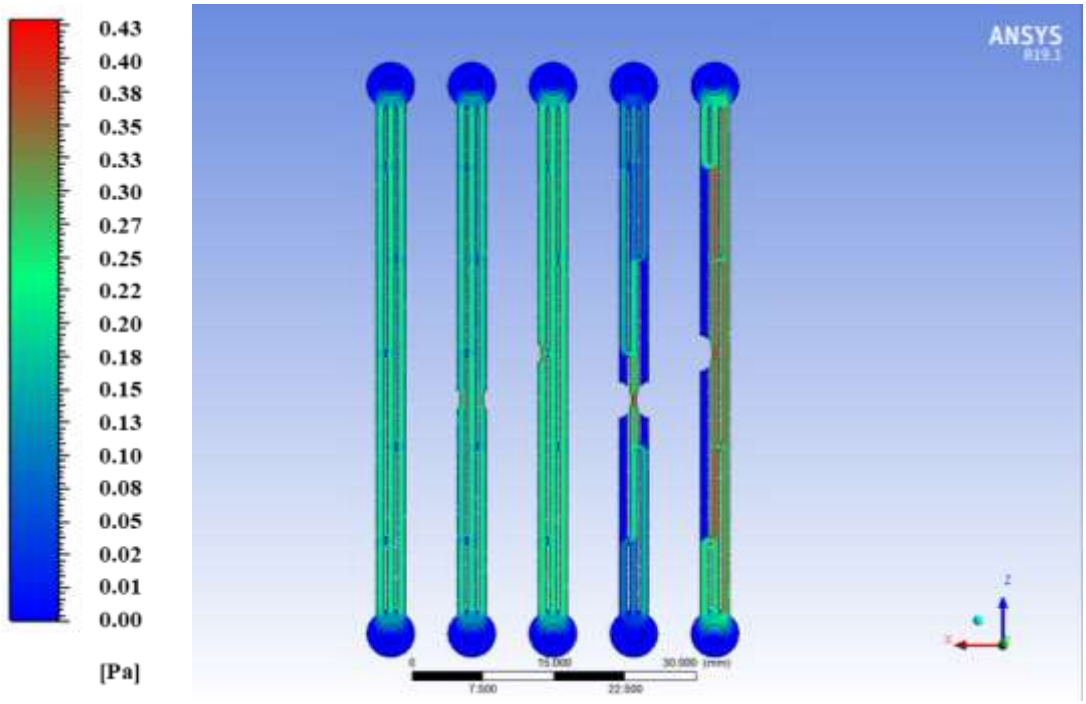


Figure 7: WSS over the bottom surface of the models. From left to right: the control, the partial occlusion in between side holes, the partial occlusion on the side hole, the total occlusion in between side holes, and the total occlusion on the side hole

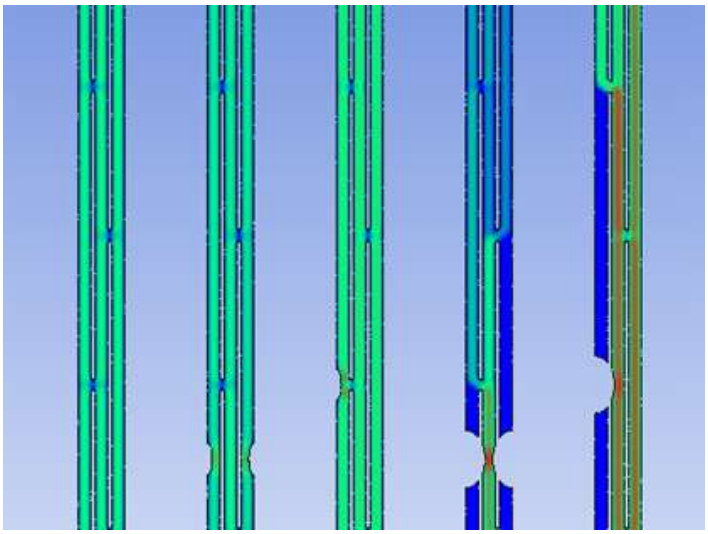


Figure 8: Zoomed-in view of the WSS contours for the different models tested

The side holes in the design 1 have a WSS of 0.010 Pa, due the absence of flow exchange between the ureter lumen and the stent; i.e. all side holes were thus 'inactive'.

A similar behaviour can be noticed in designs 2 and 3, with partial occlusion. The design 2 and 3, which differ only from the position of the occlusion, had very comparable levels of WSS.

They do not show activation of any side hole, but only an increase of WSS between the partial occlusion and the inner wall of the stent (to a value of 0.015 Pa).

An increase in WSS and a different flow distribution occurs in designs 4 and 5. The total occlusion forces an exchange of flow through the side holes located before and after it. This causes a lowering of WSS before and after the occlusion on the outer surface of the stent and an increase inside the stent. In the designs 4 and 5 the WSS reaches a maximum value of 0.430 Pa due to the occlusion.

However, it should be highlighted that these values are not reached in the side holes but in the internal

lumen of the stent.

The most relevant side holes, positioned before and after the obstruction, reached WSS mean values of 0.200 Pa and 0.300 Pa respectively for designs 4 and 5.

The results confirm that both designs, occluded or non-occluded, are prone to the deposition of encrustation in the side holes.

The designs 1, 2, and 3 do not have any active side hole, and designs 4 and 5 have 4 side holes active.

Considering that in each design there are 5 side hole holes, there is a percentage of active side holes equal to 80%. It should be noted that results provide only a partial representation of the all stented ureter system. In fact, the ureteric stent has a size of 26-27 cm, thus the results from this study suggest that only 10% of side holes in a full-scale stent would be active.

FUTURE COLLABORATIONS (if applicable)

The project has strnghtfull the existing links between Politecnico di Milano and University of Southampton.

REFERENCES

[1] A. Mosayyebi, D. Lange, Q. Yann Yue, B. K. Somani, X. Zhang, C. Manes and D. Carugo, “Reducing deposition of encrustation in ureteric stents by changing the stent architecture : A microfluidic-based investigation Reducing deposition of encrustation in ureteric stents by changing the stent architecture : A micro fl uidic-based investigation,” vol. 014101, 2019.

SHORT TERM SCIENTIFIC MISSION (STSM)
SCIENTIFIC REPORT

This report is submitted for approval by the STSM applicant to the STSM coordinator

Action number: CA16217- 43558
STSM title: Unravel the mechanisms behind the biofilm formation on ureteral stents
STSM start and end date: 18/02/2019 to 08/03/2019
Grantee name: Joana Maria Marques da Silva

PURPOSE OF THE STSM:

The main purpose of this STSM was to unravel the effect of impregnation processes with different fatty acids (e.g., capric acid, lauric acid, stearic acid and linoleic acid) and natural compounds (e.g. curcumin) on the antimicrobial properties of ureteral stents. Thereby, the antimicrobial properties were evaluated using gram positive (i.e., Staphylococcus aureus) as well as rumen fluid and pig feces. The main aim was the identification of the formulations effective at neutralizing pathogens and disrupting biofilm formation.

DESCRIPTION OF WORK CARRIED OUT DURING THE STSMS

Among the parameters that play a key role in the performance of an ideal stent are their material, design and surface coating. In this STSM, the main goal was to evaluate the biological performance of stents loaded with different compounds. The loading of different compounds was performed by supercritical fluid impregnation using similar operation conditions in which respect temperature, pressure, reaction time and venting time (i.e., depressurization). The compounds were selected due to their antimicrobial properties. Fatty acids are well-known to present antibacterial and antifungal properties. Thereby saturated and insaturated fatty acids were used such as capric acid (CA), lauric acid (LA), stearic acid (SA) and linoleic acid (LIN). Additionally, curcumin (CUR), was also used due to its remarkable properties, namely their antimicrobial properties. The experimental setup used for the impregnation process was adapted from previous works developed in our group. Briefly, the experiments, with a duration of 24 hours in order to achieve the complete saturation of supercritical carbon dioxide with compounds, were carried out at 100 bar, 50°C and with venting rates of 30-90 minutes. It should be noted that for CUR impregnation, two approaches were used with and without ethanol as co-solvent. Afterwards, the formation of biofilm on the impregnated stents was assessed via Scanning electron microscopy (SEM) and a method based on Colony forming unit (CFU) counting. Briefly, impregnated stents and a non-impregnated stent (control sample) were sectioned in half (i.e., to correctly evaluate biofilm formation nor only on stent surface but also in its inner area) and repeatedly washed using distilled water to remove non-impregnated material in the stent surface. Stents were then placed in a 1.5 ml microtube (1 stent per tube) containing a 1ml inoculation, in Mueller-Hinton broth (MHB), of the selected bacteria strain (Staphylococcus aureus ATCC 25923) at an optical density (600 nm) value of 0.1. Stents were incubated in the suspension for a total period of 24 hours (20 hours with shaking at 180 rpm and 4 hours with no agitation). Stents were then aseptically removed from the culture and washed with 2 ml of phosphate buffer to remove planktonic cells and culture media, followed by processing for downstream analysis. For SEM, stents were placed in 1 ml of a 10%(v/v) formalin solution and incubated at 4°C for 1 hour for biofilm fixation. After incubation, formalin was removed, and stents washed with PBS to remove any remaining compound. Samples were then dehydrated by a series of 10-minute incubations at increasing ethanol concentrations and left in vacuum overnight to evaporate any remaining solvent. Finally, samples were mounted and gold-sputtered for posterior analysis. For CFU counting, stents were placed in 1 ml of phosphate buffer and subjected to two cycles of vortex (45 s) intermitted by sonication at 35 kHz for 2 minutes at room temperature. Samples were then serially diluted (dilution factor of 10) and 100 µl of each dilution spread in tryptic soy agar (TSA) plates. Plates were left

incubating at 37°C for 24 hours, followed by colony counting and determination of bacterial log_{10} reduction values.
The assessment of the antibacterial was also performed by using rumen fluid and pig feces resuspended in PBS (i.e., 50 ml of phosphate buffer). In brief, each tubular formulation was incubated in both media for 3 days at 39°C. Afterwards, the samples were removed from the fluids and washed in order to remove the remaining fluid entrapped inside the stent. Then, DNA was extracted from each formulation and controls (i.e., coated and non-coated stents that were not exposed to the fluids) using FastDNA SPIN Kit.
PCRs amplifying the V1-2 region of the 16S rRNA gene were performed with the DNA to detect bacteria in each formulation and control. Amplicons were verified by agarose gel electrophoresis, purified and normalized and sequenced using 250 bp paired-end sequencing chemistry on an Illumina MiSeq platform.
Finally, ureteral stents obtained from Hospital Braga were also prepared for sequencing. In brief, urinary stents were sectioned into 3 different zones labeled upper, mid and lower. Samples were stored in RNA later solution. However, before processing the samples were kept in phosphate solution. As such 15 ml falcons containing the initial storage fluid of urinary stents were also evaluated. At the same time, a piece of each sample was kept for microorganism culturing and isolation. Samples were placed in 5 ml of tryptic soy broth and left incubating overnight at 37ºC 180 rpm. At this point, 900 µl of the various inoculations was prepared for preservation at -80°C by addition of an isovolume of glycerol 30%(v/v). Simultaneous, a sterile loop was lightly dipped in the various inoculations and used to inoculate TSA and sabouraud Dextrose Agar (SDA) plates by streaking. Plates were incubated at 37ºC for 24 hours.

DESCRIPTION OF THE MAIN RESULTS OBTAINED

The biofilm assessment was initially performed against S.aureus using SEM and colony counting from the extracted biofilm. SEM results (**Figure 1**) show that depending on the compounds impregnated the propensity for biofilm formation is clearly different. Hence, it is possible to conclude the efficacy of CA, LA, LIN, when compared with CUR and SA. The data corroborated the colony counting results present in **Figure 2**. However, it should be noted that in SEM more bacteria were observed in the inner side of stents which indicates that the impregnation may need to be optimized in order to achieved better results. Additionally, in colony counting CUR and SA did not show any log reduction, whereas at SEM it looks that a small reduction occurs. Thereby, the extraction method from biofilm will also need optimization. The bacterial log_{10} reduction values calculated are LA - 5.288; CA - 5.288; SA – 0; LIN - 1.81 and CUR -0. It should be noted that in future studies SEM analysis should also be performed after biofilm extraction to confirm the successful extraction.

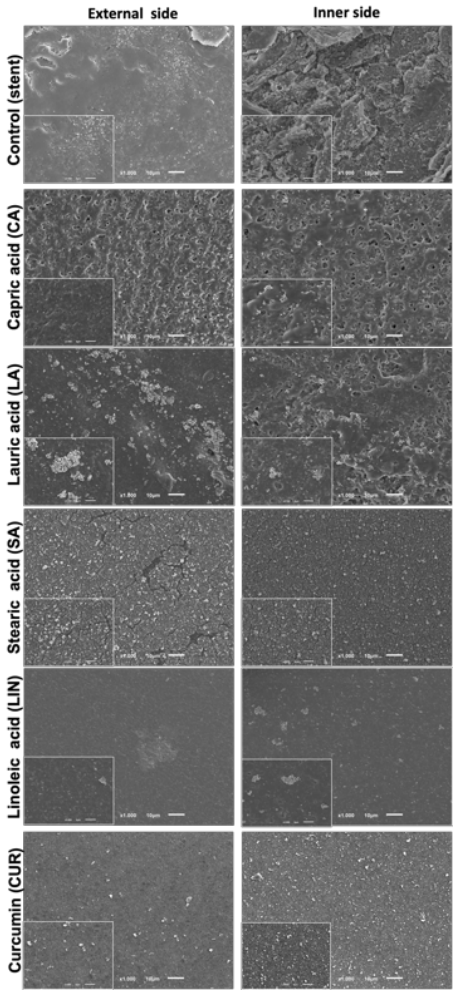


Figure 1. SEM micrographs of the surface of different stents' formulations upon contact with S.aureus.

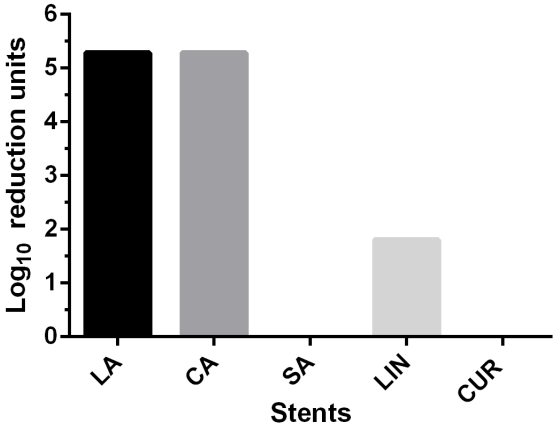


Figure 2. Log_{10} reduction values of S.aureus adhered to impregnated stents upon overnight incubation in MHB.

Additionally, our stents were also exposed to a cocktail of microorganisms by using rumen fluid and pig feces and the results are shown in **Figure 3.**

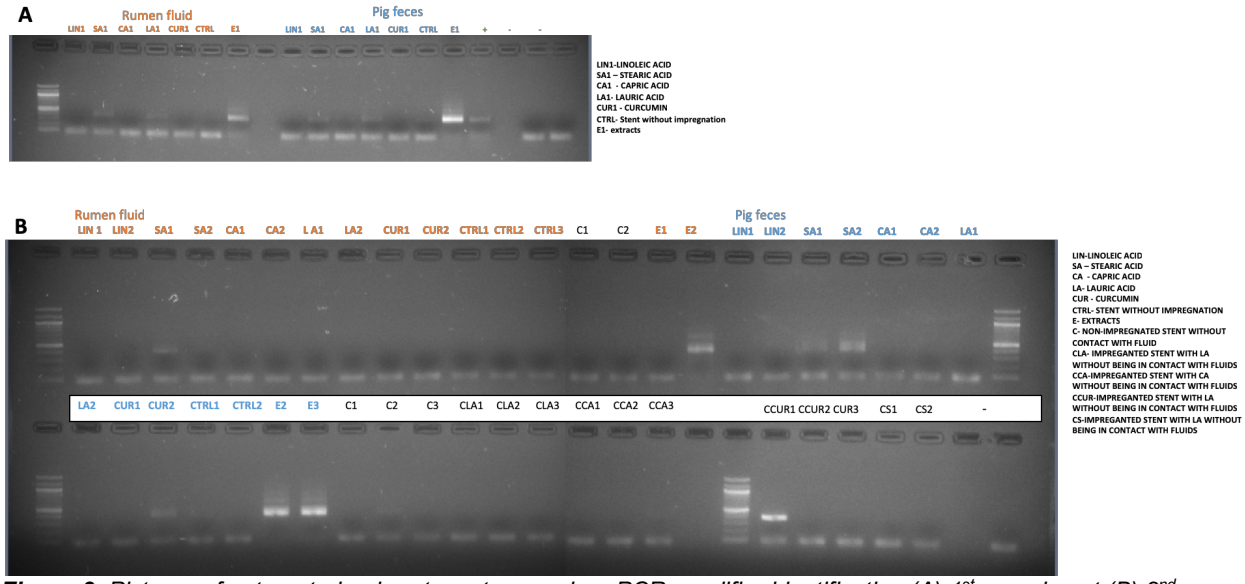
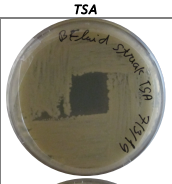
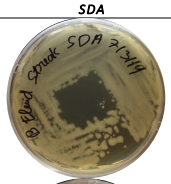
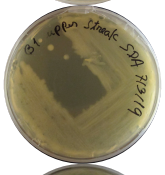
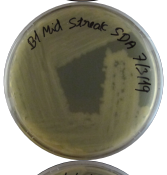
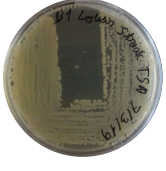
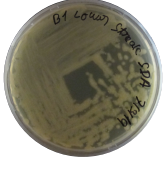


Figure 3. *Pictures of automated gel systems to speed-up PCR amplifier identification (A) 1st experiment (B) 2nd experiment.*

The 1st assay indicates that samples more susceptible to form biofilm when in contact with rumen or pig feces are SA and LA. However, in both cases, the CTRL (stent without suffering impregnation step) no amplification was detected, which was not expected. Thereby, in the second experiment more samples were evaluated and some variations between the samples occur which indicates that the biofilm extraction needs to be optimized. Additionally, it should be noted that amplification was observed in one stent impregnated with CUR that was not exposed to the fluid, which may indicate that we had some contamination during the processing of the samples. In order to make a bacterial profile of all the samples, they were sent to sequence. With this, the microorganisms that adhered to the stents should be clearly identified and differences between the controls and samples characterized. The sequencing results will be ready within few months.

The characterization of microorganisms from hospital ureteral stents was also started and till now the obtained data from the culturing indicate that different microorganisms are presented, namely different bacteria and possibly fungi (Table 1), since SDA plates are more appropriate for the growth of fungi. In order to identify the different organisms, present subsequent steps of streaking/culturing are required as well as dilution of the original inoculation to reduce total biomass which facilitates the obtention of single colonies. Furthermore, DNA was also extracted from the stents to be in the future characterized by sequencing (host institution).

Table 1. Streaking of overnight infected stent inoculations on TSA and SDA plates.

Samples	*TSA*	*SDA*
Storage fluid of extracted stent		
Upper Part of Stent		
Middle Part of Stent		
Lower Part of Stent		

FUTURE COLLABORATIONS (if applicable)

The collaboration will proceed in the future, as it will be valuable for both the host and the applicant. In the future, the microbiota of ureteral stents will be further evaluated using commercial stents previously used in clinical practice. Briefly, the microorganisms present in the stents (i.e., upper, middle, lower part) will be identified using the expertise of the host. 3B's group will extract the microorganisms and culture them in the stents impregnated with different compounds. Afterwards, the biofilm propensity will be evaluated using culturing and high throughput sequencing (amplicon sequencing and metagenomics), which may allow us to characterize the biofilm composition on the different formulations of stents, as well as, the mechanisms behind its formation.

SHORT TERM SCIENTIFIC MISSION (STSM)
SCIENTIFIC REPORT

This report is submitted for approval by the STSM applicant to the STSM coordinator

Action number: CA16217
STSM title: Investigating the time evolution of encrustation in ureteric stent: in-silico and in-vitro
STSM start and end date:15/02/2019 to 22/04/2019
Grantee name: Maryam Mosayebi

PURPOSE OF THE STSM:

Ureteral stents are one among commonly-employed urologic tools to provide urine drainage when the ureter is blocked by obstructions, such as stones or tumors. Designing a flexible tube containing multiple side holes allows for urine to bypass the occlusion. Despite the advantage of stents towards allowing urine drainage, majority of them suffer of biofilm forming and encrustation [1]. (see figure 1)

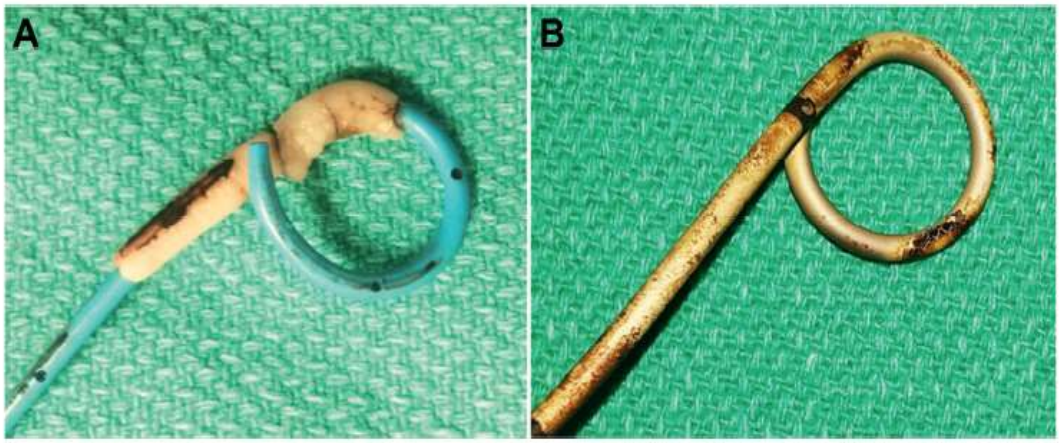


Figure 1. (A) forgotten stent after 1 year in cystinuric patient. (B) Forgotten stent after 2 years in paraplegic patient [2].

In general, the focus of this research is on the understanding and quantification of crystallization processes of chemical compounds, usually found in urine, over stents' surfaces. The long term goal is to use this wealth of knowledge to develop a simple model that is able to replicate the occurrence of crystals and their growth rate over stents' surfaces. Within the time constraints (about 3 months) of this visit, the objectives are framed as following:

1) identifying and understanding relevant processes governing inorganic crystal growth in standing fluid conditions
2) identifying and understanding processes governing crystallization in moving fluid conditions
3) Develop one (or more) simple mathematical models that relates flow properties to crystal growth rates

DESCRIPTION OF WORK CARRIED OUT DURING THE STSMS

The study included a systematic literature search using specific keywords such as: “Crystal growth, Encrustation, Crystallization in moving fluid and crystal forming in urinary system”. This literature review was carried out using “google scholar” and “ISI web of science”. This report provides a summary of the acquired knowledge.

DESCRIPTION OF THE MAIN RESULTS OBTAINED

Crystallization processes in standing fluids:
Understanding the crystallization processes in urine-solutions is a key aspect when it comes to identify strategies to reduce encrustations in urinary stents. The formation of a crystal in standing fluids includes two stages: nucleation and growth [3].
Nucleation is first step in crystallization and it relates to new structure with lower free energy [4].
As far as growth is concerned, in standing fluid conditions Vekilov [5] quantifies it using a two-factors equation (see equation 1).

$$\beta = \frac{a}{n_k} \upsilon_{+} \exp\left(\frac{\Delta S}{K_B}\right) \exp\left(\frac{-\Delta H}{K_B T}\right) \quad (1)$$

Where β (cm/s) is step velocity coefficient (Crystal growth rate relates to growth of steps in crystal); a (non dimensional) is the Miller index which dictates the shape of the crystal; n_k is the mean number of molecules between two kinks; υ related to the step velocity (cm/s); ΔS is entropy of system (KJ/mol K); ΔH is enthalpy of system (KJ/mol); K_b is Boltzmann constant (J/K) and T is absolute temperature respectively (K).

Equation 1, was derived, assuming: (i) the supersaturation level (i.e. the ratio between the concentration of the solutions and the critical concentration) is near 1; (ii) incorporation of molecules from the solution into kinks follows a first-order rate law (kinks are locations in the crystal lattice where molecules attach to).

Crystallization in moving fluids
The effects of flow conditions on crystallization is an old subject, which has been investigated extensively, although a clear predictive formula and correlation with flow metrics could not be found. Most of the literature focuses on the groth of crystals in devices that are empirically designed to obtain the largest and ourest crystals for industrial applications. The text below focuses on two papers that provide the most interesting information related to the problem of stent ureter systems that is the subject of this report.

In order to understand the relation between flow and crystallization, Zeng et al (2018) [6] investigated the relation between the velocity field and the growth rate of crystals developing in a microfluidic device.

Zeng et al (2018) report that the factors which affect the crystal growth rate are: fluid velocity (the higher the velocity the lower the growth rate); supersaturation (increasing supersaturation levels leads to increased growth rate); location in the fluid domain (downstream crystals are exposed to lower levels of supersaturation).

In another key study, Mura et al (2016) [7] identified that crystal growth rates are strongly correlated to viscous shear stress crystals are exposed to. This is quite promising as, as reported by Mosayyebi et al (2019), the crystal growth rate in a microfluidic chip mimicking stent-ureter conditions was also strongly correlated with the magnitude of viscous shear stress. This paper, therefore, confirms the conjectures of Mosayyebi et al (2019) and provides the basis for future modelling purposes focused on stent ureter systems.

FUTURE COLLABORATIONS (if applicable)

Futrue work will include the validation of the predictive model provided by Mura et al (2016) to explain the experimental data by Mosayyebi et al (2019). if succesfull, we will implement the model as a User Defined Function in Fluent as a routine that couples fluid dynamics and crystal growth rate in I n silico models of stent-ureter systems.

References

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SHORT TERM SCIENTIFIC MISSION (STSM)
SCIENTIFIC REPORT

This report is submitted for approval by the STSM applicant to the STSM coordinator

Action number: Action CA16217 - STSM ECOST-STSM-Request-CA16217-42625
STSM title: Comparison of the efficacy of nephrostomy placement to the insertion of double-J in septic patients with upper urinary tract obstruction.
STSM start and end date: 19/11/2018 to 10/12/2018
Grantee name: Sara Villarroya Castillo

PURPOSE OF THE STSM:

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The aim of this STSM was the investigation of the effect of nephrostomy in comparison to double-J in the clinical outcome of septic patients with urinary tract obstruction.

DESCRIPTION OF WORK CARRIED OUT DURING THE STSMS

A retrospective stydy was designed to evaluate the files of septic patients with urinary tract obstruction who were managed by either nephrostomy or double-J catheter in the University Hospital of Patras (Greece).

The approval of the scientific committee of the institution was obtained.

During my presence in Patras, a database was designed with the the variables that we are going to be analyzed.
The data extraction started by investigating the patient files.

I have also written a systematic review on the current status of drug eluting/coated stents/balloons in the urinary tract under the supervision of Prof. Kallidonis.

DESCRIPTION OF THE MAIN RESULTS OBTAINED

The systematic review is on the process of revisión by Dr. Kallidonis for submission in one of the international journals.
The comparative Project is progressing by continuing the data collection from the team of the University Hospital of Patras. Data Will be available within the next weeks.

FUTURE COLLABORATIONS (if applicable)

When the data of the collaborative Project are available, my task will be to analyze the data and prepare a manuscript fro publication.

SHORT TERM SCIENTIFIC MISSION (STSM)
SCIENTIFIC REPORT

This report is submitted for approval by the STSM applicant to the STSM coordinator

Action number: CA16217
STSM title: Drug Eluting Urinary stents and urine flux monitoring
STSM start and end date: 14/04/2019 to 18/04/2019
Grantee name: Valentina Cauda

PURPOSE OF THE STSM:
(max.200 words)

The collaborative study was carried out between the company HydrUStent (Porto, Portugal), acting as Host Institution for the grantee, and the Politecnico di Torino (Italy), acting as Home Institution.

The purpose of this STSM was to exchange ideas and start frutiful discussion on-the-field about the development of innovative antibacterial and drug-eluting ureteral stent materials combining the zinc oxide (ZnO) nanomaterials developed by Prof. Cauda at Politecnico di Torino and drug-eluting and biodegradable ureteral stents developed by HydrUStent, in the person of Dr. Barros.

ZnO has documented strong antimicrobial properties, is biodegradable and shows networks of nanopores which present ideal characteristics for drug-eluting functions. The polymeric materials developed by HydrUStent for producing stents can be thus loaded with such nanomaterial to obtain multipurpose functions in an ureteral stent: drug-elutilng capabiltiy sustained over time, biocompatibility, antimicrobial properties preventing bactieria colonization, and final biodegrdation in a tunable time span.

DESCRIPTION OF WORK CARRIED OUT DURING THE STSM

(max.500 words)
The work started with a meeting between the HydrUStent CEO, Dr. Alexandre Barros, and his team with the grantee, Prof. Valentina Cauda. There was a full introduction to the polymer hydrogel formulation of the stents and formation of the stents in the final double-pig tail tubular form. Thereafter the grantee exposed the synthesis procedure of ZnO nanostructures based on a sol-gel hydrothermal approach and the possibility to vary different synthesis parameters (reagents type and concentration, type of solvent, temperature of the process) which then impart different final morphologies and sizes leading to various material properties (higher surface area, useful to upload drugs, for example). The grantee brought with her two kinds of ZnO powders, the first with a size of 20 nm and a smooth surface, i.e. nanospheres, the other with a size of 4 µm and a highly nanostructured surface, i.e. desert roses-like microparticles.
The grantee was trained in the company facilities and the adjacent research group of 3B's in the university campus where the company is hosted. In particular, polymer compounding at industrial level, processing and materials formulation, instruments (in particular injection molding) used by the company to prepare the ureteral stents. Various key parameters of the process were illustrated, such as the viscosity, water content in the polymer, temperature of the process, injection speed.

Experimental integration of ZnO particles on the polymer used to fabricate the stents was developed by either compounding to form polymeric composites or coating already preformed polymers.
Validation techniques to characterize the materials were discussed and the trainee wil perform the materials characterization in the home Institution (Politecnico) once back, i.w. scanning electron microscopy, energy dispersive spectroscopy and X-ray diffraction.
Further discussion was done also about the patent on the monitoring of urinary flux from prof. Cauda and the solution that HydUStent actually proposes on the market.

DESCRIPTION OF THE MAIN RESULTS OBTAINED

The following main results in this STSM were achieved:

- Full training of the academic researcher on industrial processes for stents formulations, biomedical device positioning on the market and deals with hospitals, suppliers, distributors.
- Adoption of a strategy to synergize between the research on nanomaterials conducted at the Politecnico di Torino by Prof. Cauda and the product developed by the company HydrUStent: novel composite polymeric materials were prepared eithwr based on polymer compounding with ZnO powders or on ZnO coatings on the already pre-formed stents of the company. The experimental work served to understand the potantialities of the future stents in terms of its biocompatibility, drug-eluting properties and biodegradability, but also to focus on the possible challenges, bottlenecks and failure. In this regard, it was decided that the use of pure ZnO coatings on the stents is not a feasible and safe solution, because of the risk of cytotoxic behavior of the nanomaterials towards the ureter epithelium and potential encrustation formation of organic salts from the urine flow. In contrast the adoption of a composite materials were the ZnO particles are embedded rin the polymer show the advantages to have a more uniform material, control the release of drug in a sustainable time manner and
- Generation of synergies between the deposited patent of Prof. Cauda and the company HydrUStent concerniung the novel stents formulation and the monitoring of urine flow.
- A future action planning was also done to continue the research

FUTURE COLLABORATIONS (if applicable)

The collaboration between the grantee, prof. Valentina Cauda and her reseach group in Politecnico di Torino, and HydrUStent company, in the person of Dr. Alexandre Barros and his team will go on in different ways.

First, the experiemtal work to finish testing the novel polymer formulation for smart stents will be continued by futher researcher exchange. Possibly a new STSM application of one researcher to the Cauda's research group in order to test the dissolution and drug eluting properties will be carried out in the near future.

Secondly, technology transfer potential between the patent deposited by Prof. Cauda can be assessed thanks to the experience gained in the field by Dr. Barros, leading to industrial synergies.