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# A simple way to graft a bioactive polymer - polystyrene sodium sulfonate on silicone surfaces

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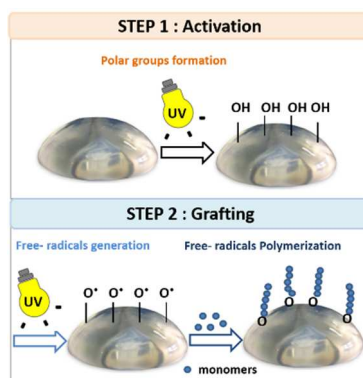
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**Abstract:** Silicone-based materials are largely used in plenty of fields, especially in the biomedical field as implantable devices for surgery purposes. However, due to recent issues, the safety of the material has been reconsidered. In this work, we aim to graft a bioactive polymer on silicone breast implants shell surface to allow better integration of the implant inside the body and also a decrease of the bacterial adhesion. To functionalize silicone material, we develop a simple method to first activate the surface only using UV irradiation allowing then the radical polymerization of sodium styrene sulfonate to occur. Different parameters such as activation time, monomer concentration or sample size, have been studied to get optimum grafting rates. All along the grafting process, surfaces are characterized using ATR-FTIR, SEM-EDS, WCA measurements, colorimetric method, and XPS. Our results indicate that with the use of UV irradiation, it is thus possible both to activate and initiate the grafting of the bioactive polymer on silicone surfaces.

**Keywords:** silicone, implant, bioactive polymer, grafting

## Graphical abstract



## 1. Introduction

Poly(dimethylsiloxane) (PDMS) more commonly known as silicone is an elastomer widely used in a large range of applications such as the fabrication of microfluidic devices [1] or as raw materials for biomedical implants for various surgery applications [2]. The material is known for its inertness, flexibility, low cost, and ease of fabrication. Silicone-based breast implants are used since the 1960s for reconstructive or plastic surgery. However, after long term implantations, several issues appeared such as ruptures and fibrosis. This leads to discuss and re-consider the PDMS non-invasive aspect and biocompatibility. Complications due to breast prosthesis implantations include hematoma, infections, inflations, silicone gel perspirations or the apparition of capsular contractures [3], in the long term.

In the past years, many cases of large cell anaplastic lymphoma (a form of cancer that most of the time requires a prosthesis replacement) have also been reported post-surgery [4] on women with breast implants after 8 -10 years. According to the American Society of plastic surgeons, 694 cases of BIA-ALCL have been identified worldwide since February 2019 (<https://www.plasticsurgery.org/>). In France, the equivalent association (ANSM) has reported 59 cases including 5 deaths in the country, thus raising the safety issues of such implants. In this study, we have been focused on smooth silicone breast implants as textured implants are being slowly removed from the international market and banned in France since February 2019 (<https://www.ansm.sante.fr/>). One of the main reasons for all the occurring problems is bacterial contamination during the first implantation surgery. Indeed, investigations of biofilms from explanted implants with capsular contracture showed the presence of *Staphylococcus epidermidis*. Researchers have proven that the severity of the contracture was directly correlated to the number of colonizing bacteria [5]. The bacterial contamination is supposed to be due to the hydrophobic behavior of silicone surfaces that promotes the adsorption of plasma proteins leading to the adhesion and proliferation of bacteria. In the long term, a deformation around the outer shell of the prosthesis is visible, which may be due to an inflammatory response. This non-controlled host response results in the generation of a fibrous shell around the implant which later leads to complications such as the calcification of the breast implant that is responsible for pains and skin deformations [6].

To overcome this problem, surface modification is needed. A lot of work has been done in the microfluidic field to improve silicone material surface wettability by grafting hydrophilic polymers [1] expecting a better flow of polar liquid through PDMS channels. However, before any modification, silicone surfaces usually need to be oxidized first to generate polar groups on its surface which is really difficult to achieve due to the low reactivity of the silicone. Mostly, PDMS surfaces are activated using UV/ozone [7-9] in dry environments or by oxygen plasma treatment to generate oxygen reactive species (-OH;-OOH) [10-12]. However, it has been shown that created oxygen species are not stable in time and thus, a quick recovery of PDMS surface hydrophobicity was observed [27].

A silicone-based quarter polymer has been developed previously in the laboratory [13]. It resulted from the UV-combination of two copolymers carrying bioactive groups: on one hand silicone-methacrylate moieties and sulfonate-carboxylate groups on the other hand. This quarter polymer has significantly reduced the bacterial adhesion (*S.Aureus* & MRSA 88244) by 80% *in vitro* and by more than a log of difference *in vivo* (1.5 to 2 log) between a non-grafted (control) sample and a grafted sample [13]. However, the synthesis required several steps which made it difficult to scale up for industrial applications. Another method to improve the biocompatibility consists of the covalent grafting of peptide molecules. But despite the comparable antibacterial activity, the system has a short lifetime and arise high cost [14].

Here we have elaborated a simple method to graft PDMS sample with a bioactive polymer, and we have studied the influence of different synthesis parameters such as surface activation conditions, monomer concentration, and sample's size. We have characterized the grafting by different

techniques such as Fourier Transform Infrared Spectroscopy in the ATR mode, ATR-FTIR, water contact angle measurements (WCA), X-ray Photoemission Spectroscopy (XPS), electronic microscopy with chemical analysis (SEM-EDS) or colorimetric measurements.

In the past years, our team has successfully developed functionalization methods of bioactive polymers on metallic surfaces (titanium and its alloys [15-17]) and polymers surfaces (PET [18], PCL [19-21]). The grafting of one specific polymer, the polystyrene sodium sulfonate (polyNaSS) on these surfaces was carried out under heating [15, 18, 19] or UV irradiation [16,17, 20, 21]. Both techniques allow the creation of radical species permitting the initiation of the polymerization. The grafting of such polymer increases the wettability of the surfaces making them more appropriated for the biological environment. Most importantly, grafted bioactive polymers have also reduced bacterial adhesion [13,22,23]. The mechanism of action can be described as follow: polyNaSS helps to modify the adsorbed plasmatic proteins conformation on the implants making them non-identifiable by the bacteria receptors. This prevents bacterial adhesion and the risk of infection [13,23]. With the described context and inspired by previous work, we aim to develop a simple way to graft polyNaSS on silicone surfaces through a radical polymerization of sodium styrene sulfonate.

In this work, we will be focused on the grafting of the bioactive polymer on the outer shell of smooth silicone breast implants, i.e. the side that is in direct contact with the biological environment. This type of implants is composed of silicone in different forms: the silicone elastomer composing the outer shell, the silicone gel filling the prosthesis and silicone liquid in the mixture of the gel.

## **2. Materials and Methods**

### **Material**

Silicone samples were extracted from the outer shells of smooth breast implants. The prosthesis was cut in half and silicone gel was rid. Gel residues were removed with two washings in tetrahydrofuran (THF, FISHER Chemical) under ultra-sonication two times, five minutes each. Then, the samples were rinsed in distilled water for five minutes three times. Finally, samples were left dried in a 37°C oven overnight and cut into 1 x 1 cm square-shaped samples before experiments.

Sodium styrene sulfonate (NaSS, Sigma), the monomer used for the grafting process is purified by a recrystallization process in a mixture of water/ethanol (Carlo Erba) (10 : 90 v/v) overnight at 70°C. A first Büchner filtration is realized and the filtrate was kept at 4°C for 48 hours before the second filtration. The purified NaSS was then dried under atmospheric pressure at 50°C overnight and then stored at 4°C [15,17].

UV source used comes from a Lot Quantum Design UV lamp delivering a power at 365 nm under normal conditions of temperature and pressure.

### **Methods: grafting process**

**Surface activation:** Silicone sample was placed in a round-bottom flask containing distilled water previously degassed and was left under UV irradiation at 365 nm for one hour at 160 mW/cm<sup>2</sup>.

**Grafting procedure (cf. Method C):** After the surface activation, the sample was transferred to another round-bottom flask containing a degassed aqueous solution of NaSS and left under UV irradiation in the same conditions as described above. The surface was then rinsed in distilled water for 24 hours and dried overnight at 37°C before analyses.

### **Surfaces characterizations:**

After each surface modification step, silicone samples were characterized by Fourier-transform infrared spectra recorded in attenuated total reflection mode (ATR-FTIR), X-ray photoelectron spectroscopy (XPS), scanning electron microscopy (SEM), colorimetry, water contact angle measurement (WCA). Changing in terms of surface compositions, physical properties or surface textures can be highlighted.

**Fourier-transform infrared spectroscopy (FTIR):** Analyses were performed on a Perkin Elmer Spectrum Two Spectrometer in the 600 to 4000  $\text{cm}^{-1}$  spectral range with a resolution of 4  $\text{cm}^{-1}$ . Silicone samples were directly pressed against the diamond crystal for acquisitions (128 scans) and obtained spectra were then fitted and analyzed.

**X-ray photoelectron spectroscopy (XPS):** To examine possible change in terms of the surface elementary composition after the functionalization, we aim to target the presence of sulfur atoms attributed to polyNaSS grafting silicon samples were analyzed by XPS. These analyses were performed using an Omicron Argus spectrometer (Taanusstein, Germany) equipped with a monochromated  $\text{AlK}\alpha$  radiation source ( $h\nu = 1486.6 \text{ eV}$ ) working at an electron beam power of 300 W. Photoelectrons emission was analyzed at a takeoff angle of  $90^\circ$ ; the analyses were carried out under ultra-high vacuum conditions ( $\leq 10^{-10}$  Torr) after introduction via a load-lock into the main chamber. Spectra were obtained by setting up a 100 eV pass energy for the survey spectrum and a pass energy of 20 eV was chosen for the high resolutions regions. Binding energies were calibrated against the C1s binding energy of aliphatic carbon atoms at 284.8 eV. Element peak intensities were corrected by Scofield factors<sup>[26]</sup> Casa XPS v.2.3.15 software (Casa Software Ltd, UK) was utilized to fit the spectra and Gaussian/Lorentzian ratio was applied (G/L ratio = 70/30).

**Scanning Electronic Microscopy - Energy Dispersive Spectroscopy (SEM-EDS):** Images were obtained with a HITACHI TM3000 with Oxford energy dispersive spectroscopy (EDS). This technic is mainly used to characterize surface topography with high-resolution image. Coupled with EDS, the device could be used for a qualitative analysis of the elementary composition on the sample's surface.

**Colorimetry:** Toluidine Blue (TB) is used as a colored indicator to quantify the concentration of polymer grafted on silicone samples. The samples, once grafted and washed were immersed in 5mL of TB (pH=10) for complexation for 6 hours at  $30^\circ\text{C}$ . After that, the excess of TB was removed with 3 washings in a solution of sodium hydroxide ( $\text{NaOH}$ - $10^{-3}\text{M}$ ), five minutes each. Then, complexed TB on sample surfaces was de-complexed by putting the samples in 5 mL of acetic acid (50/50 v/v) for 24hours. TB assays was carried out onto six samples for each conditions.

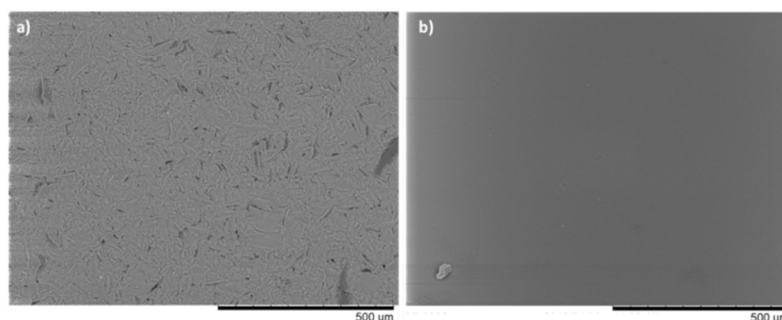
The method is based on the presence of a chromophore group on the TB molecule absorbing in the visible domain at around 633 nm. The molecule has a cationic group  $\text{N}^+(\text{CH}_3)_2$  allowing it to complex with negatively charged species such as sulfonate  $\text{SO}_3^-$ . According to Ikada *et al.*<sup>[24]</sup> work, stipulating that one mole of TB molecule complexes with one mole of carboxylate ( $-\text{COO}^-$ ). We assumed that it works for sulfonate groups ( $-\text{SO}_3^-$ ) as well. Then we are able to determine the concentration (=grafting rate) of grafted polymers on silicone surface using UV-vis spectroscopy after decomplexation.

**Water Contact Angle measurements (WCA):** Hydrophobic or hydrophilic behavior of the surface was evaluated by measuring the contact angles on the surface. This is a qualitative measurement of the wettability of the material. In our study, distilled water was used as the solvent and measurements were taken using a DSA10 contact angle measuring system from KRÜSS GmbH. Once the drop (2  $\mu\text{L}$ ) of distilled water is in contact with the surface, the measure was taken after 20 seconds with the Drop Shape Analysis (DSA) program that gives the value of the contact angle. At least three measures were taken and averaged.

### 3. Results and discussion

#### 3.1. Bare silicone

The first part of the study was the analysis of bare silicone shell samples. In terms of the surface's aspect, SEM pictures (*Figure 1*) showed that the shell exhibits two different surface textures. The inner side, ca. the one that was initially in contact with silicone gel, has a textured aspect (roughness visible in *Figure 1(a)*). On the contrary, the outer side, which is in direct contact with the biological environment once the prosthesis is implanted, is very smooth as seen in *Figure 1(b)*.



**Figure 1 : SEM analysis silicone shell's inner side (a) and silicone shell's outer side (b)**

*The observed difference may be attributed to the way the shell has been manufactured. It is supposed that the outside was in contact with the mold while the inner side was exposed to ambient air. In this work, we will be mainly focussed on grafting bioactive polymer on the outside part of the prosthesis showing almost no surface roughness.*

*PDMS material exhibits methyl groups ( $-\text{CH}_3$ ) at its surface which makes the surface very hydrophobic. Water contact measurements on bare silicone samples have confirmed the hydrophobic behavior with high contact angle values of around  $105.00^\circ \pm 1.00$ .*

*The covalent grafting of polyNaSS on such surfaces aims to increase the wettability. First, the surface needs somehow to be activated. In other words, polar groups like hydroxyl groups ( $-\text{OH}$ ), carboxyl groups ( $-\text{COOH}$ ) or hydroperoxide ( $-\text{O}-\text{O}-$ ) have to be generated through the methyl groups present on silicone surface to allow the initiation of the NaSS monomer radical polymerization.*

*The grafting process is based on the “grafting from” method. Active species such as free-radicals are created on the surface allowing then initiation of the radical polymerization. This step is followed by the propagation step where initiated radicals will attack another monomer molecule that will, in turn, generates a radical capable of attacking another monomer. Finally, the reaction ends up with the termination step. In our case, polar groups need to be generated through an activation step to allow the fixation of polyNaSS on the surfaces [17,20,21,23].*

#### 3.2. Grafting

##### 3.2.0. Standart conditions of grafting – Method A

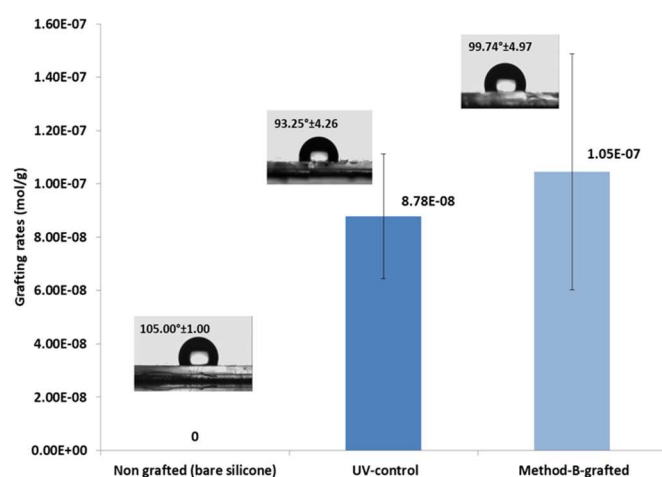
Several methods that have been developed in the laboratory to activate metallic [15-17] and polymers [18-21] surfaces were first tested on silicone surfaces. Among which acid activation and ozone activation were tested, detailed protocols are described in Appendix A (method A). Macroscopic degradation

was observed in the case of the acid activation. No significant polymer grafting was observed for both conditions according to colorimetric measurements and WCA measurements. Results are all resumed in figure A1.

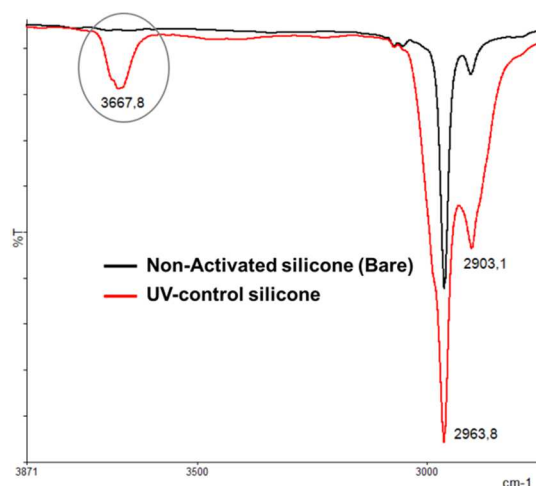
### 3.2.1. Direct grafting without activation – Method B

Inspired by previous work realized in the laboratory [20,21], where polyNaSS was successfully grafted on polymer surfaces without any activation step thanks to the presence of oxygen groups (-OH, -COOH) on the surface at the native state. Under UV irradiation, oxygen radical species are created meanwhile NaSS monomers polymerized on the surface. In the case of silicone surfaces, it was expected to maybe create carbon radicals on silicone methyl groups in the meantime of the polymerization without any previous oxidation step. Experimentally, the silicone sample was placed under UV light at 160 mW/cm<sup>2</sup> in a stirred degassed monomers solution for two hours.

Grafting rates did not show any improvement compared to the UV-control. The values remained practically the same compared:  $8.70 \times 10^{-8}$  versus  $1.05 \times 10^{-7}$  mol/g (Figure 2) and are relatively low for a significant grafting rate. Nevertheless, interestingly FTIR spectra (Figure 3) has revealed a feature at 3667 cm<sup>-1</sup> which can be attributed to free hydroxyl groups after UV treatment. Moreover, the contact angle measurements showed a slight decrease going from 105° before method-B grafting to 99.74° (Figure 2). Even if the surface remained hydrophobic.



**Figure 2 : Grafting rates of Non-grafted silicone & Method B-grafted silicone**



**Figure 3 : FTIR spectrum- Non-grafted silicone & Method B-grafted silicone = activated silicone**

These observations suggested that UV irradiation allows the creation of oxygen groups on silicone surfaces and can be used as the source of energy to effectively activate silicone surfaces.

### 3.2.2. Grafting with UV activation – Method C

Considering the previous results where UV light efficiency promotes the formation of polar groups on silicone surfaces. It is assumed for the following experiments:

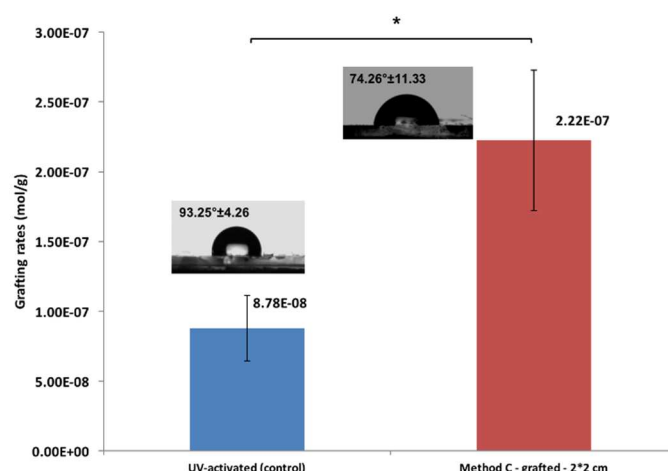
- Activation: 1 hour - UV irradiation (160 mW/cm<sup>2</sup>)
- Polymerization: 1 hour - UV irradiation (160 mW/cm<sup>2</sup>) in a degassed monomer solution (0.7M)

With this method, the obtained results are promising. First, the grafting rates (Figure 4) were much higher compared to the UV-control. More especially they are higher than the grafted samples following the previous grafting process (Method B):  $2.22 \times 10^{-7}$  mol/g versus  $8.78 \times 10^{-8}$  mol/g. Statistically, the difference is significant with a p-value equals 0.05(\*). XPS analysis also showed a difference in terms of composition : on the grafted sample, sulfur (S 2p) atoms were detected at 174 eV with a rate of 1.94% whereas it was practically undetectable on bare or UV-activated samples (Table 1).

Element	Position (eV)	Bare silicone (%atomic)	UV-activated silicone (%atomic)	Grafted silicone(%atomic)
O 1s	531	18.4	19.2	18.9
C 1s	283	50.9	49.2	52.3
<b>S 2p</b>	<b>174</b>	<b>0.4</b>	<b>0.3</b>	<b>1.9</b>
Si 2p	101	30.3	31.3	26.8

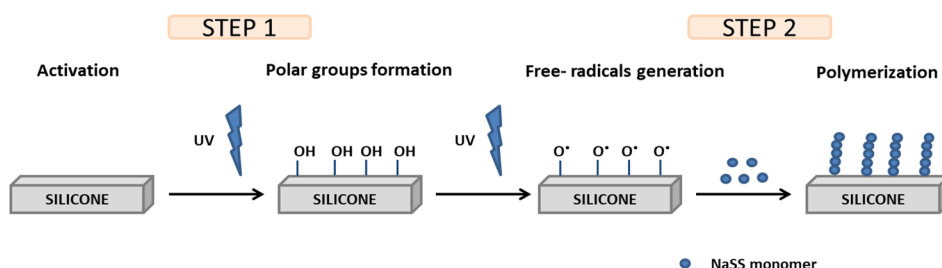
**Table 1 : elementary composition in atomic percentage obtained from XPS analysis of several silicone treated surfaces**

Contact angle measurements confirmed the observation (Figure 4). The surface went from really hydrophobic ( $\approx 105^\circ$ ) to hydrophilic surface ( $\approx 74^\circ$ ). The high standard deviations observed were supposed to be due to the non-homogeneity of grafting on the surface.



**Figure 4 : Grafting rates of UV-activated silicone & Method C-grafted silicone**

We can conclude that polyNaSS has been successfully grafted on the silicone surface with the developed two-step grafting protocol (method C) using UV irradiation. The wettability has clearly been increased. Grafting rates and XPS measurements have confirmed the presence of polyNaSS on silicone surfaces with this method. The mechanism is resumed in Figure 5.





## Figure 5 : Developed two-steps mechanism of grafting polyNaSS on silicone surface

### 3.2.3. Importance of the grafting under UV irradiation versus under heating (thermal grafting)

Thermal grafting was the initial method developed in the laboratory<sup>[15,17,18,19,20,25]</sup> to graft polyNaSS on different types of surfaces after the activation step (ozonation or acid treatment). Silicone samples were first activated under UV irradiation, placed into a degassed monomer solution and left in a thermal oil bath for 3 hours at 50°C. The results showed that compared to the previous method, thermal grafting showed any significant improvement in terms of grafting rates. It remained the same value as only UV-activated (control) samples:  $8.70 \times 10^{-8}$  mol/g (Figure A2-(3)). Contact angle results also confirmed the non-efficacy of the thermal grafting method, the grafted surfaces remained hydrophobic:  $108.13^\circ \pm 2.44$ . The explanation could be that thermal energy was not strong enough to break O-H bond (464 kJ/mol) created by UV light during the activation step. As no oxygen radical species were created, the radical polymerization cannot occur. Ozonation or acid treatment activation usually allows the formation of hydroperoxide (O-O) which required less energy (142 kJ/mol) to be broke than O-H bond to create oxygen radical species. As UV only allowed the generation of hydroxyl groups on silicone surface thermal grafting cannot be used for the radical polymerization reaction particularly on silicone surfaces.

### 3.3. Improvement of the grafting homogeneity

#### 3.3.1. Size of the sample

*When contact angle measurements were taken, we noticed that the lowest values were concentrated in the same area on the sample. Around this area, the values were slightly higher. It was proposed that it was due to the UV beam wideness which was not wide enough to equally cover the sample's surface. So, to improve the homogeneity of the grafting, the sample dimensions were reduced from 2x2 cm to 1x1 cm samples for the next experiments keeping the same grafting conditions.*

On Figure 6-(2), results showed that grafting rates were slightly increased compared to 2x2 cm samples :  $2.56 \times 10^{-7}$  mol/g versus  $2.22 \times 10^{-7}$  mol/g. More importantly, wettability was even higher with angles around  $40^\circ$  (\*p=0.05) with a lower standard deviation (Figure 7-(4)). The grafting homogeneity was improved.

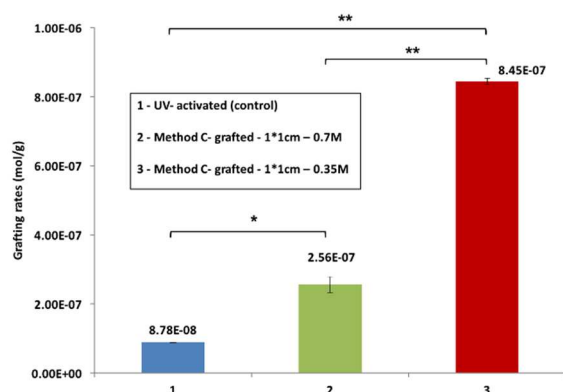
By this two-step protocol using UV to graft polyNaSS on silicone surface, we have optimal results for 1x1 cm squared-samples.

#### 3.3.2. Monomer concentration

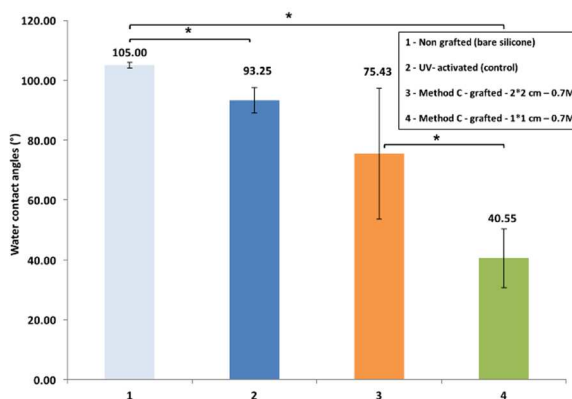
*On titanium surfaces, it has been shown that when the monomer concentration was reduced by half : 0.7M to 0.35M, grafting rates were much higher<sup>[17]</sup>. This allows both the increase of the grafting rates and economic save by using half of the quantity of monomer. In this study, a UV-activated silicone sample was immersed into a round-bottom flask containing a degassed monomer solution concentrated at 0.35M and left under UV irradiation for 1 hour at 160 mW/cm<sup>2</sup>.*

Results on Figure 6-(3) showed that with a monomer concentration of 0.35M, the grafting rates increased by four :  $8.45 \times 10^{-7}$  mol/g versus  $2.56 \times 10^{-7}$  mol/g. The variation is significant (\*\*p=0.005). The hypothesis suggested to explain such observation, is that the high density of molecules promotes the reaction of homopolymerization of monomers in solution which competes with the radical polymerization occurring on the surface of the sample. A lower density (0.35M) decreases interactions

between monomers so it decreases homopolymerization reactions. Accessibility to the surface is then better and thus, it may allow better polymerization rates on silicone surface. In terms of wettability, the contact angles remained practically the same as in the conditions of 0.7M, around 45°.



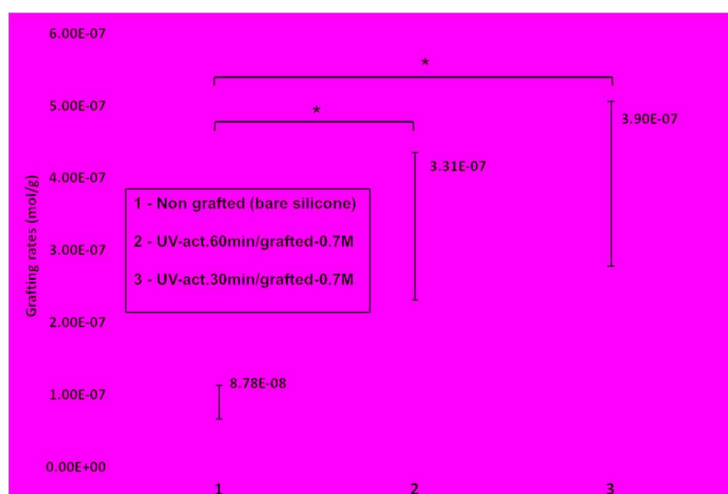
**Figure 7 : Grafting rates results of different grafting conditions**



**Figure 6 : Water contact measurements**

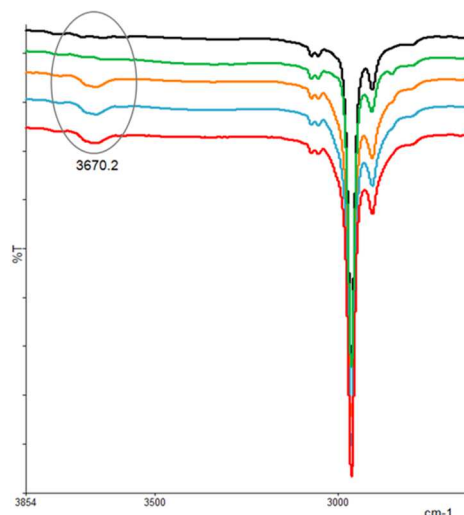
### 3.3.3. Reducing activation time

For all previous experiments, the UV-activation time was set at 1 hour. In the next experiments, the duration was reduced by half: 30 minutes. It was expected for the grafting rates to remain at least the same. Results are shown in Figure 8 where it is clear that for 60 minutes or 30 minutes of UV activation, grafting rates remained equivalent. Added to that, WCA also showed a hydrophilic surface with angles around 40°. The polymer grafting process duration time could be reduced to 30 minutes.



**Figure 8 : Grafting rates of different activation time grafted silicone (\*p=0.05)**

Furthermore, FTIR spectra shown on Figure 9 also confirm that only after 30 minutes (orange spectra) of UV-exposure hydroxyl groups are already formed with the band at  $3670\text{ cm}^{-1}$ . Obviously, after 45 or 60 minutes, these groups remain present.

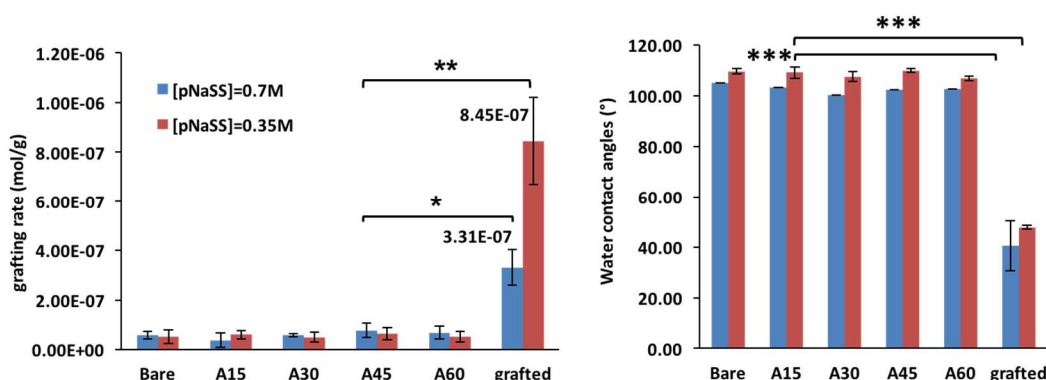


**Figure 9 : FTIR spectrum silicone samples activated at different time  
(black:bare silicone/green:15min-UV/orange:30min-UV/blue:45min-UV/red:60min-UV)**

All along with the study, we have first successfully grafted the bioactive polymer on silicone surface with an adapted activation step. Following that, we successfully refined the grafting parameters to get the highest rates of grafted polymer and also the most hydrophilic surface with optimum parameters (duration, concentration, sample's dimensions).

### 3.3. Physisorption

Physisorption tests were realized to confirm that polyNaSS has been covalently bond to the surface and not only physisorbed. First, silicone samples were activated at different time duration: 0, 15, 30, 45, 60 minutes under UV irradiation at  $160\text{ mW/cm}^2$  before they were put in an aqueous solution of polyNaSS for 24 hours under stirring. Afterward, samples were rinsed several times with distilled water for 24 hours and dried at  $37^\circ$ . Whatever the duration time of activation, surfaces remained hydrophobic with a contact angle varying around  $105^\circ$  for the physisorbed samples (Figure 10). The grafted sample clearly exhibit a hydrophilic behavior with a contact angle of  $40^\circ$ . On the other hand, grafting rates results also confirm the previous observation. Grafted samples exhibited a higher grafting rates compared to the physisorbed samples. Statistical analysis showed a significant difference between physisorbed and grafted samples, particularly at the concentration of  $0.35\text{M}$  (\* $p=0.05$ /\*\* $p=0.005$ /\*\* $p=0.001$ ). By these results, we confirm the covalent bond of polyNaSS on silicone surface following the grafting conditions we have developped.



**Figure 10 : Physisorption of polyNaSS grafting rates (left) and WCA measurements (right) compared to Method-C grafted silicone**

The developed “grafting from” method permit a covalent attachment between the polymer and the surface.

## 5. Conclusions

We have successfully developed a new approach to covalently graft polyNaSS on silicone breast implant outer shell's surface. Based on the work realized in the laboratory, several methods were tested to graft polyNaSS on silicone surface. Different techniques of activation were tested in vain: piranha solution (surface strong degradation) and ozonation. As well, different grafting methods were tested: UV grafting without activation and thermal grafting also showed really low grafting rates comparable to non-grafted samples. Experiments of UV grafted samples without activation step have did not show any improvement in terms of grafting rates compared to non-grafted sample, but FTIR spectra showed the apparition of hydroxyl groups after UV exposure. This led us to use UV irradiation to activate silicone surfaces: UV-activated samples clearly present a hydroxyl band at  $3670\text{ cm}^{-1}$  and a slight decrease of the contact angle ( $105^\circ$  to  $95^\circ$ ). Following the activation, polyNaSS was grafted onto the surface under UV irradiation at  $160\text{ mW/cm}^2$  as well.

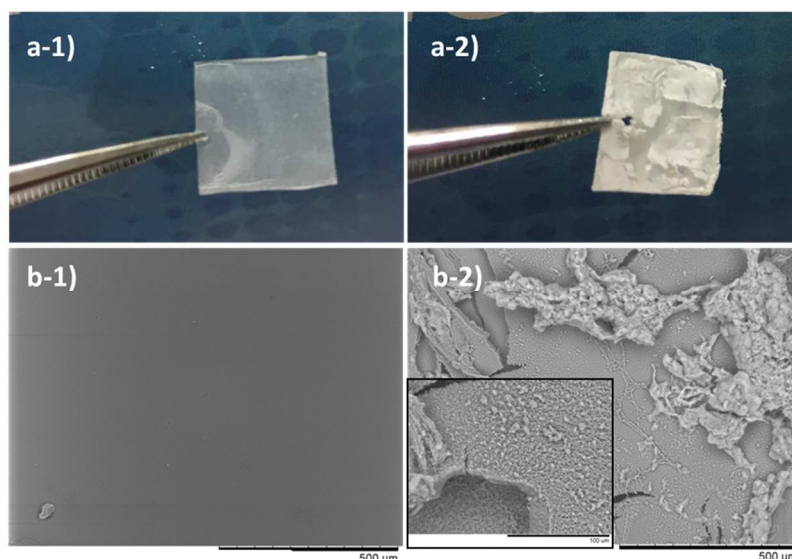
To conclude, we have fulfilled the main objective which was to find a suitable method to activate silicone surfaces by generating oxygen groups on it thanks to the use of UV irradiation delivering a power of  $160\text{ mW/cm}^2$ . We were then able to graft the bioactive polymer through a radical polymerization under UV. Once the grafting protocol was set up, the experiment parameters were refined to optimize the grafting rates: sample size, monomer concentration, and activation duration. The developed two-steps UV grafting process adapted for silicone breast implant shell is a low-cost process and most importantly is easy to realize which makes it industrially scalable.

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## Appendix A

### Standard conditions of grafting – Method A

Different activation methods that have been demonstrated efficient on metallic [15,16,17] or polymers [18-21] surfaces were tested to generate polar groups on silicone surface. Acidic oxidation and ozonation were separately tested to activate the silicone samples in a first step. For the acidic oxidation, silicone sample was immersed into a piranha solution ( $\text{H}_2\text{SO}_4/\text{H}_2\text{O}_2$  50/50 v/v). It was first soaked in sulfuric acid for 1 minute before hydrogen peroxide was added. The reaction was left for 3 minutes. An exothermic reaction occurred instantly and the surface was immediately attacked in the first seconds after the hydrogen peroxide was added. The sample was then quickly washed in distilled water three times and left dried at 37°.



**Figure A 1 : Optical pictures and SEM images of silicone sample before (a1 & b1) and after (a2 & b2) piranha oxidation**

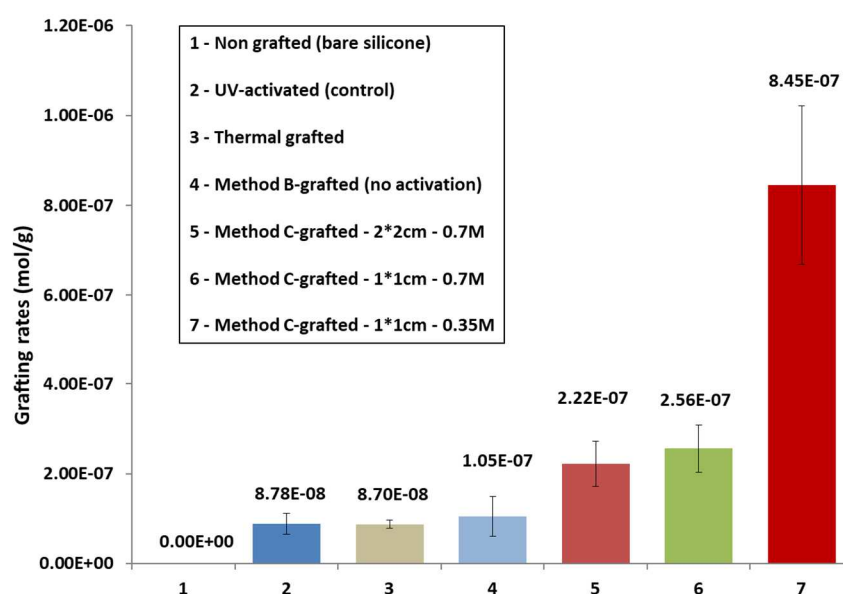
Macroscopically the surface was clearly degraded in the acidic mixture (Figure A 1). The solution strongly attacked the silicone sample with a detachment of a cracking white layer. Images of this newly formed layer were recorded with the SEM-EDS and a difference in terms of elementary composition compared to bare silicone was observed. On the piranha oxidized silicone surfaces the amount of oxygen was clearly increased while the quantity of carbon decreased by 10% (Table A 1).

We supposed that piranha solution has created an oxide layer on the sample and meantime has degraded the surface. That is why, this oxidation method cannot be used to activate this silicone material to create polar groups on it.

	Carbon (%atomic)	Oxygen (%atomic)	Silicim (%atomic)
Bare silicone	46.4 ± 0.9	32.6 ± 0,7	21.0 ± 1.0
Piranha oxidized silicone	36.8 ± 0.4	43.1 ± 0,4	20.1 ± 0.3

**Table A 1 : EDS elemental analysis (atomic%) of elements on silicone surface**

For ozonation activation, ozone was generated using an ozone generator BMT 802 N (ACW) with a pressure of 0.5 bars and an oxygen flow rate of 0.6 L.min<sup>-1</sup>. The samples were placed in distilled water where an ozone flow goes through for 20 minutes. Once activated, the sample was put under UV irradiation for 1 hour at 160 mW/cm<sup>2</sup> for the polymerization of NaSS monomers on the surface. In terms of grafting rates, no difference between bare silicone and ozone-treated-grafted silicon was observed. The surface remained hydrophobic with high contact angles around 105°. This led us to rethink the grafting process, especially the adapted activation protocol for silicone breast implant materials (Figure A 2).



**Figure A 2 : Grafting rates of the different conditions of grafting**

## References

1. I. Wong, C. Ho, *Microfluidics and Nanofluidic* **2009**, 7, 291-306.
2. F. Abbasi, H. Mirzadeh, A. Katbab, *Polymer International* **2001**, 50, 1279-1287.
3. <https://www.ecancer.fr/content/download/119635/1429833/file/Breast%20implantassociated%20anaplastic%20large%20cell%20lymphomas.pdf> (accessed on 11-09- 2019).
4. M. Mepin, H. Hu, D. Chowdhury, A. Deva, K. Vickery, *Material* **2018**, 11, 2393.
5. U. Rieger, J. Mesina, D. Kalbermatten, M. Haug, H. Frey, R. Pico, R. Frei, G. Pierer, N. Lüscher, A. Trampuz, *British Journal of Surgery* **2013**, 100, 768-774.
6. K.W. Dunn, P.N. Hall, C.T.K. Khoo, *British Journal of Plastic Surgery* **1992**, 45, 315-321.
7. H. Shahsavan, J. Quinn, J. d'Eon, B. Zhao, *Colloids and Surfaces, A: Physicochemical and Engineering Aspects* **2015**, 482, 267-275.
8. Y. Berdichevsky, J. Khandurina, A. Guttman, Y. Lo, *Sensors and Actuators B: Chemical*. **2004**, 97, 402-408.
9. J. Zhou, A. Ellis, N. Voelcker, *Electrophoresis* **2010**, 31, 2-16.
10. N. Maheshwari, A. Kottantharayil, M. Kumar, S. Mukherji, *Applied Surface Science* **2010**, 257, 333-664.
11. L. Carneiro, J. Ferreira, M. Santos, J. Monteiro, E. Giroto, *Applied Surface Science* **2011**, 257, 10317-10936.
12. S. Hemmilä, J. Cauich-Rodríguez, J. Kreutzer, P. Kallio, *Applied Surface Science* **2012**, 258, 9864-9875.
13. AC. Cremieux, G. Pavon-Djavid, A. Saleh Mghir, G. Helary, V. Migonney, *Journal of Applied Biomaterials & Biomechanics* **2003**, 1(3), 178-85.
14. C. Pinease, S. Jebors, P. Stoebner, V. Humblot, P. Verdié, L. Causse, X. Garric, H. Taillades, J. Martinez, A. Mehdi, G. Subra, *Material Today Chemistry* **2017**, 4, 73-83.
15. G. Helary, F. Noirclere, J. Mayingi, V. Migonney, *Acta Biomaterialia* **2009**, 5, 124-133.
16. C. Falentin-Daudré, V. Migonney, H. Chouirfa, J. Baumann, « Procédé de greffage de polymères bioactifs sur des matériaux métalliques. », B248515D34589, **2015**.
17. H. Chouirfa, V. Migonney, C. Falentin-Daudré, *RCS Advances* **2016**, 6, 13766-13771.
18. C. Vaquette, V. Viateau, S. Guérard, F. Anagnostou, M. Manassero, D.G. Castner, V. Migonney, *Biomaterials* **2013**, 34 (29), 7048-63.
19. A. Leroux, C. Egles, V. Migonney, *PLOS ONE* **2018**, 13 (10), e0205722.
20. G. Amokrane, C. Falentin-Daudré, S. Ramtani, V. Migonney, *IRBM* **2018**, 39 (4), 268-278.
21. G. Amokrane, V. Humblot, E. Jubeli, N. Yagoubi, S. Ramtani, V. Migonney, C. Falentin-Daudré, *ACS Omega* **2019**, 4 (17), 17194-17208.
22. S. Berlot, Z. Aissaoui, G. Pavon-Djavid, J. Belleney, M. Jozefowicz, G. Hélar, V. Migonney, *Biomacromolecules* **2002**, 3, 1, 63-68.
23. H. Chouirfa, D.M. Evans, P. Bean, A. Saleh-Mghir, AC. Crémieux, D.G. Castner, C. Falentin-Daudré, V. Migonney, *ACS Applied Materials & Interfaces* **2018**, 10 (2), 1480-1491.
24. K. Kato, Y. Ikada, *Biotechnology and Bioengineering* **1995**, 47, 557-566.

25. G. Rohman, S. Huot, M. Vilas-Boas, G. Radu-Bostan, D.G. Castner, V. Migonney, *Journal of Materials science: Materials in Medecine* **2015**, 26:1–10.
26. J.H. Scofield, *Journal of Electron Spectroscopy and Related Phenomena* **1976**, 8, 129-137.
27. J. Kim, M. Chaudhury, M.J. Owen, *Journal of Colloid and Interface Science* **2000**, 226, 231-236.