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SURFACE ACTIVE COMPOUNDS (SAC's) OF *Acinetobacter spp.:* APPLICATIONS IN BIOMEDICINE, A REVIEW

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ABSTRACT

Surface active compounds such as biosurfactants and bioemulsifiers have emerged as potential biomolecules because of their unique structure and diverse properties that are potentially useful for many therapeutic applications. Biosurfactants and bioemulsifiersof microbial origin have exhibited various biomedical activities such as antimicrobial, anti-inflammatory, anti-adhesive, antibiofilm and anticancer. Genus *Acinetobacter* have been reported decades back for production of surface-active compounds, however, there are incredibly few reports on application of surface-active compounds produced by genus *Acinetobacter* in the biomedical field. The increasing incidences of infections caused by multidrug resistant pathogens, nosocomial infections due to biofilms produced by pathogens and various types of cancer developing in the human population are posing serious health hazards to mankind. Therefore, exploring the potential of biosurfactants and bioemulsifiers to address therapeutic issues is the need of the hour. This article reviews the different types of biosurfactants and bioemulsifiersproduced by genus *Acinetobacter,* and the biomedical applications of these compounds.

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INTRODUCTION

A surfactant is an amphiphilic agent with both hydrophilic and hydrophobic structural moieties in its molecule and tends to be distributed at the interface between liquid phases with different degrees of polarity (oil/water). Surfactants reduce both surface and interfacial tension, leading to the capacity for detergency, emulsification, foaming, lubrication, solubilisation and phase dispersion. These traits make surfactants one of the most versatile process chemicals. Surfactants are commercially important due to various industrial applications of these compounds. These compounds are used in detergents, textile, leather, paper, chemical processes, pharmaceuticals, cosmetics, agriculture and food industries (Sobrinho *et al*. 2014; Harshada 2014; Gharaei-Fathabad, 2011). Most of the surfactants, that are commercially available are synthesised from petroleum derivatives. However, concerns among users and environmental legislation have led to the search for natural surfactants as a green alternative to the chemical surfactants. Several natural compounds with tensioactive properties are synthesised by living organisms. Compounds of a microbial origin that exhibit surfactant properties (emulsification capacity and a reduction in surface tension) are called biosurfactant (Sandeep and Rajasree 2017; Sobrinho *et al*. 2014; Gharaei-Fathabad 2011).

Biosurfactants (BSs) and bioemulsifiers (BEs) are thus, amphiphilic molecules mainly produced by microorganisms including bacteria, yeast and fungi. They possess both hydrophilic and hydrophobic moieties (Ohadi *et al*. 2017). They possess the characteristic property of reducing the surface and interfacial tensions using the same mechanisms as chemical surfactants. However, BS/BE show better environmental sustainability, improved foaming properties and stable activity at extremes of temperature,pH and salinity. These characteristics make BS/BE superior tothe chemical surfactants (Sandeep and Rajasree 2017; Gharaei-Fathabad2011; Satpute *et al*. 2010; Singh and Cameotra 2004). Surfactants synthesized by microbes have recently received increased attention in scientific world, due to their unique characteristics relative to their chemical counterparts. The unique features include non-toxicity, biodegradability, biocompatibility, effective at low concentrations and are synthesized from natural substrates under moderate environmental conditions (Banat *et al*. 2000, Singh and Cameotra 2004; Gharaei-Fathabad 2011; Uzoigwe *et al*. 2015). Moreover, they can be produced by microbial fermentation using several cheaper agro-based substrates and waste materials, thereby reducing the production cost (Sawant *et al*. 2021; Sandeep and Rajasree 2017; Banat *et al*. 2014; Singh and Cameotra 2004). The potential use of BSs/BEs in medical field have rapidly increased during the past few years. The antimicrobial, antifungal and antiviral activity exhibited by

the BSs/BEsmake them significant molecules for application as therapeutic agents in combating many diseases (Shah *et al*. 2016; Fracchia *et al*. 2012; Okoliegbe 2012; Kiran *et al*. 2010; Rodrigues *et al*. 2006). These molecules also exhibit anti-adhesive, anticancer, anti-inflammatoryand immunomodulatoryproperties, thus have widespread applications in medical field. These surface-activemolecules, therefore, exhibit potential candidature for new age chemotherapy (Sandeep and Rajasree 2017; Shekhar *et al*. 2015; Okoliegbe 2012; Fracchia *et al*. 2012; Rodrigues and Teixeira 2010).

***Acinetobacter*:** *Acinetobacter* are Gram negative, non-spore forming, non-fermenting often coccobacillary bacteria that belong in the family Moraxellaceae. The genome comprises a single circular chromosome sized 2.6−4.7 Mb and a strain-dependent set of plasmids. Flagella are absent; therefore, cells do not exhibit swimming motility. However, cells exhibit twitching motility because of presence of fimbriae. Cells commonly occur in pairs. Metabolism is strictly aerobic with oxygen as the terminal electron acceptor. All strains are mesophilic, grow between 20-30^{o}C, with an optimal temperature of 33-35°C for most strains. *Acinetobacter* strains are oxidase-negative, catalase-positive and most strains do not show positive nitrate reduction test. All strains show good growth on complex media. Colonies are generally nonpigmented and show mucoid appearance when the cells are encapsulated (Bergey 1930; Nemec 2022; Shete *et al*. 2015). *Acinetobacter* strains are widespread in nature, hence, inhabit variedwater and soil ecosystems and inhabit plant and animal bodies. Several species are also responsible for causing the nosocomial infections. Such strains are generally resistant to multiple antibiotics. *Acinetobacter* are a key source of infection in immunocompromised patients in the hospital, particularly*Acinetobacter baumannii*. According to the recent reports**,** the genus included 73 species (Nikolova and Gutierrez 2023).

Biosurfactants and bioemulsifiers: Surface active compounds (SACs) such as BSs/BEs are structurally diverse compounds mainly yielded by microorganisms utilizing hydrocarbons. BSs/BEsare surface active biomolecules, however, there are significant differences between them especially, based on their physico-chemical properties and physiological roles. BSs lower surface and interfacial tension at gas-liquid-solid interfaces where as, BEs lower the interfacial tension between immiscible liquids, or at the solid-liquid interface, resulting in the formation of more stable emulsions. BSs usually exhibit emulsifying capacity but BEs do not necessarily reduce surface tension (Varjani and Upasani 2017; Sandeep and Rajasree 2017; Fracchia *et al*. 2012). Thus, SACs are grouped into surfactants and emulsifiers. BSs reduce the surface tension, BEs form and stabilize of emulsions (Satpute *et al*. 2010). Biosurfactants are commonly low molecular weight produced by microorganisms and are composed of sugars, amino acids (hydrophilic moieties), saturated and unsaturated fatty acids (hydrophobic moieties) and functional moieties such as carboxylic acids e.g. glycolipids and lipopeptides (Sivapathasekaran and Sen 2017; Uzoigwe *et al*. 2015). These molecules beenamphiphiles can dissolve in both polar and non-polar solvents. BSs are well known for good surface activity which involves reducing the surface and interfacial tension between different phases such as liquid-air, liquid-liquid, and liquid-solid, exhibiting a low critical micelle concentration (CMC) and formation of stable emulsions. They can act as wetting, foaming and solubilizing agents in different industrial processes (Uzoigwe *et al*. 2015; Rahman and Gakpe 2008). Bioemulsifiers are high molecular weight biopolymers or exopolysaccharides. These are complex mixtures of heteropolysaccharides, lipopolysaccharides, lipoproteins and proteins (Uzoigwe *et al*. 2015). Like BSs, these molecules can proficiently emulsify two immiscible liquids such as hydrocarbons or other hydrophobic substrates even at low concentrations.However, these molecules are less efficient at surface tension reduction (Sandeep and Rajasree 2017; Shah *et al*. 2016). BEs are thus, useful in solubilization of poorly soluble substrates, increasing their access and availability for biodegradation. BEs can stabilize emulsions, thus increasing their use in various industries such as cosmetics, food, pharmaceutical and petroleum (Uzoigwe *et al*. 2015).

Table 1. Classification of microbial surfactants based on molecular weight and chemical nature with examples

Molecular weight	Type of the biosurfactant	Description	Examples	References
Low mol weight	Glycolipids	Carbohydrates in combination with aliphatic acids or hydrooxyaliphatic acids	Rhamnolipids (mono or di), Trehalolipids, Sophorolipids, Mannosylerythritol lipids (MELs), Trehalose tetraester, Trehalose dicorynomycolate, Cellobiolipids, Alpha-galactosylceramide, sulfoquinovosyl diacylglycerol, Polylol lipid, MyrmeKioside, Trikentoside	(Sandeep and Rajasree 2017;Kuyukina et al. 2001;Christofi and Ivshina 2002; Desai and Banat 1997; Inès and Dhouha 2015; Shah et al. 2016;Rahman and Gakpe 2008;Tanaka et al. 1990)
	Lipopeptides and lipoproteins	Cyclic lipopeptides	Surfactin, Lichenysin, Iturin family, Fengycins family, Serrawettins, Non-inoniccyclodepsipeptides, Viscosin, Subtilisin, Arthrofactin, Gramicidins, Polymyxin, Peptide-lipid, Hallobacilin, Mixirin, Somocystinamide A, Fellutamide, Pseudofactin, Rakicidin, Apratoxin	(Fracchia et al. 2012; Gharaei-Fathabad2011; Desai and Banat 1997;Dey et al. 2015; Shah et al. 2016; Rahman and Gakpe 2008; Sobrinho et al. 2014)
High molecular weight	Polymeric (lipoproteins, proteins, polysaccharides, lipopolysaccharides)		Emulsan, Liposan, Mannoprotein, Alasan, Biodispersan, Carbohydrate-protein-lipid, Aminolipids, Polysaccharide, Lipoglycan,	(Fracchia et al. 2012; Gharaei-Fathabad 2011; Desai and Banat 1997; Hyder 2015;Shekhar et al. 2015; Shah et al. 2016; Rahman and Gakpe 2008; Sobrinho et al. 2014)
	Particulate biosurfactant		Vesicles and fimbriae, Whole cells	(Desai and Banat 1997; Shah et al. 2016; Rahman and Gakpe 2008; Sobrinho et al. 2014)
	Fatty acids, neutral lipids and phospholipids		Fatty acids, Neutral lipids and phospholipids	(Desai and Banat 1997; Shah et al. 2016;Sandeep and Rajasree 2017;Rahman and Gakpe 2008; Sobrinho et al. 2014)

Classification of microbial surfactants

Microbial surfactants are classified mainly on the chemical composition (Santos *et al*. 2016; Shoeb *et al*. 2013) Table1 summarises different types of surface-active compounds produced by microbial world.

Table 2. Microbial surfactants produced by different strains of *Acinetobacter* spp. and their chemical nature

Sr. No	*Acinetobacter* spp. (Producer organism)	Isolation site	Type of the surfactant	References
1	*Acinetobacter radioresistens*	-	BE- Alasan	(Navon-Venezia *et al*. 1995)
2	*Acinetobacter calcoaceticus*	-	BE- Emulsan	(Desai and Banat 1997)
3	*Acinetobacter calcoaceticus*	-	BE- Biodispersan	(Desai and Banat 1997)
4	*Acinetobacter calcoaceticus*	-	BE- Polysaccharide/ lipopolysaccharide complexed with protein or pplypeptide	(Shabtai and Gutnick 1985)
5	*Acinetobacter baumannii* A25	-	BE- Protein- Polysaccharide complex	(Mujumdar and Chopade 2002)
6	*Acinetobacter junii*A6	-	BE- Not specified	(Mujumdar and Chopade 2002)
7	*Acinetobacter calcoaceticus*subsp. a*nitratus*SM7	oil-spilled seawater	BE- Not specified	(Phetrong *et al*. 2008)
8	*Acinetobacter* genospecies A15	Wheat rhizosphere	BE- Polysaccharide-protein-lipid complex	(Bhawsar *et al*. 2011)
9	*Acinetobacter baumannii* SS4	Marine	BE- complex of polysaccharide-protein-lipid	(Bhuyan 2011)
10	*Acinetobacter calcoaceticus* C42	Rhizosphere of corn	BE- Lipopeptide	(Bashetti*et al*. 2012)
11	*Acinetobacter baumanii*AC5	-	BE- Lipoglycan	(Hyder 2015)
12	*Acinetobacter beijerinckii* ZRS	Oil contaminated soil samples	BE- Polymeric	(Zhao *et al*. 2016)
13	*Acinetobacter* sp	Healthy human skin	BE- Not specified	(Jagtap *et al*. 2010)
14	*Acinetobacter calcoaceticus*	-	BS- Polysaccharide	(Tanaka *et al*. 1990)
15	*Acinetobacter calcoaceticus*	-	Particulate BS- Vesicles and fimbriae	(Desai and Banat 1997)
16	*Acinetobacter junii*	Soil	BS- Not specified	(Menezes Bento *et al*. 2005)
17	*Acinetobacter calcoaceticus*BU03	petroleum-contaminated soil	BS- Not specified	(Zhao and Wong 2009)
18	*Acinetobacter calcoaceticus*IMV B_7241	Petroleum contaminated soils	BS- Trehalose Mycolates (Glycolipid)	(Pirog *et al*. 2012)
19	*Acinetobacter* sp. D3-2	Petroleum contaminated soil	BS- Lipopeptide	(Bao *et al*. 2014)
20	*Acinetobacter* sp	Hydrocarbon contaminated soil	BS- Not specified	(Yuan *et al*. 2014)
21	*Acinetobacter baylyi*ZJ2	Oil-contaminated soil	BS- Lipopeptide	(Zou *et al*. 2014)
22	*Acinetbacter indicus* M6	Marine water	BS- Glycolipoprotein.	(Peele *et al*. 2016)
23	*Acinetobacter junii*	Petroleum reservior	BS- Rhamnolipid	(Dong *et al*. 2016)
24	*Acinetbacterjunii*B6	Oil excavation site	BS- lipopeptide	(Ohadi *et al*. 2017)
25	*Acinetobacter baumannii* MKS2	Oil polluted soil	BS- Glycolipids	(Muthukamalam *et al*. 2017)
26	*Acinetobacter bouvetii*UAM25	Culture collection from The National Polytechnic Institute (Mexico)	BE-lipo-heteropolysaccharide	(Ortega-de la Rosa *et al*. 2018)
27	*Acinetobacter* sp. Ab9-ES and*Acinetobacter* sp. Ab33-ES	Lipid-rich wastewater	BE- Glycoprotein	(Adetunji and Olaniran 2019)
28	*Acinetobacter venetianus*AMO1502	oil spilled off	BS- Not specified	(D'Almeida *et al*. 2024)
29	*Acinetobacter sp.* V2		BS- Complex of protein and fatty acid	(Ntshingila *et al*. 2022)
30	*Acinetobacter. junii*B6		BS- Lipopeptide	(Mehrabani *et al*. 2021)
31	*Acinetobacter calcoaceticus* P1-1A	Oil contaminated sample from offshore oil and gas platform	BS- Not specified	(Moshtagh *et al*. 2021)
32	*Acinetobacter sp.* AKBS16	Petrol pump site	BS- Emulsan	(Jadeja *et al*. 2018)
33	*Acinetobacter baumannii* MN3	Production water from crude oil reservoir	BS- Lipopeptide	(Parthipan *et al*. 2017)
34	*Acinetobacter baumannii* BJ5	Petroleum oil contaminated soil	BS- Glycolipid	(Gupta *et al*. 2020)
35	*Acinetobacter calcoaceticus*	palm oil mill facility soil	BS- Lipopeptide	(Chooklin *et al*. 2023)
36	*Acinetobacter sp. strain* JR7	Oil spilled soil	BS- Lipopeptide	(Zobaer *et al*. 2023)
37	*Acinetobacter baumanii strain* MS14413	agro-industrial wastes	BS- Not specified	(Onajobi *et al*. 2023)
38	*Acinetobacter junii*	petroleum hydrocarbon-contaminated soil	BS- Glycolipid	(Sahu and Shrivastava 2022)
39	*Acinetobacter radioresistens*(Strain S1-2)	sediment and seawater samples from the Caspian Sea	BS- Not specified	(Hassanshahian and Ravan 2018)
40	*Acinetobacter calcoaceticus*(Strain K4-2)	sediment and seawater samples from the Caspian Sea	BS- Not specified	(Hassanshahian and Ravan2018)
41	*Acinetobacter sp.* P 2(1)	Microbiology Laboratory of Biology Department, Faculty of Science and Technology, Airlangga University.	BS- Not specified	(Triawan *et al*. 2017)
42	*Acinetobacter johnsonii*ABR6	Petroleum reservoir	BS- lipopeptide	Akbari *et al*. 2020

Biosurfactants and Bioemulsifiers produced by *Acinetobacter* spp.: Several *Acinetobacter* spp. have been reported to be the potent producers of BSs and BEs till date. Table 2 summaries BS/BE produced by different species from *Acinetobacter* genus and their chemical nature.

Genetics for production of SACs by *Acinetobacter* spp: SAC's production involves operons which code for the enzymes necessary to synthesize the SAC's. Environmental conditions play a pivotal role in triggering the expression and further regulation of the genes that code to synthesize SAC's (Chabhadiya *et al*. 2024).

Studies demonstrated that genes associated with the synthesis of BE emulsan produced *Acinetobacter lwoffii* RAG-1 are clustered in the region termed as the *wee* cluster. This study reported that two genes *wzb* and *wzc*from the *wee* cluster are involved in the synthesis of emulsan (Nakar and Gutnik 2003). A study revealed that extracellular anionic lipoheteropolysaccharide emulsan, produced by *A. venetianus* RAG-1 was encoded by 27kbp *wee* gene cluster. Emulsan produced by *A. venetianus* RAG-1assisted in alkane degradation by capturing and transporting the hydrocarbon to the cell (Fondi *et al*. 2016). The bioemulsifier of *Acinetobacterradioresistens* KA53, referred to as alasan, encoded by gene *alnA* is a high-molecular-weight complex of an anionic polysaccharide containing covalently bound alanine (apoalasan) and protein (Toren *et al*. 2002).

Biomedical applications of biosurfactants and bioemulsifiers produced by *Acinetobacter* spp.: Surface active molecules such as BSs exhibit unique properties such as higher biodegradability and lower toxicity. Rigorous research on BSs/BEs have revealed interesting biological and chemical properties that divulge promising applications in various fields related to pharmaceutical and other sectors (Imtiaz *et al*. 2022; Ceresa *et al*. 2021; Sonawane *et al*. 2021; De Giani *et al*. 2021; Pendse and Aruna 2020; Naughton *et al*. 2019; Jemil *et al*. 2017; Prasad *et al*. 2015; Tomar and Singh 2014; Chakraborty *et al*. 2014; Gudiña *et al*. 2013).

Antimicrobial activity: In recent years, researchers have discoveredthatBSs/BEs exhibit various properties of biomedical importancesuch as, antibacterial, antifungal and antiviral activities. These properties make BSs/BEs promising candidates for treatment of many diseases (Sharma and Saharan 2016). In recent decades, there has been a global urge to find alternatives for currently used antibiotics and BSs/BEs have exhibited promising candidature as antimicrobial agents through research in this field (De Giani *et al*. 2021; Pendse and Aruna 2020; Naughton *et al*. 2019; Ndlovu *et al*. 2017; Prasad *et al*. 2015; Sharma *et al*. 2015; Gudiña *et al*. 2015; Chakraborty *et al*. 2014). A lipopeptide BS produced by *Acinetobacter junii* effectively exhibited antimicrobial activity against *C. albicans* and *C. utilis* and many bacterial pathogens e.g., *Pseudomonas aeruginosa, Staphylococcus aureus*, *Klebsiella pneumoniae, Escherichia coli* and *Salmonella typhi* at concentration of 5 ug/ml. The MIC values of the BS were lower as compared to the standard antifungal agent fluconazole, also according to the researchers BS form *A. junii* exhibited 100% inhibition against *C. utilis* (Ohadi *et al*. 2020). A novel BS produced by *Acinetobacter indicus* M6 reduced the surface tension of water from 72.0 to 39.8 mN/m and exhibited thermophilic, halophytic and acidophilicstability as well. The BS was purified by acetone precipitation and was recovered by column chromatography. The composition of the BS was studied by H^1NMR and LC-MS and was characterised as glycolipoprotein. Antimicrobial activity of the BS was determined by agar well diffusion assay for 50% and 100% growth inhibition for different concentrations of the BS ranging from 20-50 mg ml^{-1}. The effect of the BS on the cell membranes of the bacteria was elucidated by TEM. The BS showed antimicrobial activity against a broad range of pathogenic and non-pathogenic strains, including Gram-positive, Gram-negative bacteria and yeasts. Except for *S. aureus,* nearly complete inhibition was observed by the researchers, with different BS concentrations ranging from 20-50 mg ml^{-1}. *E. coli* showed the highest degree of inhibition at the lowest concentration of the BS. BS induced structural changes in the bacterial cell membrane as elucidated by the TEM images, whereas the cell membranes of the bacterial cells not exposed to the BS remained intact (Karlapudi *et al*. 2020). BE producing *Acinetobacter baumanii*AC5 was isolated from the sediments and the antimicrobial activity of the BEwas evaluated. BE production was growth dependent and was induced by presence of edible oil in the culture medium. Partially purified BEwas obtained by the solvent precipitation method using chloroform: methanol (2:1v/v) solvent system. Chemical composition analysis of the partially purified BE revealed that it is lipoglycan containing lipids 63%, carbohydrates 35% and a minor fraction of proteins 2%. Antibacterial activity of three different concentrations (10, 20 and 30 mg ml^{-1}) of the BEwas determined by agar disc diffusion method against *E. coli,S. aureus, Salmonella sp., and P. aeruginosa*. Lipoglycan BE exhibited antibacterial activity against the tested organisms with maximum antibacterial activity against *S. aureus* followed by *P. aeruginosa, Salmonella sp. and E. coli*. Antifungal activity of the BEwas determined on the radial growth rate of the *P. notatum* and *F. oxisporium*using potato dextrose agar (PDA) medium containing different concentrations of BE (0,10, 20 and 30 mg ml^{-1}). BE inhibited the radial growth rate of *P. notatum* and *F. oxisporium*and exhibiteddose dependent antifungal activity (Hyder 2015). Bhawsar *et al*. (2011) reported production of a polysaccharide-protein-lipid complexBE, which exhibited antimicrobial activity against *S. aureus, S. typhimurium, K. pneumoniae, A. niger, A. fumigatus, C. humicola, C. albicans*. Mostafapour *et al*.(2014) isolated BS producing *Acinetobacter*sp from oil contaminated sites. The BS produced was characterised as glycolipid and showed potent antimicrobial activity against pathogenic bacteria such as *Staphylococcus aureus*, *Staphylococcus epidermidis* and *Pseudomonas aeruginosa.*

Antibiofilm activity: "Biofilm is the unique pattern of growth exhibited by certain microbes that provides characteristic features and advantages to the microbes (Mishra *et al*. 2020). Biofilms are aggregation of microorganisms growing on biotic or abiotic surfaces. Biofilm formation is a crucial mechanismin some of the pathogenic microorganisms, which contributes to the survival of these pathogenic microorganisms in the environment (Doghri*et al*. 2020; Kiran *et al*. 2010). Biofilm-forming microbes exhibit reduced susceptibility to many antibiotics. Biofilm formation on devices used in medical facilities plays an important role causing nosocomial diseases (Kiran *et al*. 2010). Thus, biofilms produced by pathogenic microbes are alarming human health concerns because enhanced pathogenesis in causing infectious diseases. Biofilms help in survival of the microbes in a wide range of ecosystems (Doghri *et al*. 2020). Therefore, there is a necessity for promising antibiofilm agents, which can effectively control the biofilm formation or can contribute to disruption of the preformed biofilms on biotic and abiotic surfaces. BSs and BEs produced by many microorganisms can thus, be effective antibiofilm agents (Ohadi *et al*. 2020; Mishra *et al*. 2020; E Silva *et al*. 2017; Jemil *et al*. 2017; Sharma and Saharan 2016; Gudiña *et al*. 2015; Kiran *et al*. 2010). Figure1 represents different antibiofilm mechanisms exhibited by BSs/BEs.

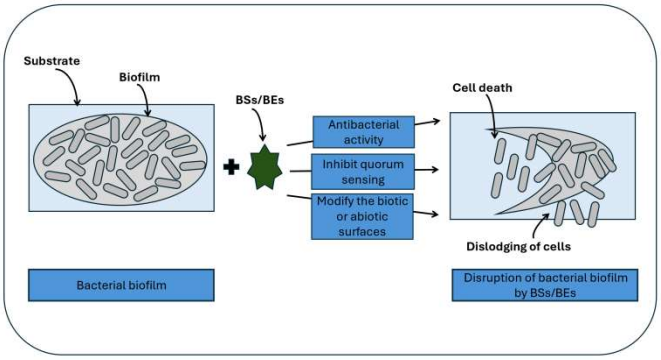

Figure 1. Schematic representation of different antibiofilm mechanisms exhibited by surface-active compounds such as BSs/BEs

Lipopeptide BS produced by *A. junii*disrupted biofilms formed by S. *aureus*, *P. mirabilis*, and *P. aeruginosa* up to 35%, 10%, and 32%, respectively, with the BS concentration at 1250 µgml^{-1}. It was observed by the researchers that further increase in the concentration of the BS up to 2500 µg ml^{-1} increased the disruption of the biofilms up to52%, 31%, and 70% respectively (Ohadi *et al*. 2020). Antibiofilm activity of the purified glycolipoproteinBS produced by *Acinetobacter indicus* M6 was determined against *P. aeruginosa* ATCC 9027 and *S. aureus* ATCC 6538. Biofilm formation was promoted in 96 well plates and the biofilm formed in the plates was exposed to the different concentrations of the BS. After incubation the biofilm was stained using crystal violet and the optical density was also measured at 600 nm. BS showed dose dependent disruption of

the biofilms of *S. aureus*. BS concentration of 500 µg ml^{-1} resulted in around 82.5% inhibition of the biofilms. Decrease in the biofilm was also demonstrated by crystal violet staining (Karlapudi *et al*. 2020).

Anticancer activity: In the last few decades, human civilization has witnessed an increase in the incidence of the different types of cancers, taking a toll of millions of lives.Chemotherapeutic agents used for treating cancers are non-specific and are highly cytotoxic (Adu *et al*. 2022; Wu *et al*. 2017). Therefore, there is an urge for chemotherapeutic agents that can specifically target the cancerous cells in the patients. Many studies and reviews have focused on surface active compounds from microbial origin such as BSs and BEs as promising anticancer agents (Ceresa *et al*. 2023; Imtiaz *et al*. 2022; Wadhawan *et al*. 2022; Walvekar *et al*. 2022; Semkova *et al* 2021; Wu *et al*. 2017).Different mechanisms by which BSs/BEs can induced cell death in cancerous cells are proposed in literature. BSs/BEs can promote cell death in cancer cells by enzyme activation pathway, mitochondrial pathway and cell cycle regulation pathway. Sophorolipids can slow cell growth and therefore promote apoptosis in cancer cells (Wang *et al*. 2024; Sonawane *et al*. 2021; Ceresa *et al*. 2021). BSs/BEs can also promote death of cancer cells by activation of WBCs such as natural killer cells, reducing the process of angiogenesis and by disrupting the cell membranes (Magalhães *et al*. 2021; Dey *et al*. 2015). Figure 2 illustrates schematically different proposed mechanism for anticancer activity exhibited by surface active compounds such as BSs/BEs.

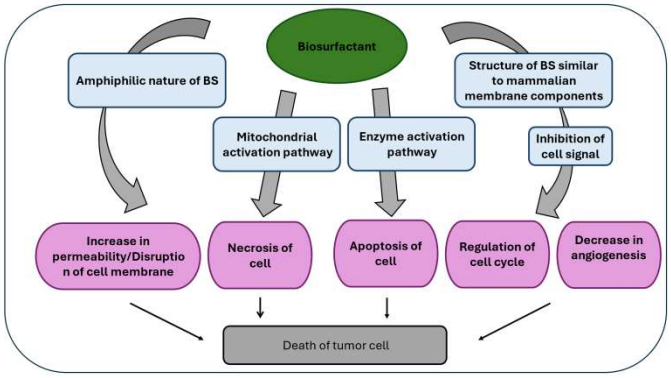

Figure 2. Different mechanisms of anticancer activity of surface-active compounds such as BSs/BEs

The cytotoxic effect of lipopeptide BS produced by *A. junii* was studied on two cancer cell lines (U87 and KB) and normal cells (HUVEC) by WST-1 assay. This study confirmed that the cytotoxic effect of the BS was higher on KB cells as compared to U87 cells. The results further exhibited that the normal cells (HUVEC) showed higher viability (74%) than the cancer cells (64% and 65% for U87 and KB, respectively) (Ohadi *et al*. 2020). Anticancer activity of the purified glycolipoproteinBS produced by *Acinetobacter indicus* M6 was determined against A549 lung cancer cell line. Non-tumorous mouse fibroblast cell line MC-3T3-E1 was used to check the cytotoxicity of the BS. Four different concentrations (50, 100, 200, 500 µg ml^{-1}) of the purified BS were tested against A549 lung cancer cell line and non-tumorous mouse fibroblast cell line MC-3T3-E1 at different incubation times. Cell viability was evaluated by MTTmethod. BS showed decrease in the percentage of the lung cancer viable cells with increase in concentration and incubation times as well. Significant decrease in the percentage of the lung cancer cells was observed at concentration of 200 µg ml^{-1} at 72 h. Different concentrations of the BS did not affect the cell viability of the non-tumour cell line. Cell cycle analysis of the cells exposed to 500 µg ml^{-1} of BS for 24 hwas performed by flow cytometry. BS showed G1 arrest and decreased the viable cells during S phase in the A549 lung cancer cell line whereas it did not affect the cell viability of the non-tumorous mouse fibroblast cell line MC-3T3-E. Thus, this illustrates the non-toxic nature of the BS against normal cells (Karlapudi *et al*. 2020).

Antioxidant activity: Biological free radicals such as ROS and RNS are highly reactive species, as these radicals have an unpaired electron that can react with various biomolecules such as lipids, DNA and proteins associated with a cell (Chaudhary *et al*. 2023; Pham-Huy *et al*. 2008). Free radicals can be endogenous or exogenous in origin, however body keeps balances production of free radicals with antioxidant defence. If this balance fails, it leads to the oxidative stress (Pedro *et al*. 2022; Sen and Chakraborty 2011). Antioxidants are compounds that can neutralize the free radicals by reducing these unstable compounds and thus protect the cells from the deleterious effect of oxidative stress (Sen and Chakraborty 2011; Pham-Huy *et al*. 2008). Action of antioxidants is illustrated in Fig. No. 3. In the last few decades, there has been increase in the exploration of biomaterial with antioxidant properties which can be put forth for the therapeutic application to manage diseases related to oxidative stress (Pedro *et al*. 2022). Recent discoveries reveal the antioxidant property of SAC's such as BS (Abdollahi *et al*. 2020).

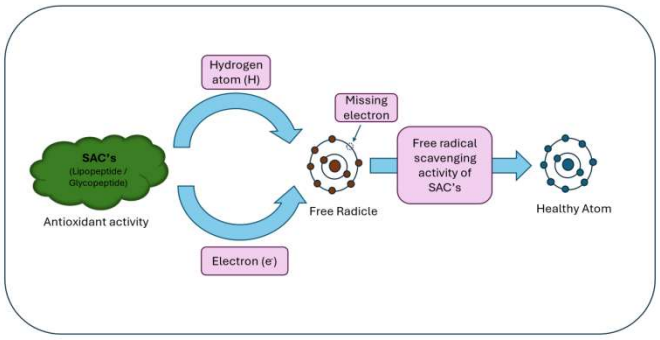

Figure 3. Diagrammatic representation of the antioxidant mechanism of surface-active compounds such as BSs/BEs

Lipopeptide biosurfactant (LBS) produced by *Acinetobacterjunii* B6 was investigated for antioxidant mediated wound healing activity. Antioxidant activity was determined by DPPH radical scavenging activitiy and FRAP assays. DPPH assay exhibited scavenging activity in dose dependent manner with IC50 value of 0.7 mg/ml. Researchers of this study reported that the antioxidant activity of the LBS played a significant role in wound healing with experiments in laboratory animals (Ohadi, *et al*. 2018). Extracellular polysaccharide (ECP's) produced by *Acinetobacter* spp. exhibit diverse medicinal properties. These ECP's are being also proved as good emulsifiers. Antioxidant activity of purifiedECP produced by *Acinetobacterindicus* M6, was studied by different methods such as, hydroxyl radical scavenging activity, super-oxide radical scavenging assay by phenazine methosul-fate (PMS)-nicotinamide adenine dinucleotide (NADH)-Nitroblue tetrazolium chloride (NBT) systemand 1,1-diphe-nyl-2-picrylhydrazyl (DPPH) radical scavenging activity. ECP produced by the *Acinetobacterindicus* M6exhibited significant antioxidant potential with 59% of hydroxyl radical scavenging activity at a concentration of 500 µg/mL, 72.4% of superoxide radical scavenging activity at a concentration of 300 µg/mL, and 72.2% of $DPPH^{\cdot}$ radical scavenging activity at a concentration of 500 µg/mL (Teja *et al*. 2021).

CONCLUSIONS

Microbial surfactants are ecofriendly molecules which exhibit diverse functional properties. Microbial world is a producer of different types of BSs/BEs. Genus *Acinetobacter* is one of the potent producers of varied BSs and BEs, synthesized by variety of different species. Though genus *Acinetobacter* is reported decades back to produce microbial surfactants, researchers even today focus to explore the different types of BSs/BEs produced by *Acinetobacter* spp. Exploring the genetics involved in surfactant produced by *Acinetobacter* will help to increase the yield of the surfactants. Various surfactants produced by genus *Acinetobacter* have showcased widespread applicationsin biomedical field, as antimicrobial, antibiofilm, anticancer and antioxidant agents. To make commercial production of microbial surfactants cost effective, researchers have focused on use

of be low-cost raw materials. Thus, BSs/BEs produced by genus *Acinetobacter* holds a promising candidature for application in medicine.

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Statements and Declarations

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REFERENCES

Abdollahi, S., Zahra Tofighi, Tahereh Babaee, Shamsi, M., Rahimzadeh, G., Hossein Rezvanifar, Elaheh Saeidi, Morteza Mohajeri Amiri, Yasaman Saffari Ashtiani and Samadi, N. (2020). Evaluation of Anti-oxidant and Anti-biofilm Activities of Biogenic Surfactants Derived from Bacillus amyloliquefaciens and Pseudomonas aeruginosa. *PubMed*, 19(2), pp.115–126. doi:https://doi.org/10.22037/ijpr.2020.1101033.

Adetunji, A.I. and Olaniran, A.O. (2019). Production and characterization of bioemulsifiers from *Acinetobacter* strains isolated from lipid-rich wastewater. *3 Biotech*, 9(4). doi:https://doi.org/10.1007/s13205-019-1683-y.

Adu, S.A., Twigg, M.S., Naughton, P.J., Marchant, R. and Banat, I.M. (2022). Biosurfactants as Anticancer Agents: Glycolipids Affect Skin Cells in a Differential Manner Dependent on Chemical Structure. *Pharmaceutics*, 14(2), p.360. doi:https://doi.org/10.3390/pharmaceutics14020360.

Akbari, E., Beheshti Maal, K., Rasekh, B., Emami, Z. D. and Omidi, M. (2020). Lubrication and oil recovery by biosurfactant produced by *Acinetobacterjohnsonii* ABR6. *Journal of Microbial World*, 13(2), pp.154-64.

Banat, I.M., Makkar, R.S. and Cameotra, S.S. (2000). Potential commercial applications of microbial surfactants. *Applied Microbiology and Biotechnology*, 53(5), pp.495–508. doi: https://doi.org/10.1007/s002530051648.

Banat, I.M., Satpute, S.K., Cameotra, S.S., Patil, R. and Nyayanit, N.V. (2014). Cost effective technologies and renewable substrates for biosurfactantsâ€™ production. *Frontiers in Microbiology*, 5. doi:https://doi.org/10.3389/fmicb.2014.00697.

Bao, M., Pi, Y., Wang, L., Sun, P., Li, Y. and Cao, L. (2014). Lipopeptide biosurfactant production bacteria Acinetobacter sp. D3-2 and its biodegradation of crude oil. *Environ. Sci.: Processes Impacts*, 16(4), pp.897–903. doi:https://doi.org/10.1039/c3em00600j.

Bashetti, S.P., Palande, P.P., Mankar, S.G., Bhuyan, S.S., Chopade, B.A. and Mujumdar, S.S. (2012). Studies on bioemulsifier production by *Acinetobactercalcoaceticus* C42 isolated from rhizosphere of corn. *International Journal of Institutional Pharmacy and Life Sciences*, 2(3), pp.95-114

Bergey, D.H. (1930). *Bergey's Manual of Determinative Bacteriology*.

Bhawsar, S.D., Patil, S.D. and Chopade, B.A. (2011). Antimicrobial Activity of Purified Emulsifier of *Acinetobacter* Genospecies Isolated from Rhizosphere of Wheat. *Agricultural science digest*, 31(4), pp.239–246.

Bhuyan, S.S. (2011). *Studies on biosurfactant_ bioemulsifier by acinetobacter genospecies and brevibacteriumhalotolerans isolated from marine environments*. [Thesis Title] Available at: http://hdl.handle.net/10603/135067.

Ceresa, C., Fracchia, L., Andrea Chiara Sansotera, Dìaz, A. and Banat, I.M. (2023). Harnessing the Potential of Biosurfactants for Biomedical and Pharmaceutical Applications. *Pharmaceutics*, 15(8), pp.2156–2156. doi: https://doi.org/10.3390/pharmaceutics15082156.

Ceresa, C., Fracchia, L., Fedeli, E., Porta, C. and Banat, I.M. (2021). Recent Advances in Biomedical, Therapeutic and Pharmaceutical Applications of Microbial Surfactants. *Pharmaceutics*, 13(4), p.466. doi:https://doi.org/10.3390/pharmaceutics13040466.

Chabhadiya, S., Acharya, D.K., Mangrola, A., Shah, R. and Pithawala, E.A. (2024). Unlocking the potential of biosurfactants: Innovations in metabolic and genetic engineering for sustainable industrial and environmental solutions. *Biotechnology Notes*, [online] 5, pp.111–119. doi: https://doi.org/10.1016/j.biotno.2024.07.001.

Chakraborty, J., Chakrabarti, S. and Das, S. (2014). Characterization and antimicrobial properties of lipopeptide biosurfactants produced by *Bacillussubtilis* SJ301 and *Bacillusvallismortis* JB201. *Applied Biochemistry and Microbiology*, 50(6), pp.609–618. doi:https://doi.org/10.1134/s0003683814060039.

Chanika SaengeChooklin, NatthapornRattanapan, AtipanSaimmai, WiboonRiansa-ngawong and SuppasilManeerat (2023). Screening of Biosurfactant-Producing Bacteria as a Potential Biological Control Agent for Fungal Orchid Pathogens in Thailand. *Science & Technology Asia*, [online] pp.292–312. doi:https://doi.org/10.1094/PHP-2010-0614-01--DG.

Chaudhary, P., Pracheta Janmeda, Anca Oana Docea, Balakyz Yeskaliyeva, Faizal, A., Babagana Modu, Calina, D. and Javad Sharifi-Rad (2023). Oxidative stress, free radicals and antioxidants: potential crosstalk in the pathophysiology of human diseases. *Frontiers in Chemistry*, 11. doi:https://doi.org/10.3389/fchem.2023.1158198.

Christofi, N. and Ivshina, I.B. (2002). Microbial surfactants and their use in field studies of soil remediation. *Journal of Applied Microbiology*, 93(6), pp.915–929. doi:https://doi.org/10.1046/j.1365-2672.2002.01774.x.

D'Almeida, A.P., Cristina, D., Maria, V., Tiago and Valderez, M. (2024). Bioemulsifier production by *Acinetobactervenetianus* AMO1502: Potential for bioremediation and environmentally friendly applications. *Marine Pollution Bulletin*, 203, pp.116436–116436. doi:https://doi.org/10.1016/j.marpolbul.2024.116436.

De Giani, A., Zampolli, J. and Di Gennaro, P. (2021). Recent Trends on Biosurfactants With Antimicrobial Activity Produced by Bacteria Associated With Human Health: Different Perspectives on Their Properties, Challenges, and Potential Applications. *Frontiers in Microbiology*, 12. doi: https://doi.org/10.3389/fmicb.2021.655150.

Desai, J.D. and Banat, I.M. (1997). Microbial production of surfactants and their commercial potential. *Microbiology and Molecular Biology Reviews*, 61(1), pp.47–64. doi:https://doi.org/10.1128/mmbr.61.1.47-64.1997.

Dey, G., Rashmi Bharti, Sen, R. and Mandal, M. (2015). Microbial amphiphiles: a class of promising new-generation anticancer agents. *Drug Discovery Today*, 20(1), pp.136–146. doi:https://doi.org/10.1016/j.drudis.2014.09.006.

Doghri, I., Brian-Jaisson, F., Graber, M., Bazire, A., Dufour, A., Bellon-Fontaine, M.-N., Herry, J.-M., Ferro, A.C., Sopena, V., Lanneluc, I. and Sablé, S. (2020). Antibiofilm activity in the culture supernatant of a marine *Pseudomonas* sp. bacterium. *Microbiology*, 166(3), pp.239–252. doi: https://doi.org/10.1099/mic.0.000878.

Dong, H., Xia, W., Dong, H., She, Y., Zhu, P., Liang, K., Zhang, Z., Liang, C., Zhang, S., Sun, S. and Zhang, G. (2016). Rhamnolipids Produced by Indigenous *Acinetobacterjunii* from Petroleum Reservoir and its Potential in Enhanced Oil Recovery. *Frontiers in Microbiology*, 7. doi:https://doi.org/10.3389/fmicb.2016.01710.

e Silva, S.S., Carvalho, J.W.P., Aires, C.P. and Nitschke, M. (2017). Disruption of *Staphylococcusaureus* biofilms using rhamnolipid biosurfactants. *Journal of Dairy Science*, 100(10), pp.7864–7873. doi:https://doi.org/10.3168/jds.2017-13012.

Fondi, M., Maida, I., Perrin, E., Orlandini, V., La Torre, L., Bosi, E., Negroni, A., Zanaroli, G., Fava, F., Decorosi, F., Giovannetti, L., Viti, C., Vaneechoutte, M., Dijkshoorn, L. and Fani, R. (2016). Genomic and phenotypic characterization of the species Acinetobacter venetianus. *Scientific Reports*, 6(1). doi: https://doi.org/10.1038/srep21985.

Fracchia, L., Cavallo, M., Martinotti, M.G. and Banat, I.M. (2012) Biosurfactants and bioemulsifiers biomedical and related applications–present status and future potentials, In: Prof.

Dhanjoo N. Ghista (Ed.). Biomedical science, engineering and technology. Jan 20;14(1), pp.1-49.

Gharaei-Fa, E. (2011). Biosurfactants in Pharmaceutical Industry (A Mini-Review). *American Journal of Drug Discovery and Development*, 1(1), pp.58–69. doi: https://doi.org/10.3923/ajdd.2011.58.69.

Gudiña, E.J., Fernandes, E.C., Teixeira, J.A. and Rodrigues, L.R. (2015). Antimicrobial and anti-adhesive activities of cell-bound biosurfactant from Lactobacillus agilis CCUG31450. *RSC Advances*, 5(110), pp.90960–90968. doi:https://doi.org/10.1039/c5ra11659g.

Gudiña, E.J., Rangarajan, V., Sen, R. and Rodrigues, L.R. (2013). Potential therapeutic applications of biosurfactants. *Trends in Pharmacological Sciences*, 34(12), pp.667–675. doi: https://doi.org/10.1016/j.tips.2013.10.002.

Gupta, B., Puri, S., Thakur, I.S. and Kaur, J. (2020). Enhanced pyrene degradation by a biosurfactant producing *Acinetobacterbaumannii* BJ5: Growth kinetics, toxicity and substrate inhibition studies. *Environmental Technology & Innovation*, 19, p.100804. doi: https://doi.org/10.1016/j.eti.2020.100804.

Harshada, K. (2014). Biosurfactant: A Potent Antimicrobial Agent. *Journal of Microbiology & Experimentation*, 1(5). doi:https://doi.org/10.15406/jmen.2014.01.00031.

Hassanshahian, M. and Ravan, H. (2018). Screening and identification of biosurfactant producing marine bacteria from the Caspian Sea. *Caspian Journal of Environmental Sciences*, 16, pp.179-189. doi:10.22124/CJES.2018.2959

Hyder, N.H. (2015). Production, Characterization and Antimicrobial Activity of a Bioemulsifier Produced by *Acinetobacterbaumanii* AC5 Utilizing Edible Oils. *Iraqi Journal of Biotechnology*, 14(2).

Imtiaz, S., Bashir, M., Banoo, S., Ahmed, M.I. and Anwar, N. (2022) Antitumor and anticancer activity of biosurfactant, In: Green sustainable process for chemical and environmental engineering and science. Academic Press, pp 495-513.

Inès, M. and Dhouha, G. (2015). Glycolipid biosurfactants: Potential related biomedical and biotechnological applications. *Carbohydrate Research*, 416, pp.59–69. doi:https://doi.org/10.1016/j.carres.2015.07.016.

Jadeja, N.B., Moharir, P. and Kapley, A. (2018). Genome Sequencing and Analysis of Strains *Bacillus* sp. AKBS9 and *Acinetobacter* sp. AKBS16 for Biosurfactant Production and Bioremediation. *Applied Biochemistry and Biotechnology*, 187(2), pp.518–530. doi:https://doi.org/10.1007/s12010-018-2828-x.

Jagtap, S., Yavankar, S., Pardesi, K., Chopade, B. (2010). Production of bioemulsifier by *Acinetobacter* species isolated from healthy human skin. *Indian Journal of Experimental Biology*, 48(1), pp.70-76.

Jemil, N., Ben Ayed, H., Manresa, A., Nasri, M. and Hmidet, N. (2017). Antioxidant properties, antimicrobial and anti-adhesive activities of DCS1 lipopeptides from *Bacillusmethylotrophicus* DCS1. *BMC Microbiology*, 17(1). doi:https://doi.org/10.1186/s12866-017-1050-2.

Karlapudi, A.P., T.C., V., Srirama, K., Kota, R.K., Mikkili, I. and Kodali, V.P. (2020). Evaluation of anti-cancer, anti-microbial and anti-biofilm potential of biosurfactant extracted from an *Acinetobacter* M6 strain. *Journal of King Saud University - Science*, 32(1), pp.223–227. doi:https://doi.org/10.1016/j.jksus.2018.04.007.

Kiran, G.S., Sabarathnam, B. and Selvin, J. (2010). Biofilm disruption potential of a glycolipid biosurfactant from marine *Brevibacterium casei*. *FEMS Immunology & Medical Microbiology*, 59(3), pp.432–438. doi: https://doi.org/10.1111/j.1574-695x.2010.00698.x.

Kuyukina, M.S., Ivshina, I.B., Philp, J.C., Christofi, N., Dunbar, S.A. and Ritchkova, M.I. (2001). Recovery of *Rhodococcus* biosurfactants using methyl tertiary-butyl ether extraction. *Journal of Microbiological Methods*, 46(2), pp.149–156. doi: https://doi.org/10.1016/s0167-7012(01)00259-7.

Magalhães, F.F., Nunes JC, Araújo, M.T., Ferreira, A.M., Almeida, M.R., Freire, M.G., Tavares, A.P. (2021) Anti-Cancer Biosurfactants. In: Inamuddin Ahamed MI., Prasad R, editors. Microbial Biosurfactants: Preparation, Properties and Applications, pp. 159-196. https://doi.org/10.1007/978-981-15-6607-3_8

Mehrabani, M., Esmaeili-Tarzi, M., Forootanfar, H., Nematollahi, M.H., Banat, I.M., Ohadi, M. and Dehghannoudeh, G. (2021). Lipopeptide Biosurfactant from *Acinetobacterjunii* B6: A Promising Natural Surfactant for Promoting Angiogenesis. *International Journal of Peptide Research and Therapeutics*, 27(2), pp.1197–1203. doi:https://doi.org/10.1007/s10989-021-10160-9.

Menezes Bento, F., de Oliveira Camargo, F.A., Okeke, B.C. and Frankenberger, W.T. (2005). Diversity of biosurfactant producing microorganisms isolated from soils contaminated with diesel oil. *Microbiological Research*, 160(3), pp.249–255. doi:https://doi.org/10.1016/j.micres.2004.08.005.

Mishra, R., Panda, A.K., De Mandal, S., Shakeel, M., Bisht, S.S. and Khan, J. (2020). Natural Anti-biofilm Agents: Strategies to Control Biofilm-Forming Pathogens. *Frontiers in Microbiology*, 11. doi:https://doi.org/10.3389/fmicb.2020.566325.

Mostafapour, M.J., S Ahmady Abchin and Safari Moein (2014). Isolation and identification of biosurfactant-producing strains from the genus *Acinetobacter* spp. and antibacterial effects of biosurfactant produced on some of the negative and gram-positive bacteria in vitro. *New Cellular and Molecular Biotechnology Journal*, 4(14), pp.79–91.

Mujumdar, S. and Chopade, B.A. (2002). *Studies on isolation distribution biotyping characterization production of antibiotic bioemulsifier and plasmid pUPI 126 encoded indole acetic acid _IAA_ production and its role in plant growth promotion by Acinetobacter species from rhizosphere of wheat*. [Thesis Title] Available at: http://hdl.handle.net/10603/139790.

Muthukamalam, S., Sivagangavathi, S., Dhrishya, D. and Sudha Rani, S. (2017). Characterization of dioxygenases and biosurfactants produced by crude oil degrading soil bacteria. *Brazilian Journal of Microbiology*, 48(4), pp.637–647. doi:https://doi.org/10.1016/j.bjm.2017.02.007.

Nakar, D. and Gutnick, D.L. (2003). Involvement of a Protein Tyrosine Kinase in Production of the Polymeric Bioemulsifier Emulsan from the Oil-Degrading Strain *Acinetobacter lwoffii* RAG-1. 185(3), pp.1001–1009. doi:https://doi.org/10.1128/jb.185.3.1001-1009.2003.

Naughton, P.J., Marchant, R., Naughton, V. and Banat, I.M. (2019). Microbial biosurfactants: current trends and applications in agricultural and biomedical industries. *Journal of Applied Microbiology*, 127(1), pp.12–28. doi:https://doi.org/10.1111/jam.14243.

Navon-Venezia, S., Zosim, Z., Gottlieb, A., Legmann, R., Carmeli, S., Ron, E.Z. and Rosenberg, E. (1995). Alasan, a new bioemulsifier from *Acinetobacterradioresistens*. *Applied and Environmental Microbiology*, 61(9), pp.3240–3244. doi:https://doi.org/10.1128/aem.61.9.3240-3244.1995.

Ndlovu, T., Rautenbach, M., Vosloo, J.A., Khan, S. and Khan, W. (2017). Characterisation and antimicrobial activity of biosurfactant extracts produced by *Bacillusamyloliquefaciens* and *Pseudomonasaeruginosa* isolated from a wastewater treatment plant. *AMB Express*, 7(1). doi:https://doi.org/10.1186/s13568-017-0363-8.

Nemec, A. (2022). *Acinetobacter*. *Bergey's Manual of Systematics of Archaea and Bacteria*, pp.1–78. doi:https://doi.org/10.1002/9781118960608.gbm01203.pub2.

Nikolova, C. and Gutierrez, T. (2023) Novel approaches in the use of biosurfactants in the oil industry and environmental remediation. In: Gloria Soberón-Chávez editors. Foundations and Frontiers in Enzymology, Biosurfactants. Academic Press, pp.107-128. https://doi.org/10.1016/B978-0-323-91697-4.00006-5Ntshingila, N.P., Jimoh, A.A. and Lin, J. (2021). Production and characterisation of a biosurfactant based on *Acinetobacter* sp. V2 and its potential use for environmental applications. *International Journal of Environmental Studies*, 79(6), pp.1099–1122. doi:https://doi.org/10.1080/00207233.2021.1977535.

Ohadi, M., Dehghannoudeh, G., Shakibaie, M., Banat, I.M., Pournamdari, M. and Forootanfar, H. (2017). Isolation,

characterization, and optimization of biosurfactant production by an oil-degrading *Acinetobacterjunii* B6 isolated from an Iranian oil excavation site. *Biocatalysis and Agricultural Biotechnology*, 12, pp.1–9. doi:https://doi.org/10.1016/j.bcab.2017.08.007.

Ohadi, M., Forootanfar, H., Dehghannoudeh, G., Eslaminejad, T., Ameri, A., Shakibaie, M. and Adeli-Sardou, M. (2020). Antimicrobial, anti-biofilm, and anti-proliferative activities of lipopeptide biosurfactant produced by *Acinetobacterjunii* B6. *Microbial Pathogenesis*, 138, p.103806. doi: https://doi.org/10.1016/j.micpath.2019.103806.

Ohadi, M., Forootanfar, H., Rahimi, H.R., Jafari, E., Shakibaie, M., Eslaminejad, T. and Dehghannoudeh, G. (2018). Antioxidant Potential and Wound Healing Activity of Biosurfactant Produced by Acinetobacter junii B6. *Current Pharmaceutical Biotechnology*, 18(11), pp.900–908. doi:https://doi.org/10.2174/1389201018666171122121350.

Okoliegbe, I. N. and Agarry, O. O. (2012). Application of microbial surfactant (a review). *Scholarly journals of biotechnology*, *1*(1), pp.15-23.

Onajobi, I.B., Adeyemi, J.O., Orji, F.A., Samson, O.J., Egberongbe, H.O., Aina, S.A., Afolabi, O.T., Adebajo, L.O. and Fagade, O.E. (2023). Characterization of biosurfactant-producing bacterial strains isolated from agro-industrial wastes in southwestern, Nigeria. *Microbes, Infection and Chemotherapy*, 3, p.e1586. doi:https://doi.org/10.54034/mic.e1586.

Ortega-de la Rosa, N.D., Vázquez-Vázquez, J.L., Huerta-Ochoa, S., Gimeno, M. and Gutiérrez-Rojas, M. (2018). Stable bioemulsifiers are produced by *Acinetobacterbouvetii* UAM25 growing in different carbon sources. *Bioprocess and Biosystems Engineering*, 41(6), pp.859–869. doi:https://doi.org/10.1007/s00449-018-1920-5.

Parthipan, P., Elumalai, P., Sathishkumar, K., Sabarinathan, D., Murugan, K., Benelli, G. and Rajasekar, A. (2017). Biosurfactant and enzyme mediated crude oil degradation by *Pseudomonasstutzeri* NA3 and *Acinetobacterbaumannii* MN3. *3 Biotech*, 7(5). doi:https://doi.org/10.1007/s13205-017-0902-7.

Pedro, A.C., Paniz, O.G., Fernandes, I. de A.A., Bortolini, D.G., Rubio, F.T.V., Haminiuk, C.W.I., Maciel, G.M. and Magalhães, W.L.E. (2022). The Importance of Antioxidant Biomaterials in Human Health and Technological Innovation: A Review. *Antioxidants*, [online] 11(9), p.1644. doi: https://doi.org/10.3390/antiox11091644.

Peele, K.A., Ch., V.R.T. and Kodali, V.P. (2016). Emulsifying activity of a biosurfactant produced by a marine bacterium. *3 Biotech*, [online] 6(2). doi:https://doi.org/10.1007/s13205-016-0494-7.

Pendse, A. and Aruna, K. (2020). The Antimicrobial Activity of Biosurfactant Isolated from *Serratiarubidaea* KAP Against β-Lactamase Producers and its Enviromental Application. *International Journal of Research and Analytical Reviews*, 7(1), pp.553-568.

Pham-Huy, L. A., He, H., & Pham-Huy, C. (2008). Free radicals, antioxidants in disease and health. *International journal of biomedical science*, 4(2), pp.89–96.

Phetrong, K., Aran, H. and Maneerat, S. (2008). Production and characterization of bioemulsifier from a marine bacterium, *Acinetobactercalcoaceticus* subsp. *anitratus* SM7. *Songklanakarin Journal of science & technology*, 30(3), pp.297.

Pirog, T.P., Konon, A.D., Shevchuk, T.A. and Bilets, I.V. (2012). Intensification of biosurfactant synthesis by *Acinetobactercalcoaceticus* IMV B-7241 on a hexadecane-glycerol mixture. *Microbiology*, 81(5), pp.565–572. doi:https://doi.org/10.1134/s0026261712050128.

Prasad, B., Kaur, H. P., & Kaur, S. (2015). Potential biomedical and pharmaceutical applications of microbial surfactants. *World J Pharm Pharm Sci*, 4, pp.1557-75.

Rahman, P.K.S.M. and Gakpe, E. (2008). Production, Characterisation and Applications of Biosurfactants-Review. *Biotechnology (Faisalabad)*, 7(2), pp.360–370. doi: https://doi.org/10.3923/biotech.2008.360.370.

Rodrigues, L., Banat, I.M., Teixeira, J. and Oliveira, R. (2006). Biosurfactants: potential applications in medicine. *Journal of Antimicrobial Chemotherapy*, 57(4), pp.609–618. doi: https://doi.org/10.1093/jac/dkl024.

Rodrigues, L.R. and Teixeira, J.A. (2010). Biomedical and therapeutic applications of biosurfactants. *Advances in Experimental Medicine and Biology*, pp.75–87. doi:https://doi.org/10.1007/978-1-4419-5979-9_6.

Sahu, L. and Shrivastava, R. (2022). Characterization of biosurfactant produced by hydrocarbon-degrading Bacteria *Acinetobacterjunii* isolated from petroleum hydrocarbon-contaminated soil. *Bulletin of Environment, Pharmacology and Life Sciences*, 12(1), pp.110-117.

Sandeep, L. and Rajasree, S. (2017). Biosurfactant: Pharmaceutical Perspective. *Journal of Analytical & Pharmaceutical Research*, 4(3). doi:https://doi.org/10.15406/japlr.2017.04.00105.

Santos, D.K.F., Rufino, R.D., Luna, J.M., Santos, V.A. and Sarubbo, L.A. (2016). Biosurfactants: Multifunctional Biomolecules of the 21st Century. *International Journal of Molecular Sciences*, [online] 17(3). doi:https://doi.org/10.3390/ijms17030401.

Satpute, S.K., Banpurkar, A.G., Dhakephalkar, P.K., Banat, I.M. and Chopade, B.A. (2010). Methods for investigating biosurfactants and bioemulsifiers: a review. *Critical Reviews in Biotechnology*, 30(2), pp.127–144. doi: https://doi.org/10.3109/07388550903427280.

Sawant, R., Devale, A., Mujumdar, S., Pardesi K. and Shouche, Y. (2021). Promising Strategies for Economical Production of Biosurfactants the Green Molecules. In: Sayyed RZ, El-Enshasy HA, Hameeda B, editors.Microbial Surfactants Volume I: Production and Applications. CRC Press, pp. 266-286.

Semkova, S., Antov, G., Iliev, I., Tsoneva, I., Lefterov, P., Christova, N., Nacheva, L., Stoineva, I., Kabaivanova, L., Staneva, G. and Nikolova, B. (2021). Rhamnolipid Biosurfactants—Possible Natural Anticancer Agents and Autophagy Inhibitors. *Separations*, 8(7), p.92. doi: https://doi.org/10.3390/separations8070092.

Sen, S. and Chakraborty, R. (2011). The Role of Antioxidants in Human Health. In: S. Andreescu and M. Hepel, eds., *Oxidative Stress: Diagnostics, Prevention, and Therapy.* [online] American Chemical Society, pp.1–37. Available at: doi:10.1021/bk-2011-1083.ch001.

Shabtai, Y. and Gutnick, D.L. (1985). Exocellular esterase and emulsan release from the cell surface of *Acinetobactercalcoaceticus*. *Journal of Bacteriology*, 161(3), pp.1176–1181. doi:https://doi.org/10.1128/jb.161.3.1176-1181.1985.

Shah, N., Nikam, R., Gaikwad, S., Sapre, V. and Kaur, J. (2016). Biosurfactant: Types, Detection Methods, Importance and Applications. *Indian Journal of Microbiology Research*, 3(1), p.5. doi:https://doi.org/10.5958/2394-5478.2016.00002.9.

Sharma, D. and Saharan, B.S. (2016). Functional characterization of biomedical potential of biosurfactant produced by *Lactobacillushelveticus*. *Biotechnology Reports*, 11, pp.27–35. doi:https://doi.org/10.1016/j.btre.2016.05.001.

Sharma, D., Ansari, M.J., Gupta, S., Al Ghamdi, A., Pruthi, P. and Pruthi, V. (2015). Structural Characterization and Antimicrobial Activity of a Biosurfactant Obtained From*Bacilluspumilus* DSVP18 Grown on Potato Peels. *Jundishapur Journal of Microbiology*, 8(9). doi:https://doi.org/10.5812/jjm.21257.

Shekhar, S., Sundaramanickam, A. and Balasubramanian, T. (2014). Biosurfactant Producing Microbes and their Potential Applications: A Review. *Critical Reviews in Environmental Science and Technology*, 45(14), pp.1522–1554. doi:https://doi.org/10.1080/10643389.2014.955631.

Shete, A.M., Mujumdar, S.S., Bhuyan-Pawar S. and Chopade, B.A. (2015). Isolation, biotyping, biochemical and physiological characterization of marine *Acinetobacter* isolated from west coast of India. *International Journal of Current Microbiology and Applied Sciences*, 2, pp.277-301.

Shobe, E., Akhlaq, F., Badar, U., Akhter, J. and Imtiaz, S. (2013). Classification and industrial applications of biosurfactants. *Academic Research International*, 4(3), pp.243.

Singh, P. and Cameotra, S.S. (2004). Potential applications of microbial surfactants in biomedical sciences. *Trends in*

Biotechnology, 22(3), pp.142–146. doi:https://doi.org/10.1016/j.tibtech.2004.01.010.

Sivapathasekaran, C. and Sen, R. (2017). Origin, properties, production and purification of microbial surfactants as molecules with immense commercial potential. *Tenside Surfactants Detergents*, 54(2), pp.92-107.

Sobrinho, H., Luna, J., Rufino, R., Porto, A. and Sarubbo, L. (2014). Biosurfactants: classification, properties and environmental applications. In: J.N. Govil, editors. Recent Developments in Biotechnology. Studium Press LLC, USA, pp. 303-330.

Sonawane SS, Kumbhare SV, Patil NP (2021) Biomedical Application of Biosurfactants. In: Sayyed RZ, El-Enshasy HA, Hameeda B, editors.Microbial Surfactants Volume I: Production and Applications. CRC Press, pp. 19-173.

Tanaka, Y., Fukui, T. and Negi, T. (1990). *Novel Acinetobacter calcoaceticus and novel biosurfactant*. No. EP0401700A2. December 12, 1990

Teja, C.R., Karlapudi, A.P., Neeraja Vallur, K. Mamatha, Babu, D.J., T.C. Venkateswarulu and Kodali, V.P. (2021). Antioxidant potential and optimization of production of extracellular polysaccharide by Acinetobacter indicus M6. *Journal of Genetic Engineering and Biotechnology*, 19(1), pp.39–39. doi:https://doi.org/10.1186/s43141-021-00137-y.

Tomar, S. and Singh, B.P. (2014). Microbial Origin, Classification and Application of Biosurfactant. *Indian Journal of Animal Nutrition*, 7(16), pp.2060-2069.

Toren, A., Orr, E., Paitan, Y., Ron, E.Z. and Rosenberg, E. (2002). The Active Component of the Bioemulsifier Alasan from *Acinetobacter radioresistens* KA53 Is an OmpA-Like Protein. *Journal of Bacteriology*, 184(1), pp.165–170. doi:https://doi.org/10.1128/jb.184.1.165-170.2002.

Triawan, A., Ni'matuzahroh and Supriyanto, A. (2017). Effects of the combination between bio-surfactant product types and washing times on the removal of crude oil in nonwoven fabric. *AIP Conference Proceedings*. doi:https://doi.org/10.1063/1.4985428.

Uzoigwe, C., Burgess, J.G., Ennis, C.J. and Rahman, P.K.S.M. (2015). Bioemulsifiers are not biosurfactants and require different screening approaches. *Frontiers in Microbiology*, 6. doi:https://doi.org/10.3389/fmicb.2015.00245.

Varjani, S.J. and Upasani, V.N. (2017). Critical review on biosurfactant analysis, purification and characterization using rhamnolipid as a model biosurfactant. *Bioresource Technology*, [online] 232, pp.389–397. doi:https://doi.org/10.1016/j.biortech.2017.02.047.

Wadhawan, A., Singh, J., Sharma, H., Handa, S., Singh, G., Kumar, R., Barnwal, R.P., Pal Kaur, I. and Chatterjee, M. (2022). Anticancer Biosurfactant-Loaded PLA–PEG Nanoparticles Induce Apoptosis in Human MDA-MB-231 Breast Cancer Cells. *ACS Omega*, 7(6), pp.5231–5241. doi:https://doi.org/10.1021/acsomega.1c06338.

Walvekar, S., Yasaswi, S., Shetty, K. and Yadav, K.S. (2022). Chapter 11 - Applications of surfactin and other biosurfactants in anticancer activity. In Inamuddin, C. O. Adetunji. and M. I. Ahamed (Eds.), Green Sustainable Process for Chemical and Environmental Engineering and Science, Academic Press, pp. 223–234.

Wang, X., An, J., Cao, T., Guo, M. and Han, F. (2024). Application of Biosurfactants in Medical Sciences. *Molecules*, 29(11), pp.2606–2606. doi:https://doi.org/10.3390/molecules29112606.

Wu, Y.-S., Ngai, S.-C., Goh, B.-H., Chan, K.-G., Lee, L.-H. and Chuah, L.-H. (2017). Anticancer Activities of Surfactin and Potential Application of Nanotechnology Assisted Surfactin Delivery. *Frontiers in Pharmacology*, 8. doi:https://doi.org/10.3389/fphar.2017.00761.

Yuan, H., Yao, J., Masakorala, K., Wang, F., Cai, M. and Yu, C. (2013). Isolation and characterization of a newly isolated pyrene-degrading *Acinetobacter* strain USTB-X. *Environmental Science and Pollution Research*, 21(4), pp.2724–2732. doi:https://doi.org/10.1007/s11356-013-2221-9.

Zhao, Y.-H., Chen, L.-Y., Tian, Z.-J., Sun, Y., Liu, J.-B. and Huang, L. (2015). Characterization and application of a novel bioemulsifier in crude oil degradation by *Acinetobacter beijerinckii*ZRS. *Journal of Basic Microbiology*, 56(2), pp.184–195. doi:https://doi.org/10.1002/jobm.201500487.

Zhao, Z. and Wong, J.W.C. (2009). Biosurfactants from *Acinetobacter calcoaceticus*BU03 enhance the solubility and biodegradation of phenanthrene. *Environmental technology* 30(3), pp.291–299. doi:https://doi.org/10.1080/09593330802630801.

Zobaer, M., Ali, F., Anwar, Md.N., Bappi, M.S.H., Bakar, T.B. and Hossain, T.J. (2023). Isolation of Biosurfactant Producing Bacteria from Oil-Spilled Soil and Characterization of Their Secreted Biosurfactants in Pathogen-Inhibition and Oil-Emulsification. *International Journal of Pharmaceutical Sciences and Research.SSRN Electronic Journal*. doi:https://doi.org/10.2139/ssrn.4320992.

Zou, C., Wang, M., Xing, Y., Lan, G., Ge, T., Yan, X. and Gu, T. (2014). Characterization and optimization of biosurfactants produced by *Acinetobacterbaylyi* ZJ2 isolated from crude oil-contaminated soil sample toward microbial enhanced oil recovery applications. *Biochemical Engineering Journal*, 90, pp.49–58. doi:https://doi.org/10.1016/j.bej.2014.05.007.

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