Effect of Blending Sodium Polyethylene-5-Sulfoisophthalate on Adhesion of Clinical Bacteria on Polyethylene Terephthalate

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ABSTRACT: This work studies the adhesion of clinical infecting bacteria, *S. aureus* and *E. coli,* on prosthetic polymeric materials. Membranes were prepared from polyethylene terephthalate (PET) blending at various ratios with sodium polyethylene-5-sulfoisophthalate (SPES). The membranes were characterized by measuring the contact angle, equilibrium water content, and the surface concentration of sodium sulfonate. The results show that sulfonate makes the membrane more hydrophilic. The surface properties of bacteria were determined by measuring the adhesion to *n*-octane (*B*%) and the contact angles to water and α-bromonaphthalene. For the four bacteria studied, encapsulated *S. aureus* was the most hydrophobic and had the highest amount of bacteria attached to the surface of SPES/PET membrane. Furthermore, the attached amount decreased with the increase of the content of SPES. Empirical correlations for predicting the attached amount from the surface properties of both polymer and bacteria were obtained from linear regression. © 2004 Wiley Periodicals, Inc. J Appl Polym Sci 91: 3587–3594, 2004

Key words: bacterial; adhesion; blends; polyesters; interfaces

INTRODUCTION

Bacterial adherence on biomaterials is an important step of prosthetic infection. Although the exact mechanism of infection is still unclear, the slime secreted by bacteria does form a biofilm on the prosthetic materials. This biofilm can protect bacteria from the human defense system and reduce the antibiotics susceptibility.[1] To prevent the bacterial adherence onto biomaterials, the mechanism, the characteristics, and the process of adhesion ought to be studied. During the implant of biomaterials, bacterial infection often occurs and results in the failure of the implantation. This infection may range from mild or asymptomatic to recurrent and catastrophic. The combined rates of death and morbidity associated with the infection of cardiac, abdominal, and extremity vascular prostheses may be higher than 30%. Infection is a major complication in the long-term use of total artificial heart. Long-term use of catheters used for intravenous access, peritoneal dialysis, or urinary tract access frequently succumbs to infection.[2]

The interaction of bacteria and the biomaterial surface leads to the bacterial adherence. Hydrophobicity of the biomaterial can promote the adhesion of bacteria and result in infection. The adhesion is favored when the difference of surface-free energy between the biomaterial and the bacteria is small.[3] The closer the surface-free energy of bacteria to that of the biomaterial surface, the more possible causes of adhesion.[4–6]

When studying the adhesion of bacteria to poly(tetrafluorethylene-*co*-hexafluorpropylene) (FEP), Hogt et al. discovered that hydrophobic encapsulated *Staphylococcus epidermidis* is more prone to adhere to FEP, and less prone to adhere to the hydrophilic surface of plasma-exposed FEP.[7,8]

In the literature, recent studies reported that sulfonation can prolong the thrombin time as well as detach some bacteria. Most of the biomaterials using sulfonation to increase hydrophilicity are polystyrene (PS) and polyurethane (PU). Sulfonated PS showed less attachment of *Escherichia coli* than untreated PS in phosphate buffer solution.[9] The adhering number of living L1210 cells increased with the sulfonic group content on the surface of sulfonated styrene/methyl methacrylate copolymer.[10] For sulfonated PU, the thrombin time of human plasma can be prolonged and increase with the sufonate content of the polymer.[11] Detachment of *Staphylococcus aureus* was observed on sulfonated PU.[12]

Polyethylene terephthalate (PET) is a transparent material with good mechanical properties, chemical

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resistance, abrasion resistance, and flexibility. It is frequently used for making vascular, laryngeal, and esophageal prostheses.[13] However, PET is a hydrophobic material and can interact with bacteria to cause a large number of attachments. The attachment of marine pseudomonad can be reduced by cleaning the surface with radiofrequency plasma to increase the wettability of PET.[14]

In this work, the hydrophilicity of PET is improved by blending with sodium polyethylene-5-sulfoisophthalate (SPES). SPES is a copolymer of PET and 5-sulfoisophthate and has properties similar to PET.[15] To study the bacterial adherence, four strains of clinical infective bacteria, two Gram-negative *E. coli,* encapsulated Gram-positive *S. aureus,* and nonencapsulated Gram-positive *S. aureus,* are chosen. The results of this work can hopefully be useful to elucidate the adherence of biomaterials containing sulfonic groups.

EXPERIMENTAL

Polymeric biomaterials

Preparation of polymer films

Polyethylene terephthalate (M_w, 20,000) and SPES (M_w, 18,000) were obtained from Tuntext Co. (Taipei, Taiwan). These two polymers were blended in the ratios of 0, 20, 50, 80, and 100 wt % with a twin-screw extruder (Sino-Alloy Machinery, Inc., Taiwan) and injection-molded into films with dimensions of 10 × 10 × 3 mm.

Characterization

Determination of sulfonic group content. A piece of the film was immersed in 1*N* HCl and shaken for 1 h at 37°C, and then left to stand for 1 h. The solution was filtered with 0.45-μm cellulose triacetate filter (Type SC, Millipore Co., USA), and then the sodium concentration was determined by using a graphite furnace atomic absorption (GFAA) spectrometer (Analyst 300, Perkin–Elmer, USA). Calibration curve was established from standard solutions of 0.1 to 0.5 ppb, which were diluted from the standard stock (Merck & Co., USA). To determine the bulk content of sulfonic group, SPES was dissolved in a 1 : 1 mixture of phenol and 1,1,2,2-tetrachloroethane. The sodium concentration in the solution was then determined by using the GFAA.

Determination of equilibrium water content. Films were immersed in a water bath at 37°C for 5 h and weighed as W_0. Afterward, these films were placed in a vacuum oven at 40°C for 24 h and weighed as *W*. The equilibrium water content (EWC%) was then calculated according to the following formula:

$$\mathrm{EWC\%} = \frac{W - W_0}{W_0} \times 100\% \quad (1)$$

Surface free energy. Contact angles were measured by the sessile drop technique by using a contact angle tester (DSA 100, Krüss GmbH, Germany) with water and *n*-octane at 25°C. The solid surface-free energy between polymer and air (γ_{SV}) was automatically calculated from a series of contact angles according to the Wu method.[16] The interfacial free energy (γ_{SL}) between the polymer and water is calculated according to the harmonic mean equation.[17]

Bacteria

Preservation and culture

Table III lists those four strains of bacteria used in this work. After isolating a single cell by the streak method, the bacteria were cultured with brain–heart infusion broth (BHI, Difco Laboratories, USA) at 37°C for 1 month and then preserved in 10% glycerol at −80°C. For short-term preservation, the broth was kept at 4°C. The frozen sample was thawed at 25°C; then 0.1 ml was pipetted and streaked into quadrants on sheep blood agar plate (Difco Laboratories) and cultured overnight at 37°C. Afterward, a single colony was scraped with a loop and swabbed to a 15° slant medium (10 ml of nutrient agar) and incubated at 37°C. After culturing for 18–24 h, 20 ml of PBS was added. After mixing, 1 ml of the solution was moved into 9 ml of nutrient broth (concentration = 8 g/l) and mixed with a vortex mixer. Eight consecutive dilute solutions were prepared by taking 1 ml of the previous solution and mixed with 9 ml of PBS.

Staining of capsules

The presence of capsules was demonstrated in Indian ink wet-film preparation of washed bacterial cells according to Duguid.[18] Examining under ×400 microscope, the bright halo indicated the presence of capsules. To observe the slime, *E. coli* was cultured with Macconkey agar at 18°C for 14 days, while *S. aureus* was cultured with mannitol salt agar at 37°C for 10 days. After staining by the India ink wet-film method, and examining under ×400 microscope, slime showed a large light area, although darker than capsule but lighter than pure ink background.[18,19] Table IV shows the observed results.

Hydrophobicity test

The hydrophobicity of bacteria was measured by the microbial adhesion to hydrocarbon (MATH) technique.[4] In this method, 3 ml of bacteria suspension in PBS was vortexed for 1 min with 0–1 ml of *n*-octane in

TABLE I
Surface and Bulk Compositions of SPES/PET Blends

Sample	Blending ratio	Surface content of —SO_3Na (μmol/cm^2)	Bulk content of —SO_3Na (μmol/g-solid)	EWC (%)
PET	0%	0	0	0.39 ± 0.03
SPES02	20%	0.57 ± 0.01	19.0	0.52 ± 0.04
SPES05	50%	1.29 ± 0.02	47.4	0.69 ± 0.03
SPES08	80%	2.23 ± 0.01	75.8	0.88 ± 0.05
SPES	100%	3.16 ± 0.01	94.8	0.98 ± 0.07

a 5 ml cuvette. The mixture was then left at rest for 30 min to let the two phases separate completely. The absorbance of the lower aqueous phase was determined at 600 nm by using a spectrophotometer (UV2100, Shimadzu, Japan). The ratio of bacterial adhesion to *n*-octane (B%) was calculated as

$$B\% = (1 - A/A_0) \times 100\% \quad (2)$$

where A is the absorbance of the aqueous phase after mixing octane, and A_0 is the initial absorbance without mixing *n*-octane.

Surface-free energy of bacteria

The bacterial substrata for measuring contact angles were prepared by collecting bacterial cells to a density of 10^8 cells/mm^2 on a 0.45-μm filter membrane after washing three times with PBS. To maintain the moisture content, the filter with bacteria was placed on a nutrient agar plate until the filter was fixed onto a glass plate with a piece of 0.4-cm^2 double-sided adhesive tape.[20] The glass plate was then mounted on the contact angle tester using water and α-bromonaphthalene as wetting agents. The measurement of the height H and diameter D of the drop was performed within 3 s at 25°C. The surface-free energy of bacteria (γ_{BV}) was calculated according to the Wu method.[16] The interfacial free energy (γ_{BL}) between bacteria and liquid was calculated according to the harmonic mean equation.[17]

Bacterial adherence to polymer surfaces

Bacteria suspension was centrifuged at 500 rpm for 10 min and washed three times with 5 ml PBS. The suspension was then filtered with a 0.45-μm filter membrane and bacteria were resuspended with 20 ml of PBS to form a suspension of concentration of 2 $\times$ 10^9 cells/ml. In each test tube, a piece of the polymer film was immersed in 2 ml of the bacteria suspension and incubated at 37°C for 6 h (the log-growth phase). Afterward, the film was washed three times with 5 ml of PBS to remove unattached bacteria. The washed film was then vortexed in 20 ml of PBS at 1500 rpm for 5 min and sonicated for 30 s to detach adhered bacteria.[21] This sonicated suspension was then diluted 10-fold. Six consecutive 10-fold dilutions were performed. From each diluted suspension, 1 ml was withdrawn, mixed well with 14 ml of nutrient agar at 45°C, and poured into a 9-cm Petri dish. After cooling to room temperature, the dish was incubated at 37°C for 18–24 h. The number of bacteria cells was then counted.

TABLE II
Surface Characteristics of SPES/PET Membrane

Sample	$\theta_{water,P}$ (°)	$\theta_{octane,P}$ (°)	γ_{SV} (erg/cm^2)	γ_{SL} (erg/cm^2)
PET	64.6 ± 1.5	82.6 ± 2.7	40.7	9.7
SPES02	60.7 ± 2.2	76.3 ± 2.1	42.7	7.4
SPES05	55.6 ± 3.1	67.4 ± 3.2	45.6	4.8
SPES08	51.6 ± 1.7	59.1 ± 2.6	47.9	3.1
SPES	46.9 ± 1.8	51.1 ± 2.4	51.2	1.9

RESULTS AND DISCUSSION

Characteristics of polymer

The sodium content (i.e., the sulfonic content) on the surface of the film ranged between 0 and 3.16 μmol/cm^2, as listed in Table I. This is equivalent to 0–19.6

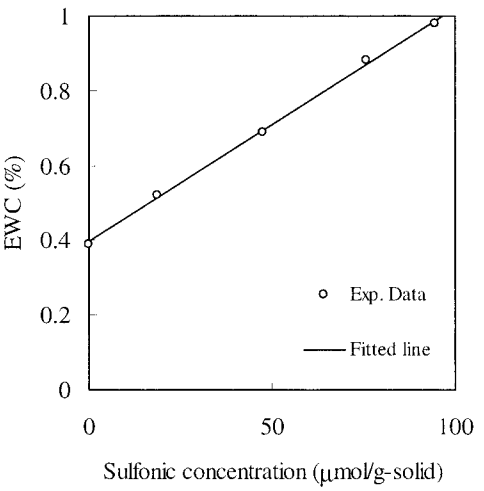

Figure 1 The linear dependence of EWC% of PET membrane on the bulk concentration of sufonic group. R^2 = 0.999.

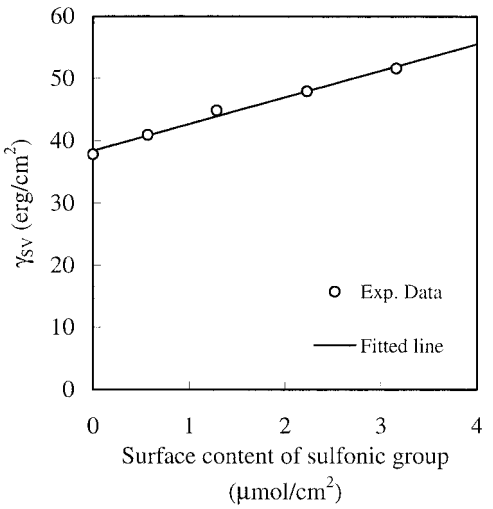

Figure 2 The linear dependence of surface-free energy of PET on the surface concentration of sulfonic group. $R^2 = 0.990$.

$\times 10^{18}$ molecules/cm^2. By taking linear regression on these numbers, the surface content of the sulfonic group was increased by 3×10^{-8} mol/cm^2 for every 1% of SPES blended. In addition, the bulk sodium content in SPES is 0.218 wt %; hence, the bulk concentration of —SO_3Na in SPES is 9.48×10^5 mol/g SPES. Therefore, the sulfonic group is very dilute in SPES. Because the molecular weight of each repeating unit for PET (and SPES) is 192 g/mol, there is one sulfonic group for every 55 repeating units of the SPES chain.

The EWC% of each polymer blend is given in Table II and ranges from 0.39 to 0.98 wt %. The EWC% is linearly dependent on the bulk content of the sulfonic group, as depicted in Fig. 1. This suggests the hydrophobicity of PET membranes increases with the blending ratio.

The surface-free energy is linearly correlated to the surface concentration of sulfonic group, as depicted in Fig. 2. By applying linear regression to the surface-free energy with respect to the surface content of sulfonic group, the slope was 4.29 erg/μmol SPES with a coefficient of determination R^2 of 0.990; thus the surface-free energy is linearly correlated to the sulfonation. The surface-free energy of PET was 40.7 erg/cm^2, which is very close to the values (41–44 erg/cm^2) in the literature.[14,16] By blending 50 wt % of SPES, 1.3 μmol/cm^2 of the sulfonic group would be introduced to the surface of PET membrane, and the surface-free energy would be increased by 15%. Similar results were reported for the sulfonation of PU and PS.[11–13]

Surface characteristics of bacteria

Those four clinical bacteria used in this work are listed in Table III, and their surface characteristics are listed in Table IV. Among these four bacteria, *S. aureus*-1 has capsule, while the rest do not. All these bacteria do not have slime. Both *E. coli*-1 and *E. coli*-2 do not have extracellular capsules. Colonies of *S. aureus* can often be found on prostheses such as artificial hip joints, vascular grafts, and artificial cardiac valves. The acute abscess infective bacterium used in this work, *S. aureus*-1, frequently infects implanted and nonimplanted devices and caused death. Thus, it is important to reduce the adhesion of clinical infective bacteria to biomaterials.

Bacterial adhesion to *n*-octane

The hydrophobicity of bacteria is evaluated by the ratio of bacterial adhesion to *n*-octane (*B*%) and the surface-free energy (γ_{BV}) of bacteria calculated from the contact angle measurement. Higher *B*% means the bacterium is more hydrophobic.

Among these four bacteria, both *S. aureus* strains have higher *B*% than both *E. coli* strains, as depicted in Figure 3 and Table IV. This indicates that *S. aureus* strains are more hydrophobic than *E. coli* strains. In addition, encapsulated *S. aureus*-1 has much higher *B*% than the other three bacteria. The reason that more *S. aureus*-1 cells adhere to *n*-octane is that the capsule can be adsorbed to the inert surface,[22,23] and that the encapsulated bacteria carry less negative charge.[24] This makes hydrophobic bacteria more prone to attach to the surface with little or no charge, such as Teflon, polyethylene (PE), poly propylene (PP), PS, and PET.

TABLE III
Clinical Source of Bacteria Used in This Study

Bacteria	Clinical source
Gram-positive bacteria	
Staphylococcus aureus strain-1 (*S. aureus*-1)	Acute abscess infection culture
Staphylococcus aureus strain-2 (*S. aureus*-2)	Wound infection culture due to suture
Gram-negative bacteria	
Escherichia coli strain-1 (*E. coli*-1)	Blood culture
Escherichia coli strain-2 (*E. coli*-2)	Urine tract culture

TABLE IV
Hydrophobic Characteristics of Bacteria

Bacteria	Capsule	B (%)	$\theta_{water,B}$ (°)	$\theta_{\alpha\text{-bp},B}$ (°)	γ_{BV} (erg/cm^2)	γ_{BL} (erg/cm^2)
S. aureus-1	+	53.6 ± 1.6	48.8 ± 4.6	59.5 ± 4.7	52.0	4.05
S. aureus-2	−	40.2 ± 1.2	41.6 ± 1.2	45.2 ± 1.5	59.3	4.78
E. coli-1	−	35.3 ± 0.7	39.9 ± 1.2	38.2 ± 1.5	61.3	5.35
E. coli-2	−	24.5 ± 0.4	33.0 ± 1.2	40.5 ± 2.0	69.8	3.05

By adding ionizing groups to the surface, the negativity would reduce the attachment of bacteria.[14]

Bacterial surface-free energy

The results of contact angle measurements are listed in Table IV. Because of the extracellular capsule, the surface-free energy of *S. aureus*-1 is lower than the other three bacteria. This value (52 erg/cm^2) is lower than the reported values of 66–69 erg/cm^2 in the literature.[25,26] On the other hand, *E. coli*-2 has the highest surface-free energy of 69.8 erg/cm^2, which is higher than the reported values of 43–69 erg/cm^2 in the literature.[26,27]

The linear dependence of B% and γ_{BV} on θ_{water} is shown in Figure 4. Linear regression of B% with respect to the θ_{water} gives a coefficient of determination R^2 of 0.994, and that for γ_{BV} is 0.996. This indicates the linear dependence of B% and γ_{BV} on θ_{water}. In the literature, bacteria used for studying the adhesion included *A. calcoaceticus, Lactobacilli, S. mitis,* and *S. salivarius*.[28–31] In these reports, linear dependence of B% on θ_{water} was not observed. The difference may be because their measurement of contact angle was performed on dry bacteria, whereas in this work, the measurement was performed on wet bacteria layer.

Bacterial adherence to polymeric surface

Interfacial free energy of adhesion

The interfacial free energy of adhesion is defined as[20]

$$\Delta F_{adh} = \gamma_{SB} - \gamma_{SL} - \gamma_{BL} \quad (3)$$

where γ_{SB} is the interfacial free energy between polymer and bacteria. These interfacial free energies were calculated according to the harmonic mean equation.[16,17] Table VI lists the results of calculation.

The occurrence of bacterial adherence to polymeric surface can be predicted from ΔF_{adh}.[26] Negative ΔF_{adh} suggests adhesion is favored, while positive ΔF_{adh} is unfavored. This point is illustrated in Figure 5, where the number of bacteria attached to the polymer surface (N_B) increases with the increase of $-\Delta F_{adh}$. In addition, Figure 5 also shows that N_B is linearly dependent on $-\Delta F_{adh}$. Linear regression on these data gives R^2 between 0.959 to 0.993.

According to the surface thermodynamics of adhesion, when γ_L (=72.1 erg/cm^2) < γ_{BV}, bacterial adherence to polymeric surface is increasingly possible to occur with increasing γ_{SV}; when $\gamma_L > \gamma_{BV}$, bacterial adherence to polymeric surface becomes less possible with increasing γ_{SV}. All four bacteria have γ_{BV} less

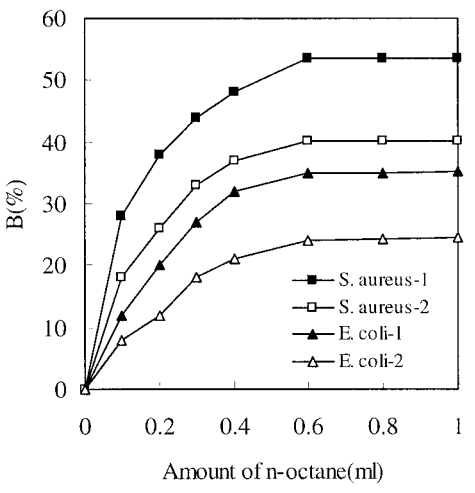

Figure 3 The influence of an added amount of *n*-octane added on the bacteria adhesion to hydrocarbon B%.

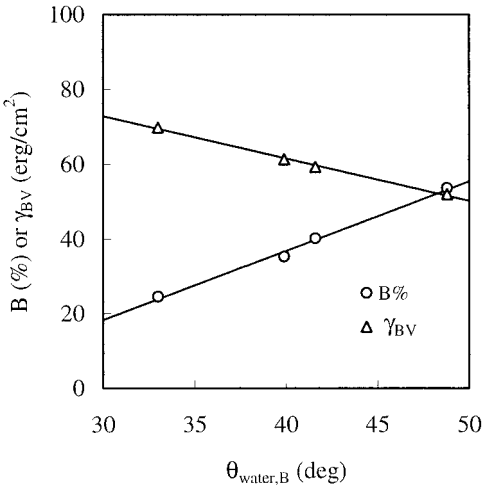

Figure 4 The linear correlations of B%, γ_{BV}, and $\theta_{water,B}$. For B% versus $\theta_{water,B}$, $R^2 = 0.994$; for γ_{BV} versus $\theta_{water,B}$, $R^2 = 0.996$.

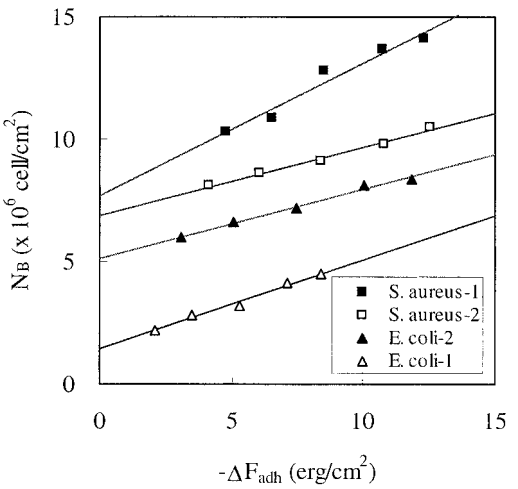

Figure 5 The linear dependence of the number of bacteria attached on the interfacial free energy of adhesion (ΔF_{adh}). $R^2 = 0.959–0.993$.

than 72.1 erg/cm^2. Therefore, ΔF_{adh} becomes less negative with the increase of γ_{SV}, as shown in Figure 6. This implies that the bacterial adherence becomes less favored as the polymer surface-free energy increases. The linear regression of γ_{SV} versus $-\Delta F_{adh}$ of each bacterium yields $R^2 > 0.996$; thus, they are linearly dependent, which is shown in Figure 6. Because N_B decreases linearly with the increase of $-\Delta F_{adh}$, N_B must also linearly decrease with the increase of γ_{SV}, as depicted in Figure 7. In other words, the hydrophobicity of bacteria can be represented by *B*% and the contact angle. The number of bacteria attached increased with the increase of *B*% of bacteria, as depicted in Figure 4. Among these four bacteria, *S. aureus*-1 has the highest *B*%, which indicates that it is the most hydrophobic. This explains why it has the highest number of attached bacteria. Furthermore, with the increase of sulfonic group, the attached amount decreased. The other measure of hydrophobicity of bacteria is the contact angle. Larger contact angle represents higher hydrophobicity. As shown in Figure 5, the number of bacteria attached increased with the increase of contact angle.

From the above results, the number of bacteria attached to the polymer surface depends linearly on γ_{SV},

TABLE V
Interfacial Free Energies (γ_{SB}) Calculated by Geometric Mean Approximation

Sample	*S. aureus*-1	*S. aureus*-2	*E. coli*-1	*E. coli*-2
PET	1.41	1.92	2.18	4.35
SPES02	0.71	1.36	1.69	3.32
SPES05	0.32	1.20	1.64	2.54
SPES08	0.60	1.82	2.36	2.64
SPES	1.15	2.52	3.14	2.85

TABLE VI
Free Energy of Adhesion of Bacteria (ΔF_{adh}) and Polymer Surfaces

Sample	*S. aureus*-1	*S. aureus*-2	*E. coli*-1	*E. coli*-2
PET	−12.34	−12.56	−11.84	−8.40
SPES02	−10.74	−10.82	−10.03	−7.13
SPES05	−8.53	−8.38	−7.48	−5.31
SPES08	−6.55	−6.06	−5.06	−3.51
SPES	−4.80	−4.16	−3.08	−2.10

B%, θ_{water}, and ΔF_{adh} in this work. However, an other study did not report such a linear dependence.[4] This is probably because in this work γ_{SV} is in a narrower range.

By blending SPES with PET, sulfonic groups were introduced to the polymer surface. In PBS, pH is 7, and thus sulfonic group carries negative charge. Bacteria in general also carry negative charge. Therefore, the adhesion was reduced by the electrostatic repulsion. This is evidenced by the data in Table VII. As the blending ratio of SPES increased, the attached amount of bacteria decreased. The difference between γ_{BV} of *S. aureus*-1 and γ_{SV} of PET was the smallest; thus more *S. aureus*-1 attached to PET than in other cases. This result is in agreement with other studies.[4–6]

Empirical correlation for predicting the adhesion amount

In Figures 5 and 7, the number of bacteria attached to the polymer surface is linearly dependent on γ_{SV} and ΔF_{adh}. This implies that for this system, there exists a linear correlation between the number of bacteria attached and the surface properties of the surface as well as the bacteria. From an engineering perspective, a

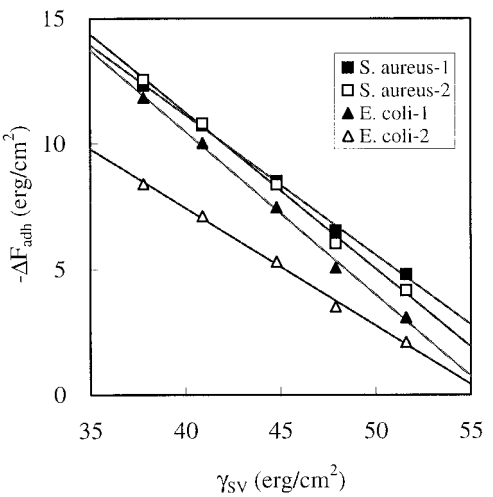

Figure 6 The linear dependence of interfacial free energy of adhesion of bacteria to polymer (ΔF_{adh}) on the surface-free energy of polymer (γ_{SV}). $R^2 = 0.996–0.998$.

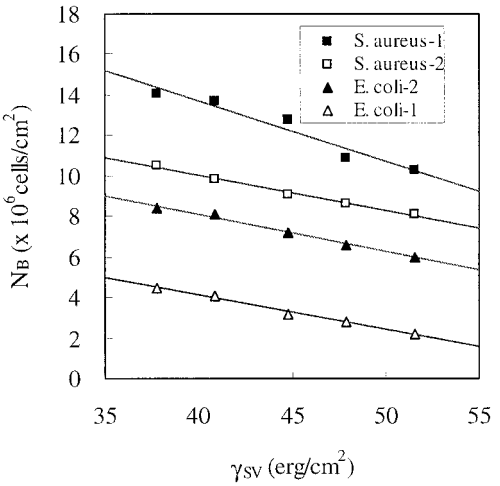

Figure 7 The linear dependence of the number of bacteria attached on the surface-free energy of polymer (γ_{SV}). R^2 = 0.945–0.982.

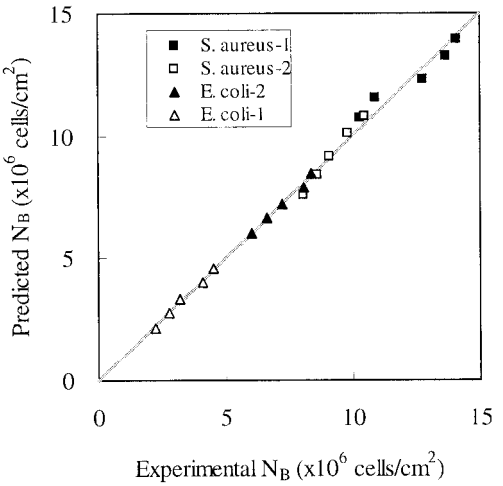

Figure 8 The goodness of prediction from empirical correlations with respect to experimental data.

correlation is useful for predicting the adhering behavior of bacteria. The simplest parameter to represent the surface property is the contact angle, which can be directly measured by using a goniometer without invoking any theoretical model. Thus, by taking linear regression on the experimental data, a correlation was obtained for *S. aureus*

$$N_B = 0.436\ \theta_{\text{water},B} + 0.184\ \theta_{\text{water},P} - 19.20 \qquad (4)$$

where $\theta_{\text{water},B}$ is the contact angle of bacteria against water, and $\theta_{\text{water},P}$ is the contact angle of polymer against the water. The R^2 for this correlation was 0.959. Similarly, for *E. coli,* the correlation was

$$N_B = 0.565\theta_{\text{water},B} + 0.137\theta_{\text{water},P} - 22.97 \qquad (5)$$

The R^2 of this correlation was 0.998. If two more parameters (θ_{octane} and $\theta_{\alpha\text{-bp}}$) were added to perform the linear regression, the R^2 was not significantly changed. Therefore, a two-parameter correlation could sufficiently describe the adhering behavior of those four bacteria. The values of N_B predicted from these correlations were very close to the experimental data, as shown in Figure 8. By comparing the regression factors in these two formulas, we can see that the surface property of bacteria is more influential to the adhering behavior of bacteria than the surface property of polymer.

CONCLUSION

The surface-free energy of PET membrane increases linearly with the surface content of are sulfonic group, which is introduced by blending PET with SPES. When the surface content of the sulfonic group increased from 0 to 3.16 μmol/cm^2, the surface energy γ_{SV} increased from 37.8 to 51.6 erg/cm^2. The adhesion of bacteria on polymer surface depends on their interfacial free energy, which is thus affected by the blending of SPES. Because of the hydrophobic nature of SPES/PET, more hydrophobic *S. aureus* adheres more than less hydrophobic *E. coli.* When γ_{SV} increased from 37.8 to 51.6 erg/cm^2, N_B of the most hydrophobic *S. aureus*-1 decreased by 3.8 $\times$ 10^6 cells/cm^2, whereas that of the least hydrophobic *E. coli*-2 decreased by 2.3 $\times$ 10^6 cells/cm^2.

For this particular system, linear dependence of N_B on ΔF_{adh} and γ_{SV} is observed for *S. aureus* and *E. coli.* Because both ΔF_{adh} and γ_{SV} were derived from contact angles, two linear correlations were obtained by applying linear regression on N_B against $\theta_{\text{water},B}$ and

TABLE VII
Number of Bacteria Attached (×10^6 cells/cm^2) on Polymer Surface After 6 h of Incubation

Sample	*S. aureus*-1	*S. aureus*-2	*E. coli*-1	*E. coli*-2
PET	14.1 ± 0.1	10.5 ± 0.4	8.4 ± 0.4	4.5 ± 0.1
SPES02	13.7 ± 0.2	9.8 ± 0.3	8.10 ± 0.05	4.09 ± 0.04
SPES05	12.8 ± 0.1	9.1 ± 0.2	7.2 ± 0.1	3.23 ± 0.07
SPES08	10.9 ± 0.1	8.60 ± 0.05	6.6 ± 0.2	2.80 ± 0.04
SPES	10.30 ± 0.05	8.11 ± 0.03	6.00 ± 0.04	2.21 ± 0.05

$\theta_{water,P}$ for both types of bacteria (*S. aureus* and *E. coli*). Predicted values show a good match with the experimental data. Regression factor shows that N_B depends more on $\theta_{water,B}$ than $\theta_{water,P}$.

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