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TE WĀHANGA PŪTAIAO

The effect of silver ion-implantation of stainless steel on bacterial adhesion and biofilm formation

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The effect of silver ion-implantation of stainless steel on bacterial adhesion and biofilm formation

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ABSTRACT

Biofilms comprise an assembly of microbial communities attached to a surface and enclosed in a polysaccharide matrix. Biofilms are a problem in dairy manufacturing plants where they cause biofouling of the stainless steel surface, resulting in product contamination and the need to shorten manufacturing runs for frequent cleaning. The equipment has to be thoroughly cleaned at regular intervals to remove biofilms. Clean-In-Place (CIP) procedures used in dairy manufacturing plants are not effective enough to remove all the biofilm cells as the extracellular polysaccharide (EPS), an integral part of a biofilm, reduces the penetration of cleaning agents. One possible alternative method to control biofilm growth involves surface modification of the stainless steel by implanting silver ions to prevent the attachment of viable bacteria that would otherwise form biofilms.

Stainless steel coupons, implanted with 1×10^{16} silver ions per cm^2, and control stainless steel coupons were tested for the attachment of *Streptococcus thermophilus* and *Pseudomonas fluorescens* in various media for up to 30 minutes. Biofilm formation and EPS production for up to 24 hours was studied on the silver-implanted and control coupons in whole milk, skimmed milk and whey. It was found that there was higher attachment (0.49 and 0.18 log CFU per cm^2 of *S. thermophilus* and *P. fluorescens*, respectively) on the stainless steel coupons than on the silver-implanted coupons in saline. In the presence of milk and whey, the difference in the attachment of bacteria on the two coupons reduced. Biofilm studies showed that the number of bacteria colonising both types of coupons was not statistically significantly different ($P > 0.05$). While the Live/Dead® BacLight™ Bacterial Viability stain showed that there were a few dead cells on the silver-implanted surfaces, scanning electron micrographs showed that the bacteria attached to a conditioning layer formed by the milk and whey proteins. Furthermore, both bacteria produced EPS, which, along with the conditioning film, might have masked the effect of silver ions from bacteria, resulting in similar numbers of bacteria present on the test and control coupons. Thus, due to the shielding effect of EPS and the conditioning film, the silver-implanted surfaces may be of limited practical value in the dairy industry.

LIST OF PRESENTATIONS

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LIST OF ABBREVIATIONS

%	percentage
℃	degree celsius
µl	microlitre
µg	microgram
Ag	silver
Al^{2+}	aluminium ion
Co^{2+}	cobalt ion
cm	centimetre
Cu^{2+}	copper ion
$C_6H_5K_3O_7.2H_2O$	tri-potassium citrate dihydrate
$CaCl_2.2H_2O$	calcium chloride dihydrate
CDC	centre for disease control
CIP	clean-in-place
CFU	colony forming units
DLC	diamond-like carbon
DNA	deoxyribonucleic acid
EPS	extracellular polysaccharide
g	acceleration due to gravity
g	gram
g/l	gram per litre
H_2SO_4	sulphuric acid
keV	kiloelectronvolt
KH_2PO_4	potassium dihydrogen phosphate
KOH	potassium hydroxide
L	litre
LPS	lipopolysaccharides

m	metre
M	molar
mbar	millibar
mm	millimetre
Mg	milligram
ml	millilitre
mM	millimolar
$MgCl_2.6H_2O$	magnesium chloride hexahydrate
MoS_2^{2+}	molybdenum disulphide ion
N^+	nitrogen ion
Nm	nanometre
$Na_3C_3H_5O(CO_2)_3.2H_2O$	sodium citrate dihydrate
Ni	nickel
O^+	oxygen ion
O.D.	optical density
Pb^{2+}	lead ion
PTFE	polytetrafluoroethylene
R_a	average roughness
Rpm	revolutions per minute
SAS	statistical analysis system
SiF_3^+	silicon trifluoride ion
SEM	scanning electron microscopy
SS	stainless steel
TiC	titanium carbide
TSB	tryptic soy broth
UHT	ultra-high temperature
USFDA	United States food and drug administration

UV	ultraviolet
v/v	volume by volume
Zn^{2+}	zinc ion

LIST OF FIGURES