ORIGINAL CONTRIBUTION

Escherichia coli adhesion to surfaces–a thermodynamic assessment

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Abstract Several studies have tried to correlate bacterial adhesion with the physicochemical properties of the surface with limited success. Most often, the obtained correlations seem to be only applicable to a particular set of experimental conditions making it difficult to obtain guidelines for the design of antibiofouling surfaces. The ratio between Lifshitz van der Waals apolar component and the electron donor component ($\gamma^{LW}\!/\!\gamma^-\!)$ was recently shown to correlate with bacterial adhesion to the surfaces of ship hulls and heat exchangers. In this work, four materials with biomedical application (polystyrene, poly-L-lactide, cellulose acetate, and polydimethylsiloxane) and glass were characterized and Escherichia coli adhesion to those materials was assayed with a parallel-plate flow chamber operating in physiological shear stress conditions. Adhesion was correlated with the γ^{LW}/γ^{-1} ratio, further extending the application range tested on the original study. Additionally, results from other studies were also evaluated to confirm the applicability of this correlation to other surfaces, microorganisms, and experimental conditions. Results show that bacterial adhesion is reduced in surfaces with lower γ^{LW}/γ^{-} and enhanced otherwise. This finding may be helpful in the design of new coatings by controlling γ^{LW}/γ^{-} or in the selection of existing materials according to the desired application.

Keywords Adhesion · *Escherichia coli* · Surface properties · Thermodynamics · Contact angle

Introduction

Microorganisms have a natural tendency to adhere to surfaces and form biofilms [1]. Beneficial biofilms can be found in bioremediation processes, wastewater treatment, and in the production of various chemicals [2, 3]. However, bacterial adhesion and subsequent biofilm growth is a common problem in industry since it can lead to food spoilage by bioconversion or efficiency loss in heat exchangers [4, 5]. In the biomedical field, biofilms are responsible for many infections in humans [6] and can cause deterioration of the functionality of medical devices [7]. Therefore, in industry, inhibiting or delaying the onset of detrimental biofilms can represent a reduction in operational costs, since fewer stops are required for sanitation [4, 8]. In the biomedical field, delaying the onset of biofilms in medical devices may reduce the need for antimicrobial treatment and the costs associated with the replacement of infected implants during revision surgery, which may triple the cost of the primary implant procedure [9].

Researchers all over the world are trying to understand bacterial adhesion in order to inhibit or promote biofilm development [10, 11]. Several strategies have been evaluated in order to control biofilm development [9, 12, 13] and one of the most promising is to control bacterial adhesion [8, 14–17].

Bacterial adhesion begins with the attraction between cells and surfaces, followed by adsorption and attachment [18]. The physicochemical forces involved in the initial approach of cells to surfaces are primarily van der Waals, electrostatic, hydration, and hydrophobic interactions [18]. Therefore, the correct selection of materials to be used in industrial and biomedical settings can be determinant to the onset of bacterial biofilms on these surfaces.

Researchers are trying to define criteria for selection of new materials according to their surface properties [16, 17, 19]. This methodology has been used intensively since accessible and fast methods such as contact angle measurements are

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available, enabling time and cost reduction in the laboratory [20-22]. However, finding a correlation between surface properties and bacterial adhesion rates has been challenging [23–25]. Li and Logan [26] studied the contribution of surface charge and hydrophobicity on the adhesion of three Escherichia coli strains, two Pseudomonas aeruginosa strains, and two Burkholderia cepacia strains on metal oxide-coated and uncoated glass surfaces. These authors observed that adhesion was not significantly correlated with bacterial charge and contact angle. Liu and Zhao [27] used the ratio between apolar Lifshitz van der Waals components (γ^{LW}) and electron donor components (γ^{-}) of modified stainless steel (Ni-P-TiO2-PTFE nanocomposite coatings) as a surface property parameter to correlate with Pseudomonas fluorescens, Cobetia marina, and Vibrio alginolyticus adhesion under static and dynamic conditions. Their results demonstrated that coatings with the lowest γ^{LW}/γ^{-} had the lowest bacterial adhesion values, and increasing γ^{LW}/γ^{-1} led to higher bacterial adhesion. That study was conducted with surfaces that may be used in ship hulls and heat exchangers but the authors suggested that their results are transferable to the biomedical field. This hypothesis was tested on this work by using four polymeric surfaces (polystyrene (PS), poly-Llactide (PLLA), cellulose acetate (CA), and polydimethylsiloxane (PDMS)) which can be used in biomedical devices in the human body [18, 28-30] and glass. Thermodynamic surface properties were evaluated in order to find if they could be correlated with bacterial adhesion. The hydrodynamic conditions used are similar to those found in the bladder, urinary tract, and reproductive system [31, 32] where biomedical devices constructed with the selected materials are used [28, 29, 33, 34] and where E. coli is the major cause for infection [35, 36]. These surfaces were also selected due to their different γ^{LW}/γ^{-} values which extend the range tested by Liu and Zhao [27]. The applicability of this correlation was also tested using data from other authors studying bacterial adhesion or protein adsorption to different materials (soil minerals, synthetic materials, plasma-treated surfaces, and metallic materials) in different systems and operational conditions. Thus, the rationale for this work was to find out a selection/design criteria to predict bacterial adhesion to materials used in the industrial and biomedical fields.

Materials and methods

Bacteria and culture conditions

A starter culture of *E. coli* JM109(DE3) was obtained by inoculation of 500 μ L of a glycerol stock (kept at -80 °C) to a total volume of 0.2 L of inoculation media with 5.5 g L⁻¹ glucose, 2.5 g L⁻¹ peptone, and 1.25 g L⁻¹ yeast extract in phosphate buffer (1.88 g L⁻¹ KH₂PO₄ and 2.60 g L⁻¹

Na₂HPO₄) at pH 7.0 [37]. This culture was grown in a 1-L shake-flask, incubated overnight at 30 °C with orbital agitation (120 rpm). A volume of 60 mL from the overnight grown culture was used to harvest cells by centrifugation (10 min, 3202*g*). Cells were washed twice with citrate buffer 0.05 M [38], pH 5.0 and the pellet was resuspended and diluted in the same buffer in order to reach a cell concentration of 7.6×10^7 cell mL⁻¹.

Surface preparation

Five materials, PS, glass, PLLA, CA, and PDMS, were prepared for adhesion assays. PS surface and microscope glass slides (VWR) were firstly washed with a commercial detergent (Sonasol Pril, Henkel Ibérica S A) and immersed in sodium hypochlorite (3 %). After rinsing with distilled water, part of the microscope glass slides was coated with the polymers. These were prepared by mixing the polymer in solid form with solvents. Dichloromethane was added to PLLA at 5 % (w/w), acetone was added to CA at 8 % (w/w), and a curing agent (Sylgard 184 Part B, Dow Corning) was added to PDMS (at a 1:10 ratio) (polymers from Sigma, solvents from Normapur). This mixture was prepared in a beaker where it was manually stirred with a glass rod to homogenize the two components without introducing bubbles. The polymers were then deposited as a thin layer on top of glass slides by spin coating (Spin150 PolosTM), for PDMS at 2000 rpm for 60 s and for the other surfaces at 5000 rpm for 50 s.

Surface characterization

The surface charge of bacteria and material surfaces was characterized by zeta potential and surface hydrophobicity using the contact angle method. One E. coli suspension was prepared as described before, and particle suspensions of each material [39] were also prepared in order to measure the electrophoretic mobility, using a Nano Zetasizer (Malvern Instruments, UK). The hydrophobicity of bacteria and surfaces was evaluated considering the Lifshitz van der Waals acid base approach [40]. Contact angles were determined automatically by the sessile drop method in a contact angle meter model (OCA 15 Plus; Dataphysics, Filderstadt, Germany) using water, formamide, and α bromonaphtalene (Sigma) as reference liquids with surface tension components taken from literature [41]. For each surface (PLLA, PS, CA, PDMS, and glass), at least 10 measurements with each liquid were performed at 25±2 °C. One E. coli suspension was prepared in the same conditions as for the adhesion assay and its physicochemical properties were also determined by sessile drop contact angle measurement as described by Busscher et al. [42].

According to van Oss [40], the total surface energy (γ^{Tot}) of a pure substance is the sum of the apolar Lifshitz van der Waals components of the surface free energy (γ^{LW}) and polar

$$\gamma^{\rm TOT} = \gamma^{\rm LW} + \gamma^{\rm AB} \tag{1}$$

The polar AB component comprises the electron acceptor γ^+ and electron donor γ^- parameters, and is given by:

$$\gamma^{\rm AB} = 2\sqrt{\gamma^+ \gamma^-} \tag{2}$$

The surface energy components of a solid or bacterial surface (s) are obtained by measuring the contact angles (θ) with the three different liquids (l) with known surface tension components, followed by the simultaneous resolution of three equations of the type:

$$(1 + \cos\theta)\gamma_{\rm l} = 2\left(\sqrt{\gamma_{\rm s}^{\rm LW}\gamma_{\rm l}^{\rm LW}} + \sqrt{\gamma_{\rm s}^{+}\gamma_{\rm l}^{-}} + \sqrt{\gamma_{\rm s}^{-}\gamma_{\rm l}^{+}}\right) \quad (3)$$

The degree of hydrophobicity of a given surface (solid and bacterial surface) is expressed as the free energy of interaction

 $(\Delta G \text{ mJ m}^{-2})$ between two entities of that surface immersed in polar liquid (such as water (w) as a model solvent).

If the interaction between the two entities is stronger than the interaction of each entity with water, $\Delta G < 0 \text{ mJ m}^{-2}$, the material is considered hydrophobic, if $\Delta G > 0 \text{ mJ m}^{-2}$, the material is hydrophilic. ΔG was calculated from the surface tension components of the interacting entities, using the equation:

$$\Delta G = -2 \left(\sqrt{\gamma_s^{LW}} - \sqrt{\gamma_w^{LW}} \right)^2 + 4 \left(\sqrt{\gamma_s^+ \gamma_w^-} + \sqrt{\gamma_s^- \gamma_w^+} - \sqrt{\gamma_s^+ \gamma_s^-} - \sqrt{\gamma_w^+ \gamma_w^-} \right); \quad (4)$$

When studying the interaction (free energy of adhesion) between surface (s) and bacteria (b) that are immersed in water, the total interaction energy, ΔG^{Adh} , can be expressed as:

$$\Delta \mathbf{G}^{\mathrm{Adh}} = \gamma_{\mathrm{sb}}^{\mathrm{LW}} - \gamma_{\mathrm{sw}}^{\mathrm{LW}} - \gamma_{\mathrm{bw}}^{\mathrm{LW}} + 2 \left[\sqrt{\gamma_{\mathrm{w}}^{\mathrm{+}}} \left(\sqrt{\gamma_{\mathrm{s}}^{\mathrm{-}}} + \sqrt{\gamma_{b}^{\mathrm{-}}} - \sqrt{\gamma_{\mathrm{w}}^{\mathrm{-}}} \right) + \sqrt{\gamma_{\mathrm{w}}^{\mathrm{-}}} \left(\sqrt{\gamma_{\mathrm{s}}^{\mathrm{+}}} + \sqrt{\gamma_{b}^{\mathrm{+}}} - \sqrt{\gamma_{\mathrm{w}}^{\mathrm{+}}} \right) - \sqrt{\gamma_{\mathrm{s}}^{\mathrm{+}}} \gamma_{b}^{\mathrm{-}} - \sqrt{\gamma_{\mathrm{s}}^{\mathrm{+}}} \right]$$
(5)

Thermodynamically, if $\Delta G^{Adh} < 0$ mJ m⁻² adhesion is favored, while adhesion is not expected to occur if $\Delta G^{Adh} > 0$ mJ m⁻².

Flow chamber experiments

A parallel-plate flow chamber (PPFC) with dimensions of 25.4×1.6×0.8 cm (L×W×H) was connected to a centrifugal pump by a tubing system. It contained a bottom and a top opening at the exit for the introduction of the test surfaces. The PPFC was mounted in a microscope (Nikon Eclipse LV100, Japan) to monitor E. coli attachment to each surface for 30 min. The cellular suspension was circulated at 2 mL s^{-1} and images were acquired with a camera (Nikon digital sight DS-RI 1, Japan) connected to the microscope. The hydrodynamic conditions were simulated by computational fluid dynamics and the results have shown that in the viewing point, the conditions are of steady flow and the average shear stress was of 0.01 Pa (not shown). Approximate shear stresses can be found in the bladder, urinary tract, and reproductive system [31, 32]. Temperature was kept constant at 37 °C using a recirculating water bath. All adhesion experiments were performed in triplicate for each surface.

The microscopy images recorded during the cell adhesion assays were analyzed with the program ImageJ (v1.46r). The number of adhered cells after 30 min was then divided by the surface area of the field of view to obtain the density of bacteria per square centimeter. Statistical analysis

Paired *t* test analyses were performed to estimate whether or not there was a significant difference between the results obtained on each surface. Results were evaluated individually using the three independent results obtained with one surface and the three individual results obtained with other surface. Results were considered statistically different when a confidence level greater than 95 % was reached (P<0.05). Standard deviation between the three values obtained from the independent experiments was also calculated.

Re-plotted data

Relevant works, where some authors had tried to find a correlation between surface properties of different materials and bacterial adhesion (as well as protein adsorption to those surfaces), were selected, and data was re-plotted in this work in order to compare with the new data here presented. Bacterial adhesion and protein adsorption data were represented as a function of the ratio between the Lifshitz van der Waals component and the Lewis acid–base electron donor γ^- component (γ^{LW}/γ^-) for each tested surface.

Results and discussion

In this work, five materials (PLLA, PDMS, PS, CA, and glass) were tested in order to evaluate *E. coli* adhesion after

Surface	Contact angle/(⁰) Water F	<u>(</u>		Surface nrone	irties			E. coli - surface interaction		Zeta potential/mV	Adhered cells cm z	í
Surface				surrace properties						•		4
		Formamide	lpha-Bromonaphtalene	$\gamma^{LW/} (mJ m^{-2})$	$\gamma^{+/}$ (mJ m $^{-2}$)	$\gamma^{-/}$ (mJ m ⁻²)	$\Delta G/$ (mJ m ⁻²)	$\Delta G A dh/(m J m^{-2})$	²)			
PLLA	88.03±1.01 6	68.49 ± 0.95	25.59 ± 1.54	40.15	0.000	4.374	-65.32	29.90		-27.90	$1.82 \times 10^{6} \pm 2.76 \times 10^{4}$	10^{4}
PDMS	113.6 ± 0.62	111.2 ± 0.61	87.62 ± 1.77	12.04	0.000	4.544	-61.82	32.60		-29.30	$1.29 \times 10^{6} \pm 3.79 \times 10^{5}$	10^{5}
PS	80.81±0.68	64.33 ± 1.24	24.64 ± 1.11	40.45	0.000	8.290	-49.56	37.80		-29.80	$1.36 \times 10^{6} \pm 1.35 \times 10^{5}$	10^{5}
CA	65.24±0.49 3	36.63 ± 2.05	22.47 ± 1.05	41.09	1.441	9.629	-37.58	25.50		-23.40	$1.35 \times 10^{6} \pm 1.32 \times 10^{5}$	10^{5}
Glass	16.38±0.35	17.19 ± 0.35	44.48 ± 0.71	32.59	2.586	52.43	27.99	62.90		-37.00	$1.18 \times 10^{6} \pm 7.47 \times 10^{4}$	10^{4}
E. coli	19.13±0.88 7	73.34 ± 0.65	58.54 ± 2.01	25.71	0.000	123.2	121.90	n/a		-17.00	n/a	
Organism	Organism/compound	Surface	Surface material	Platform		T/∘C	Hydrodynamics	ics	Assay time/h	h Correlated parameter	ameter Reference	nce
Bacillus subtilis	subtilis	Soil minerals	herals	Conical flask		25	Shaking at 1.2 g	2 g	2	SESA	[49]	
Staphylou	Staphylococcus epidermis		Helium plasma-treated PET	Well-tissue cu	Well-tissue culture plates and	37	Shear rate 5, 2	Shear rate 5, 50, and 200 s^{-1}	2.5	$\Delta G^{\rm Adh}$	[50]	
<i>Listeria r.</i> Bovine se	<i>Listeria monocytogenes</i> Bovine serum albumine	Synthetic	ic	a raural rrow Capped bottles	e chamber	37	Gentle shaking	lg	24 and 1 ^a	Surface chemistry	stry [51]	
Cytochrome c	me c											
Pseudomonas fl Cobetia marina Vihrio aleinolyt	Pseudomonas fluorescens Cobetia marina Vibrio alginolyticus	Ni	Ni-P coatings with TiO ₂ and PTFE, stainless steel	Static tank ar	Static tank and dynamic PPFC	28	Static, dynamic–shear stress 0.98, 0.46, and 0.21 mPa	nic–shear , 0.46, and	6 and 24 ^b	γ^{LW}/γ^{-}	[27, 52]	[2
Escherichia coli	iia coli	Polymeı	Polymeric coatings, glass	PPFC		37	Shear stress 0.01 Pa	.01 Pa	0.5	$\gamma^{\rm LW}/\gamma^-$	This work	rork

SESA specific external surface area, PET polyethylene terephthalate

^a Referent to microorganism adhesion and proteins adsorption, respectively

^b Referent to static and dynamic conditions, respectively

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determination of thermodynamic surface properties. Table 1 shows the contact angle measurements for each surface, the thermodynamic surface energy properties, the zeta potential values, and the cell adhesion results.

Based on contact angle values, surfaces can be classified into hydrophilic or hydrophobic if the contact angle of water with the surfaces is, respectively, lower or higher than 65° [43]. From the results in Table 1, it is possible to anticipate that glass and E. coli have hydrophilic surfaces and the other surfaces are hydrophobic. Regarding the values determined for the van der Waals forces apolar component (γ^{LW}) [44], it is possible to observe that CA has the highest attractive apolar component value and PDMS the lowest. In what concerns the polar surface components (γ^{-}, γ^{+}) , results showed that PLLA, PDMS, PS, and E. coli are monopolar surfaces, being electron donors (Table 1). Conversely, CA and glass are polar surfaces, being electron donors and acceptors. From the total free energy results, it is also possible to observe that PLLA, PDMS, PS, and CA are hydrophobic surfaces ($\Delta G <$ 0 mJ m⁻²) whereas glass and *E. coli* are hydrophilic (ΔG > 0 mJ m^{-2}). Therefore, results obtained with the determination of surface properties support the preliminary evaluation made by water contact angle measurement.

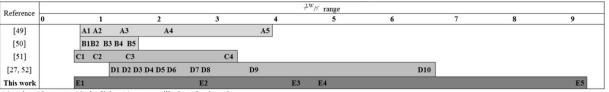
From the cell adhesion results (Table 1), it is possible to observe that a higher number of adhered cells was obtained on the PLLA surface (the most hydrophobic) and a lower bacterial adhesion value was observed on glass (P<0.05) (the most hydrophilic). Previous studies have shown that *E. coli* adhesion is enhanced in hydrophobic surfaces and decreased in hydrophilic materials [45, 46]. However, if hydrophobicity was the only relevant factor, an increase in the ΔG values should have led to a consistent decrease in bacterial adhesion and this was not observed for PDMS. Thus, a correlation between surface hydrophobicity and bacterial adhesion was not found.

The thermodynamic theory indicates that a system with a lower interacting energy (ΔG^{Adh}) usually leads to a higher affinity between bacteria and surfaces [21]. Therefore, based on the results in Table 1, *E. coli* should have adhered more to CA and PLLA and have a lower affinity to glass. Thus, it seems that cell adhesion is also not directly correlated with

 ΔG^{Adh} . Other authors have also tried to find a correlation between bacterial adhesion and surface hydrophobicity or surface free energy of adhesion without success. In a study by Oliveira et al. [24], a correlation between the hydrophobicity of materials (polyethylene, polypropylene, and granite) used in kitchens and the adhesion of four *Salmonella enteritidis* strains was also not found. Barton et al. [47] were also not successful in finding a correlation between the free energy of adhesion of orthopedic implant polymers (poly(orthoester), poly(L-lactic acid), polysulfone, polyethylene, and poly(ether-ether ketone)) and *S. epidermidis* or *E. coli* adhesion.

In this work, a correlation between electron donor character (γ^{-}) and bacterial adhesion was also not observed particularly for glass which showed a very high value of γ^{-} (52.43 mJ m⁻²) compared to the other surfaces (Table 1). Additionally, for the zeta potential data, negative values indicate electrical repulsion between negative-charged bacteria and surfaces [48] but a correlation was not found for this parameter either.

Several studies have been performed by other research groups in order to find a good correlation between bacterial adhesion (and adsorption of organic/inorganic particles) and some physicochemical parameter from the surface. A literature survey was performed in order to find such works where complete information about the thermodynamic properties was included or where these properties could be calculated from reported data (Table 2). Hong et al. [49] studied the role of surface properties in the adhesion of Bacillus subtilis to soil minerals. These authors observed a significant correlation between adhesion capacity and the specific external surface area of the minerals, but they did not find a correlation between surface hydrophobicity (ranging from -32.2 to 33.2 mJ m^{-2}) and adhesion. Katsikogianni et al. [50] studied the role of the free energy of adhesion (from -10.5 to 17.2 mJ m^{-2}) in the attachment of *Staphylococcus epidermidis* to plasma-modified PET films under quasi-static (5 s^{-1}) and dynamic conditions (50 and 200 s^{-1}). A strong correlation between the thermodynamic predictions and the measured values of bacterial adhesion under guasi-static conditions was observed. Moreover, the authors reported that the polar



A1 - mica, A2 - quartz, A3 - kaolinite, A4 - montmorillonite, A5 - simessite B1 - 1 h helium plasma treated polyethylene terephthalate (PET), B2 - 8 days helium plasma treated PET, B3 - 17 days helium plasma treated PET, B4 - 58 days helium plasma treated PET, B5 -

B1 - 1 h helium plasma tretaed polyethylene terephtha 30 days helium plasma treated PET

C1 -SiO-(CH₂)₂NH-CO-PEO-5000-OCH₃ C2 - SiO-(CH₂)₂NH-CO-NH₃, C3- SiO-(CH₂)₂NH-CO-PNMeAm, C4 - SiO-(CH₂)₂NH-CO-(CF₂)CF₃

D1 - Ni-P-TiO2-PTFE 3, D2 - Ni-P-TiO2-PTFE 4, D3 - Ni-P-TiO2-PTFE 1, D4 - Ni-PPTFE 3, D5 - Ni-P-PTFE, D6 - Ni-P-TiO2-PTFE 2, D7 - Ni-P-PTFE 1, D8 - Ni-P-PTFE 2, D9 - Ni - P, D10 - stainless steel

E1 - glass, E2 - polydimethylsiloxane, E3 - cellulose acetate, E4 - polystyrene, E5 - poly-L-lactide

Fig. 1 Surfaces used and γ^{LW}/γ^{-} tested in different works attempting to find a correlation between adhesion and thermodynamic properties

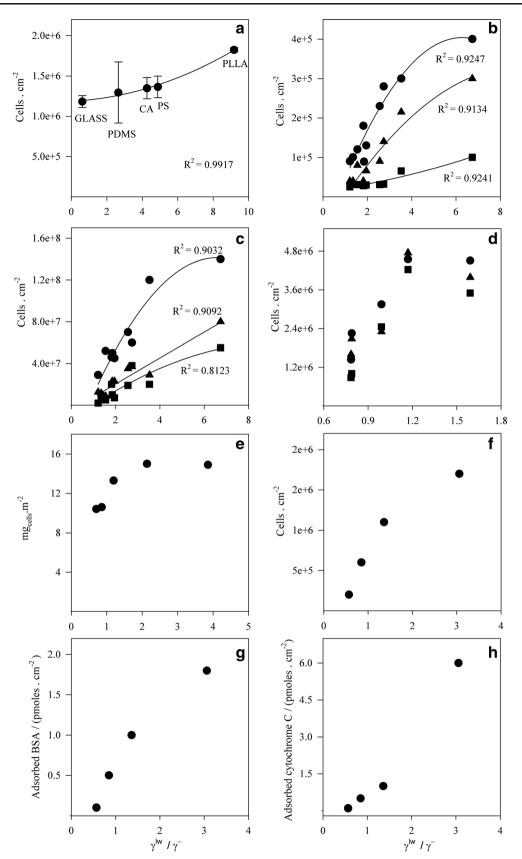


Fig. 2 Relationship between bacterial adhesion or protein adsorption and the ratio between apolar Lifshitz van der Waals components (γ^{L}) and electron donor component (γ^{-}). **a** *E*. *coli* adhesion on polymeric and glass surfaces. b Vibrio (circle), Cobetia (triangle), and P. fluorescens (square) adhesion on Ni-P coatings with TiO2 and PTFE and stainless steel, replotted from Liu and Zhao [27]. c Vibrio adhesion at 0.21 (circle), 0.46 (triangle), and 0.98 (square) mPa on Ni-P coatings with TiO₂ and PTFE and stainless steel, re-plotted from Liu and Zhao [27]. d Staphylococcus epidermis adhesion at 5 (circle), 50 (triangle), and 200 s^{-1} (square) on helium plasma-treated PET, re-plotted from Katsikogianni et al. [50]. e Bacillus subtilis adhesion on soil minerals, re-plotted from Hong et al. [49]. f Listeria monocytogenes adhesion on synthetic surfaces, re-plotted from Cunliffe et al. [51]. g Bovine serum albumin adsorption on synthetic surfaces, re-plotted from Cunliffe et al. [51]. h Cytochrome c adsorption on synthetic surfaces, re-plotted from Cunliffe et al. [51]. Whenever a correlation was reported by the original authors, it was also represented in this figure and the correlation factor (R^2) is indicated (**a**, **b**, and **c**)

acid-base interactions dominated the interactions of bacteria with the substrates in aqueous media. However, under flow conditions, the increase in the shear rate reduced the predictability of the thermodynamic models. Cunliffe et al. [51] used synthetic materials with energies ranging from 15 to 42 mJ m^{-2} for bacterial adhesion and adsorption of bovine serum albumin (with a net negative charge) and cytochrome c (with a positive charge). Protein adsorption and Listeria monocytogenes adhesion also showed some correlation with the chemistry of the surfaces. Liu and Zhao [27] have suggested a ratio between Lifshitz van der Waals apolar component and the electron donor component (γ^{LW}/γ^{-}) as a good correlation factor for cell adhesion. These authors have used P. fluorescens, C. marina, and V. alginolyticus and Ni-P-TiO₂-PTFE coatings in different hydrodynamic conditions (Table 2). This ratio was also tested for the adhesion values obtained in the present work as well as for the results reported by other groups comprising 29 different surfaces, 7 organisms, 2 proteins, and different shear stress conditions (Table 2). The (γ^{LW}/γ^{-}) range covered in each study as well as the identification of the tested surfaces is provided in Fig. 1.

In the present work, surfaces with the highest γ^{LW}/γ^{-1} values had the highest bacterial adhesion (Fig. 2a). This may be due to a lower surface electron donor component (γ^{-1} , repulsive) or a high apolar component (γ^{LW} , attractive) [44]. The highest adhesion value was observed for PLLA (P < 0.05) which has the lowest repulsive forces (lower γ^{-1} , Table 1) when compared with the adhesion values observed for PS, CA, and PDMS. Regarding PDMS, it is possible to note that a similar γ^{-1} value was observed for this surface and PLLA. However, PDMS exhibited the lowest apolar attractive forces value (γ^{LW}) and this may have led to a lower adhesion than observed for CA and PS (with higher γ^{-1} , Table 1). Glass has the strongest repulsive force value (γ^{-1}) which can explain the lowest adhesion.

In the work of Liu and Zhao [27], the second order equation $y=a+bx+cx^2$ was used to correlate experimental data and the obtained correlation coefficients varied between 0.8123 and 0.9247 (Fig. 2b, c). In this work, the same equation was applied

to the adhesion results and a correlation factor of 0.9917 was obtained (Fig. 2a). Additionally, results from all these works from the literature survey (Table 2 and Fig. 1) were re-plotted in Fig. 2, where it is possible to see that the γ^{LW}/γ^{-} parameter has a strong correlation with bacterial adhesion results from the work of Katsikogianni et al. [50] (Fig. 2d), Hong et al. [49] (Fig. 2e), and Cunliffe et al. [51] (Fig. 2f) and with the values obtained for protein adsorption by the same author (Fig. 2g, h).

Liu and Zhao [27] were able to correlate cell adhesion to the γ^{LW}/γ^{-} ratio and their working range was between 1.21 and 6.74 (Fig. 1). Although these authors have tested metallic surfaces that can be used in heat exchangers and ship hulls, they have suggested that their results could also be applied to biomedical surfaces. With the results obtained in the present work, this hypothesis was confirmed since a good correlation between *E. coli* adhesion to biomedical polymers and the γ^{LW}/γ^{-} range. Additionally, and considering data obtained from other works, it was possible to observe the validity of this correlation under diversified conditions.

Therefore, the available data seem to indicate that the γ^{LW}/γ^{-} ratio can be a good parameter for rapid material selection that can be used either to promote (higher γ^{LW}/γ^{-} values) or to decrease bacterial adhesion (lower γ^{LW}/γ^{-} values). These results may also be helpful in the design of new materials by controlling the ratio γ^{LW}/γ^{-} according to the desired application.

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