

Surface Roughness Value Recommended for the Manufacture of Antibacterial Metal Surfaces: A Review

Rugosidad superficial recomendada en la manufactura de superficies metálicas antibacterianas: una revisión

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ABSTRACT

The manufacturing of antibacterial metal surfaces has been widely studied in the elaboration dental and orthopedic implants. Surface characteristics such as wettability, chemistry, electrostatics, and roughness have been described as factors for avoiding bacterial adhesion. However, surface roughness is still debated among authors regarding its effect on antibacterial surfaces. This paper reviews the existing literature to identify the recommended surface roughness values for metal implants to avoid bacterial adhesion, and it evaluates the different roughness parameters used in this regard. This compilation found no agreement when it comes to the exact roughness that a metal implant's surface should have to avoid bacterial adhesion and the subsequent formation of biofilms. In general, different authors recommend manufacturing smooth surfaces with a nanoscale roughness, smaller than the size of the target bacterium.

Keywords: bacterial adhesion, metal implant surface, surface roughness, bacterial biofilm

RESUMEN

La manufactura de superficies metálicas antibacterianas ha sido ampliamente estudiada en la elaboración de implantes ortopédicos y dentales. Ciertas características de las superficies, tales como mojabilidad, química, electrostática y rugosidad, han sido señaladas como factores para prevenir la adhesión bacteriana. Sin embargo, la rugosidad superficial aún se encuentra en debate en cuanto a su efecto en las superficies antibacterianas. Este artículo realiza una revisión de la literatura existente para identificar los valores de rugosidad superficial recomendados para evitar la adhesión bacteriana en implantes metálicos, y evalúa los distintos parámetros de rugosidad utilizados en este contexto. Esta compilación no encontró un consenso en términos de la rugosidad exacta que la superficie de un implante metálico debería tener para evitar la adhesión bacteriana y la posterior formación de *biofilms*. En términos generales, diferentes autores recomiendan manufacturar superficies con rugosidades de escala nanométrica, menores que el tamaño de la bacteria objetivo.

Palabras clave: adhesión bacteriana, superficie de implantes metálicos, rugosidad superficial, *biofilm* bacteriano

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Introduction

Bacterial surface adhesion has been widely studied for many years, seeking to understand and prevent possible future infections [1], [2]. Nosocomial infection, also called *hospital-acquired infection*, is "an infection acquired in hospital by a patient who was admitted for a reason other than that infection. An infection occurring in a patient in a hospital or other health care facility in whom the infection was not present..." [3, p. 1].

Every year, 1 to 5% of orthopedic implants end up with an infection [4]. The number one reason for persisting infections has been the formation of bacterial biofilms [5]. According to the reports, biofilms have been present in 60 to 80% of chronic infections [6], [7].

Treating this type of infection increases the cost of healthcare as well as the recovery times [6], [8], [9]. Among other factors, roughness, wettability, chemistry, and electrostatic charge influence the adhesion of bacteria to a surface. The

values of these factors differ if metal, polymer, or ceramic is used. Knowing the exact values of these factors helps to manufacture an antibacterial surface that reduces the patient's probability of having a nosocomial infection [2], [10].

This paper aims to review one of the factors that influence bacterial adhesion in metallic surfaces: surface roughness. It aims to summarize the recommended values for avoiding bacterial adhesion to orthopedic metal implants according to different authors.

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This article is divided into four sections. *What is biofilm?* elucidates the process of bacterial adhesion to the implant surface, the difficulty of eradicating it, and the role of surface roughness in it. *Surface roughness parameters used to measure bacterial adhesion* outlines various roughness parameters utilized for measuring surface roughness, and it discusses why specific parameters are more suitable than S_a to measure bacterial adhesion, as per the ISO 25178 – 2 standard [33]. *How do nano- and micro-roughness surfaces affect adhesion?* compares the findings of different authors regarding the surface roughness ranges that prevent bacterial adhesion. In this section, the lack of consensus on these values is addressed. The *Conclusions* section provides a comprehensive overview based on this review, offering a general recommendation for manufacturing antibacterial metal surfaces. By synthesizing the findings from various sections, valuable insights for creating effective antibacterial metal surfaces are presented.

What is biofilm?

Peri-implant infections are a particular type of infection produced by a biomaterial in the body. The microorganisms in these infections take advantage of the weak interface between the antibodies and the surface of the implant [11]-[13].

Because planktonic bacteria are associated with orthopedic implants, these infections create multicellular communities on the implant's surface, which are challenging to eradicate. Bacterial adhesion, in general terms, has five stages [5], [14]-[16]:

- *Stage one:* Planktonic bacteria adhere reversibly to the surface implant. This adhesion depends on characteristics such as the surface material, hydrodynamics, electrostatic interactions, surface roughness, etc. [1].
- *Stage two:* The adhered bacteria send information to the nearby bacteria and secrete proteins with adhesive components, which makes the adhesion irreversible in a few hours [1].
- *Stage three:* The bacterial community colonizes the surface and creates a biofilm matrix that will encapsulate the bacteria. This results in a complex structure which will cover the colony [1].
- *Stage four:* The bacterial colony starts to secrete extracellular polymeric substances (EPS) and develops a complex microorganism known as a *mature biofilm*. This biofilm consists mainly of bacteria, polysaccharides, proteins, and DNA. In this last stage, some ducts are created to spare more bacteria, spreading the infection to different body parts [1].

Preventing bacterial adhesion and anchorage to the implant's surface is crucial for preventing further infections [17]. Biofilms protect bacteria from the immune system and antibiotics, and, once they have created a biofilm, treating the infection with antibiotics becomes harder [1]. It has

been found that the dose of antibiotics needed to treat some bacteria in biofilms is 1 000 times higher than that needed in the first adhesion stage [14], [18]. Many authors such as [19] and [20] have stated that modifying the implant surface, either chemically or physically, may lead to antibacterial surfaces.

Regarding the physical modification of the surface, micro- and nano-topographies are essential in creating antibacterial surfaces. However, the exact values are still unknown [21]-[23].

Surface roughness parameters used to measure bacterial adhesion

Surface roughness is the way in which the topography of a surface can be described in terms of how high the peaks are and how deep the valleys are on a surface after manufacturing. Surface roughness is just one of the many characteristics that influence bacterial adhesion [24]-[28].

Surface roughness can be measured with different parameters: profile roughness parameters (R_a , R_q ...) calculated in a profile (line), and area roughness parameters (S_a , S_q ...) calculated in a completed area. Profile parameters, especially R_a , are the most commonly reported, but they cannot describe the surface thoroughly, providing a reduced portion of surface information [26]-[28].

Area roughness parameters better describe the surface as a whole. Some parameters can provide specific information about the surface, and their use depends on the application [29]-[32]. The surface roughness parameters described below are reported in the ISO 25178 – 2 standard and are used to measure surface roughness for antibacterial adhesion [33].

S_a – Arithmetic mean deviation: This parameter is the arithmetic mean of an absolute value corresponding to the height of the sampling area. This parameter does not adequately describe the surface topography because different surfaces with different peaks and valleys can have the same S_a .

S_q – Squared mean height: This parameter is similar to S_a and is related to the surface energy.

S_{sk} – Asymmetry: This parameter describes the distribution of peaks of the topography. The height difference is distributed uniformly if a surface has a $S_{sk} = 0$.

S_{ku} – Kurtosis: This parameter describes the spikiness of the surfaces if the peaks are uniformly distributed.

S_{dr} – Surface area ratio: This is one of the most important parameters to describe an antibacterial surface. A surface with a $S_{dr} = 0$ is ideally 'smooth'. This parameter can also identify the differences between two surfaces with the same S_a .

The measurement of these parameters is affected by the equipment used. Some studies describe how contact and no-contact equipment affect measurements, so this criterion should be considered in future research [32], [34].

How do nano- and micro-roughness surfaces affect adhesion?

Surface roughness has been widely studied with the aim of preventing bacterial adhesion to metal surfaces. Many authors have described the roughness values found to be the most appropriate in this regard. However, they disagree on each type of topography [13], [35].

Some authors have stated that some surface feature patterns potentially determine the bacteria's early location and adhesion. In this vein, nanoscale topographies with an organized pattern reduce bacterial adhesion by 40-95% [36], [37].

In a study on bacterial adhesion to titanium nitride, [38] found that surfaces with a roughness lower than 0,066 μm had better anti-adhesion properties. Other studies such as [39] argue that the adhesion of bacteria is minimal if the surface has a roughness value or Ra of 160 nm. This study evaluated stainless steel surfaces with Ra between 30 and 890 nm. A different study [41] evaluated five types of surfaces with Ra between 172,5 and 45,2 nm, finding that bacterial adhesion was restrained on the nanoscale surface.

While investigating different polishing surfaces, [41] found that bacteria had less adhesion in metal surfaces with higher porosity. The authors found increased bacterial adhesion in surfaces with a Ra smaller than 0,2 μm . In another study, no relation between superficial roughness and bacterial adhesion could be found [42].

In contrast, the authors of [43] argue that there is a negative correlation between bacterial adhesion and metal surfaces with a roughness of less than 6 nm. They found that bacteria prefer a smoother surface under static culture conditions if the roughness is smaller than 0,23-6,13 nm. When the roughness is more significant than 6-30 nm, bacteria prefer surfaces with higher roughness values. The authors also found that a higher roughness does not necessarily benefit adhesion; it will increase the production of EPS to create a biofilm. It will help protect the bacteria because deep valleys trap them and provide cover from shear force [16].

On the other hand, some authors such as [44], [45], and [46], among others, have stated that nano-topography has an antibacterial effect, as peaks with sizes similar to those of the bacteria exert strain forces on the bacterial membrane, causing it to break. This phenomenon is known as the *cicada wings effect*.

[43] suggests that increasing the surface roughness more significantly than 1,24 μm will increase bacterial adhesion. [47] support this idea, stating that surfaces with a roughness

higher than 0,4 μm increase the risk of peri-implant diseases. They support the conclusions of [48], i.e., metal surfaces of 130-360 nm are desired in avoiding bacterial adhesion. A similar idea is shared by [49]: surfaces with flaws such as grooves, gaps, or cracks provide a favorable environment for bacteria [39]. These authors suggest that bacteria adhere to surfaces that correspond to their size since this maximizes their contact area [50]-[52].

[20] suggest that the adhesion of the bacterium *Staphylococcus aureus* decreases by using surfaces with a roughness value under 1,51 nm and a unidirectional surface texture. Some related results were obtained [53]. In this study, the adhesion of *Escherichia coli* and *Staphylococcus aureus* was reduced by 55,6 and 40,5%, respectively, by using a titanium surface smaller than 6 nm (surface peak density). The study by [15] with the same bacteria found similar results using pattern surfaces of 1 μm . A similar result with the same value of roughness was found by [54].

As is the case of [55], some authors agree that, despite the vast amount of works, there is not enough information about the adequate topography. However, in general, the best approximation would be a nanoscale surface roughness with a structure similar structure to that of bone tissue [56].

In [57], by analyzing the way in which bacterial adhesion is evaluated, the authors found that using parameters such as Ra may not be the most accurate approach. Ra is an arithmetic mean or the absolute value of the vertical deviation from the mean line of the profile [31], [58], [59]. In other words, Ra is described as the arithmetic mean deviation of an assessed profile, which is the average of the peaks and valleys of the surface with regard to a centerline [35]. According to this, there could be different surfaces with the same Ra and significant differences in peaks and valleys [60]. Therefore, other superficial roughness parameters should be considered in future research, such as Rq, Rsk, or even area roughness parameters such as Sq, Sq, Ssk, and Sku [61].

Other works [61], [66] have used Sq, Ssk, and Sku to measure surface roughness, finding, for instance, that *Staphylococcus epidermis* creates biofilms on surfaces with high Ssk values and that smoother surfaces have the best antibacterial properties. Despite not providing an exact value for the surface parameters to create antibacterial surfaces, the findings are consistent with the idea that a surface with low peaks and valleys hinders the adhesion of bacteria.

Moreover, bacteria will avoid surfaces with a roughness smaller than their size, as this increases their contact area, which can damage the bacterial membrane [46], [66].

Conclusions

According to this review, there are two things to consider while creating antibacterial surfaces.

Table I. Different roughness values suggested by different authors to create antibacterial surfaces

Authors	Title	Results
Songze Wu et al. [18]	Influence of surface topography on bacterial adhesion	The height of the peaks on the topography should not be greater than the flagella size. The adequate roughness should be $R_a > 172,5-45,2$ nm or $R_q > 217,9-56,6$ nm.
Kurup et al. [38]	Surface modification techniques of titanium and titanium alloys for biomedical, dental applications	Surfaces with a roughness lower than $0,066 \mu\text{m}$ had better anti-adhesion properties than treated ones.
Vadillo-Rodríguez et al. [37, p. 347]	Bacterial response to spatially organized microtopographic surface patterns with nanometers scale roughness	"Surfaces with shorter or less deep features than $21,1$ nm inhibited the growth of the bacteria..."
Costa et al. [63, p. 4]	Fitting pieces into the puzzle: The impact of titanium-based dental implant surface modifications on bacterial accumulation and polymicrobial infections	"Nanostructured surfaces have been demonstrated to be more efficient at reducing bacterial attachment..." Roughness values of $R_a = 0,51-1,36 \mu\text{m}$ or $S_a = 0,66-2,91 \mu\text{m}$ have no antibacterial effects, although they induce osteoblast adhesion.
Yuan et al. [14, p. 53]	Bacterial anti-adhesion surface design: surface patterning, roughness, and wettability: A review	A R_a smaller than 6 nm has a negative correlation with bacterial adhesion. If the roughness is low ($R_a: 0,23-6,13$ nm), "... bacteria prefer to adhere on the smoother surface under static culture conditions and the increased production of EPS to create a biofilm..." When the roughness has a value between 6 and 30 nm, bacteria prefer to adhere to the rougher surface.
Dantas et al. [47, p. 2]	Bacteria co-culture adhesion on different texturized zirconia surfaces	"Roughness surfaces higher than $0,4 \mu\text{m}$ increase the affinity of microorganisms and the risk of peri-implant diseases..." Surfaces between 130 and 360 nm are more likely to be antibacterial surfaces.
Lu et al. [20]	Effects of surface roughness and texture on the bacterial adhesion on the bearing surface of bio-ceramic joint implants: An <i>in vitro</i> study	The R_a to decrease the number of adherent bacteria (<i>Staphylococcus aureus</i>) should be under $1,51$ nm.
Filipović et al. [49, p. 5]	Bacterial adhesion on orthopedic implants	"A tiny increase in surface roughness ($R_a = 0,04-1,24 \mu\text{m}$) resulted in a pronounced increase of bacteria..." Bacteria adhere to surfaces similar in size since this allows them to maximize the contact area.
Annunziata et al. [64]	Bacterial adhesion to direct laser metal formed and mildly acid-etched implants surfaces	Roughness should be less than $0,2 \mu\text{m}$. The texture should be $S_a < 0,5 \mu\text{m}$.
Lüdecke et al. [53]	Nanorough titanium surface adhesion of <i>Escherichia coli</i> and <i>Staphylococcus aureus</i> via nano adhesion points	<i>Escherichia coli</i> and <i>Staphylococcus aureus</i> adhesion was reduced by $55,6$ and $40,5\%$, respectively, by using a titanium surface smaller than 6 nm (surface peak density).
Yang et al. [15]	Control of bacterial adhesion and growth on honeycomb-like patterned surfaces	<i>Escherichia coli</i> and <i>Staphylococcus aureus</i> , in patterns of R_a between 1 and $10 \mu\text{m}$, showed bacterial growth inhibited to a greater extend than on a flat surface.
Lüdecke et al. [54 p. 587]	Physical vapor-deposited titanium thin films for biomedical applications: Reproducibility of nanoscale surface roughness and microbial adhesion properties	" <i>Escherichia coli</i> adhesion decreased from approximately 23% to 19% with increasing surface nano roughness from 2nm to 6 nm..."

- Bacteria have fewer probabilities of initial adhesion and biofilm formation in surfaces with nanoscale roughness.
- Bacteria have fewer odds of adhesion in surfaces with a roughness value less than their size.

In conclusion, antibacterial surfaces should be smooth, avoiding high peaks and valleys where the bacteria could be protected from shear forces. The surface roughness should be smaller than the bacteria's size. This parameter depends on the type of bacteria studied, but *Escherichia coli* and *Staphylococcus aureus* can serve as a reference since they are the most common in nosocomial infections. The sizes of these two bacteria are $1-5$ and $1 \mu\text{m}$ in diameter, respectively.

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Conflicts of interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

CRedit author statement

All authors: conceptualization, methodology, software, validation, formal analysis, investigation, writing (original draft, writing, review, and editing), data curation.

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