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Review Article

Simple glycolipids of microbes: Chemistry, biological activity and metabolic engineering

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ABSTRACT

Glycosylated lipids (GLs) are added-value lipid derivatives of great potential. Besides their interesting surface activities that qualify many of them to act as excellent ecological detergents, they have diverse biological activities with promising biomedical and cosmeceutical applications. Glycolipids, especially those of microbial origin, have interesting antimicrobial, anticancer, antiparasitic as well as immunomodulatory activities. Nonetheless, GLs are hardly accessing the market because of their high cost of production. We believe that experience of metabolic engineering (ME) of microbial lipids for biofuel production can now be harnessed towards a successful synthesis of microbial GLs for biomedical and other applications. This review presents chemical groups of bacterial and fungal GLs, their biological activities, their general biosynthetic pathways and an insight on ME strategies for their production.

Contents

1. Introduction

Lipid biotechnology research has focused to date on developing sustainable alternatives to depleting fossil fuels. One strategy was plant-derived fuel, biodiesel [1]. A main drawback of this approach

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is that oil and land allocated for biodiesel production compete with those allocated for human food consumption. Moreover, replacement of natural vegetations with plants used for biodiesel production generates long-term environmental concerns. Another strategy is to use lipids originating from microbes, called "single cell oil" (SCO) as substrates for biodiesel production. We believe that accumulating knowledge and developed biomolecular tools obtained from lipid engineering of oleogenic microbes can now be harnessed for the microbial production of lipid derivatives of added-value.

Glycosylation of organic molecules, including lipids, usually leads to derivatives of new and/or better physicochemical properties and biological activities [2,3] that reflect in higher market prices. Metabolic engineering of lipid derivatives has previously investigated polyunsaturated fatty acids [4] and fatty acid derivatives that are used as substrates for oleochemical industries, e.g. heterologous production of ricinoleic acids by *Y. lipolytica* [5]. Other added-value lipid derivatives of commercial interest include wax esters, polyhydroxyalkanoates (bioplastics), hydroxylated fatty acids, carotenoids, polyenic polymers [6] and glycolipids.

This review focuses on simple glycolipids (SGLs) as an important family of glycolipids (GLs) class. The importance of SGLs stems from the fact that this family of GLs comprises a wide range of bioactive molecules with potential biomedical, pharmaceutical and cosmetic applications [7,8]. Nonetheless, many simple GLs are limited commercially because of their still low yield and high cost of production, particularly of high purity simple GLs aimed for biopharmaceutical purposes.

We present the chemical groups of simple GLs, their microbial producers and their biological activities. Then, we describe the key biosynthetic enzymes and metabolic precursors involved in biosynthesis of simple GLs. Finally, we discuss metabolic engineering strategies for simple GLs production in native and heterologous hosts.

2. Definition and classification of simple glycolipids

The term glycolipids (GLs), in general, encompasses a wide diversity of structurally heterogeneous biological compounds that are produced by microbes, plants, animals and humans [9]. As their names suggest, they are composed of glycosyl and lipid moieties. The IUPAC uses the term GLs to broadly designate any compound containing one or more monosaccharide residues bound by glycosidic linkage to a hydrophobic moiety [10]. Our definition of GLs is even broader to include glycoside and non-glycoside GLs in which the sugar and lipid residues are linked together via glycosidic (e.g. *O*- or *N*-glycosidic linkages) and non-glycosidic linkages (e.g. ester or amide linkages), respectively (Fig. 1). The glycosyl residue can be mono-, di-, oligo or polysaccharides (e.g. glucose, cellobiose or glycan, respectively), alcohol sugars/polyols (like mannitol, erythritol or arabinol, etc.), amino sugars (like desosamine, etc) or sugar acids (like glucuronic acids). The lipid residue of GLs ranges from fatty acids, fatty alcohols, fatty amino alcohols, polyketides, sterols, hopanoids and carotenoids with different substitutions, chain lengths, saturation levels, branching and di-/oligo-/polymerizations.

Numerous classifications exist for GLs [10], the most convenient of which is their classification into simple and complex GLs [11–13] (Fig. 1). Simple GLs (SGLs), sometimes called saccharolipids [14], are two-component (glycosyl and lipid moieties) GLs in which the glycosyl and lipid moieties are directly linked to each other. Complex glycolipids (CGLs) are, however, structurally more heterogeneous, as they contain, in addition to the glycosyl and lipid moieties, other residues like glycerol (glycoglycerolipids), peptide (glycopeptidolipids), acylated-sphingosine (glycosphingolipids), or other residues (Fig. 1). Polysaccharide-containing GLs, although containing no residues other than glycosyl and lipid moieties, are classified under complex glycolipids because of the complex nature of their polysaccharide residues; however, oligosaccharide-containing GLs are classified as simple GLs [13] (Fig. 1). Simple glycolipids addressed in this review are those of natural microbial origin, therefore, SGLs of synthetic or other biological origins are not mentioned.

3. Surfactant properties of simple glycolipids

Simple glycolipids (SGLs) are amphiphilic molecules as they comprise both the hydrophilic glycosyl and the lipophilic lipid residues. This amphiphilic nature confers surfactant activity to most GLs; those of which with pronounced surfactant activity are called biosurfactant. Compared to petroleum-derived (e.g. alkylbenzene sulfonates) or plant-based (e.g. alkyl polyglycosides) synthetic surfactants [15], microbially-produced SGL biosurfactants are mostly of higher surface activity, higher emulsifying power, lower critical micelle concentrations, higher biodegradability (compared to petroleum-derived surfactants), lower ecotoxicity [16] and lower protein denaturing potency [17–19]. The advanced properties of microbial SGLs are suggested to be attributed to a peculiar mosaic distribution of regions of polarity over the GL molecule, as well as to their branched or sometimes circular structures compared to synthetic surfactants [18]. Moreover, most SGLs are naturally produced as complex mixtures of congeners or homologues that vary in the number of glycosyl units and extent of their acylation, the number of conjugate lipid chains, their lengths, the extent of unsaturations and substitutions; these factors together contribute to their unique surfactant properties and behaviors [18].

Although the unique surface properties of some SGLs qualified some of them to be marketed as ecological surfactants [20,21], yet, their competitiveness in the detergent market is limited because of their higher prices compared to alkyl polyglycosides synthetic surfactants which are at least 50% less expensive. For example, the estimated cost of large-scale production of the SGLs: sophorolipid and rhamnolipid biosurfactants, are about US$ 2.5–3/Kg [21,22] and US$ 5–20/Kg [23], respectively, compared to US$ 1–3/Kg for the synthetic alkyl polyglycoside surfactants [23].

Aside from their surface activities, nearly all natural SGLs have interesting biological activities, as described later, that let them occupy market niches not approachable by synthetic surfactants [24]. Noteworthy, the biological activities of SGLs are thought to stem from their surface activities [25].

4. Chemical groups and origins of microbial simple glycolipids

Microbially produced SGLs are classified in chemical groups based on their chemical structures so that every group comprises SGLs members sharing unique glycosyl and/or lipid moieties for SGLs produced by bacteria (Table 1) and fungi (Table 2). In this classification, some SGLs congeners are classified in separate groups when they originate from different microbial origins and vice versa. Under each SGL group, exhaustive list of its members, together with their chemical names, their microbial producers as well as their taxonomic phyla is mentioned (Tables 1 and 2). Furthermore, the confirmed chemical structures of representative or prototypic members of each SGL group are presented (Fig. 3).

Based on our survey of microorganisms producing SGLs, we found that 50% of all known microbial SGLs are produced by microbes belonging to the phylum *Actinobacteria* (Fig. 2). Second in rank to *Actinobacteria*, comes phylum *Proteobacteria* followed by

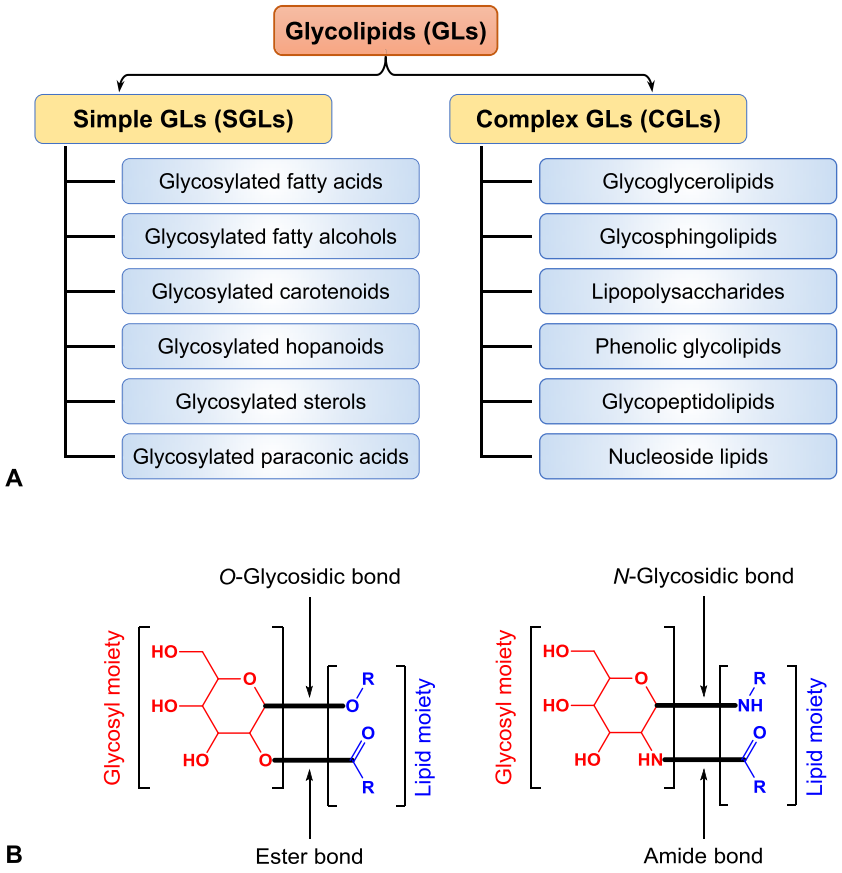

Fig. 1. Classification of glycolipids and main types of linkages between their glycosyl and lipid residues. **(A)** Simple glycolipids (SGLs) comprise glycolipids consisting of glycosyl and lipid residues only, whereas, complex glycolipids (CGLs) contain glycerol, ceramide, phenolic, peptide, nucleoside or polysaccharide residues in addition to the glycosyl and lipid residues. **(B)** Glycosyl and lipid residues are mainly linked via *O*-glycosidic and/or ester bonds, and less frequently via *N*-glycosidic and/or amide bonds.

the two major fungal phyla, *Ascomycota* and *Basidiomycota,* consecutively (Fig. 2).

4.1. Bacterial simple glycolipids

Overall, bacterially produced simple glycolipids (SGLs) outnumbers fungally-produced ones (Fig. 2). A previous survey of about 16000 pooled natural bacterial metabolites revealed that about 20% of them are glycosylated, about 30% of these glycosylated metabolites are glycosylated lipids of which glycosylated macrolactones/-lactams take a share of about 20% and other glycosylated lipids (including SGLs) take a share of 10% [26]. Nearly all glycosylated macro-lactones/-lactams are produced by members of the phylum *Actinobacteria*. We classified baterially produced SGLs in 10 groups (Table 1).

4.2. Fungal simple glycolipids

Fungally produced simple glycolipids (SGLs) are less numerous than bacterially-produced ones (Fig. 2). Fungal SGLs are classified in 10 groups that are mainly produced by members of the phyla *Ascomycota* and *Basidiomycota* (Table 2).

5. Physiological roles of simple glycolipids

For most SGLs, the exact physiological roles to their native producers are not clearly known. Generally, SGLs are secondary metabolites that are not essential for cell viability. Nonetheless, given their antimicrobial properties, SGLs are suggested to help producing organism dominate environmental niches by inhibiting the growth of other organisms [153]. In addition, SGLs are required to coordinate multicellular or group behaviors (biofilm formation and swarming) and enhance growth of producing organisms on hydrophobic carbon sources [154–156]. Some additional roles are assigned to specific SGLs like rhamnolipids, which are considered as virulence factors that modulate host immune response [29]. Similarly to their unglycosylated counterparts, glycosylated carotenoids are postulated to act as photoprotectants and antioxidants to protect organisms from injuries caused by free radicals and active oxygen species [106]. In thermophiles, glycocarotenoids are thought to stabilize and reinforce cell membranes [113]. Hopanoids are sterol analogues in bacteria. Similarly to sterol in eukaryotes, hopanoids and their glycosylated derivatives are thought to help stabilize and regulate membrane fluidity and permeability particularly during shifts in pH and other physicochemical conditions [117,157]. Sophorolipids are suggested to act as extracellular forms of carbon storage that can be recycled later under starvation conditions [154].

6. Bioactivities of simple glycolipids

Simple glycolipids (SGLs) have very interesting biological activities on other organisms ranging from viruses to human cells. Although the mechanism of these bioactivities is not definitively known, it is suggested that most of SGLs bioactivities arise from their surface activities. Collectively, many of them have antiviral,

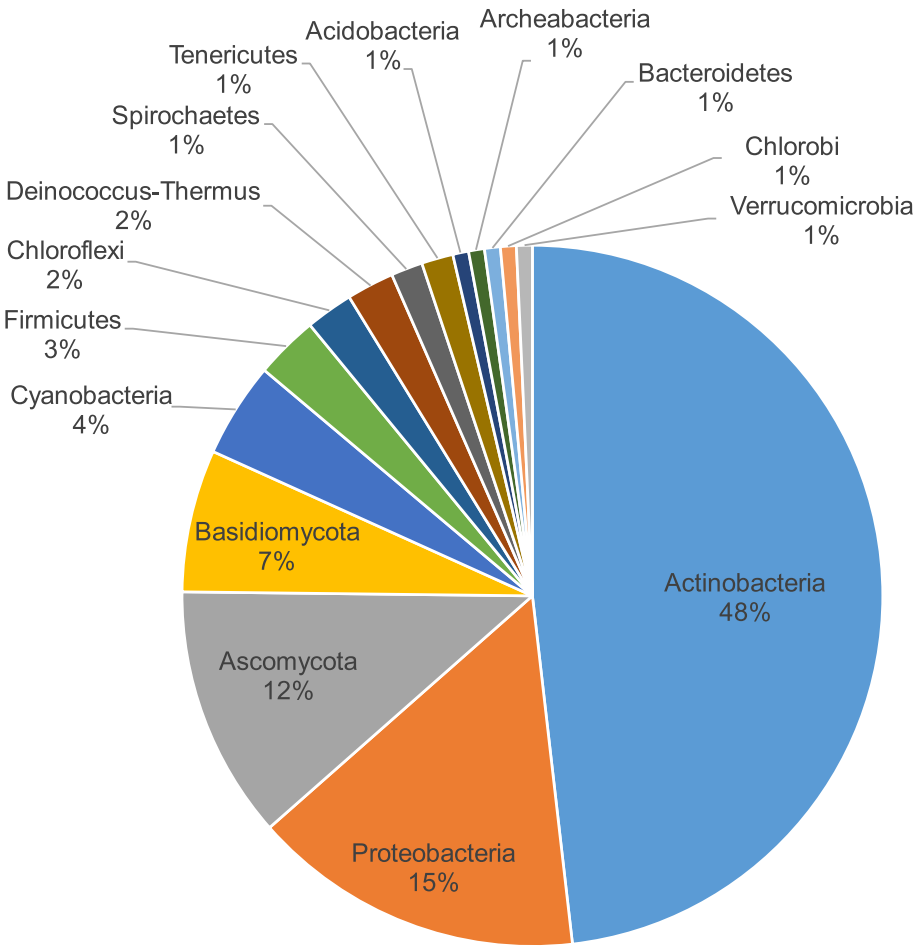

Fig. 2. Approximate distribution of microbial producers of simple glycolipids in different bacterial and fungal phyla. Incidences of microbial production of chemically unique SGLs in every microbial phylum were counted. Homologues or stereoisomers of the same chemically unique SGLs did not add into these calculations to avoid false overestimations. As an example, rhamnolipids (RLs) exist in two unique structures known so far, mono-rhamnolipids and di-rhamnolipids containing one and two rhamnose moieties, respectively, and are produced by proteobacterial species. Although these two RL congeners has several homologues varying in chain length of their lipid moiety, they were counted as two chemically unique SGLs in our calculations. Only the phylum of the microbial producer and not its genus and species identity that was taken into account; for example, although di-rhamnolipids are produced by different species of the genus *Pseudomonas* and *Burkholderia*, all di-rhamnolipids scored one hit in our calculations because all these di-rhamnolipids producers belong to the same phylum, *Proteobacteria*.

antimicrobial, anti-inflammatory and anticancer activities (Table 3). Many reviews are found in literature detailing the potential biomedical and cosmeceutical applications of biosurfactants in general, many of which are simple glycolipids [8,158–160].

7. Biosynthesis of simple glycolipids

With few exceptions, the exact biosynthetic steps of majority of simple glycolipids (SGLs) are not yet fully understood. Generally however, biosynthesis of SGLs implicates the supply and linking of glycosyl and lipid precursors. Pathways supplying glycolipid precursors are depicted later (Fig. 5) and are thought to play an important role in regulation of SGLs biosynthesis. Linking of glycosyl and lipid precursors is mostly via *O*-glycosidic or ester bonds (Fig. 1B) that are formed by glycosyltransferases (GT) (Fig. 4 B1, B2) [215] or acyltransferases (AT) [216] (Fig. 4 A1, A2), respectively. Glycosyltransferases catalyze the transfer of the sugar moiety from an activated glycosyl donor, usually sugar-nucleotide (Leloir GTs) or −phosphate (non-Leloir GTs), to a lipid acceptor (or a sugar acceptor for extending the sugar backbone of glycolipids), by making glycosidic bonds between the hydroxyl groups (nucleophile) of the acceptor and the anomeric carbon of the sugar donor (Fig. 4 B1, B2) [215]. Acyltransferases (AT) catalyze the transfer of the lipid moiety from an activated acyl donor, mostly acyl-CoA or -ACP, to a glycosyl acceptor (or a lipid acceptor for extending the lipid backbone of the glycolipid) by making an ester bond between the hydroxyl group (nucleophile) of the acceptor and the acyl donor's carbonyl group [216] (Fig. 4 A1, A2).

Concerning the fate of SGLs, one report showed that the flocculosin GL can be degraded by its producing yeast, *Pseudozyma flocculosa*, which feeds on it under nutrient limitations [153]. Glycolipids could theoretically be hydrolyzed by one or more of the following enzymes. First, glycoside hydrolases (GH) that hydrolyze the sugar-sugar or sugar-lipid glycosidic bonds (Fig. 4 B1, B2) [153,217]. Second, carbohydrate esterases (CE) hydrolyze the sugar-lipid ester bonds (Fig. 4 A2). Lipid esterases (LE), also known as lipases, hydrolyze lipid-lipid ester bonds (Fig. 4 A1) in glycolipids with multimeric hydroxyalkanoate lipid moieties e.g. rhamnolipids (Fig. 3). This hypothesis is corroborated by reports showing the hydrolysis of polymeric hydroxyalkanoates, which share the same lipid moieties as rhamnolipids, by the action of microbial lipases/esterases [218–220]. Nonetheless, the metabolic fate of SGLs is one of the subjects that require thorough investigations.

Among the poorly studied aspects in SGLs metabolism also are the transport SGLs across microbial membranes. Some SGLs require active transport for their exportation out of the cell, like cellobiose

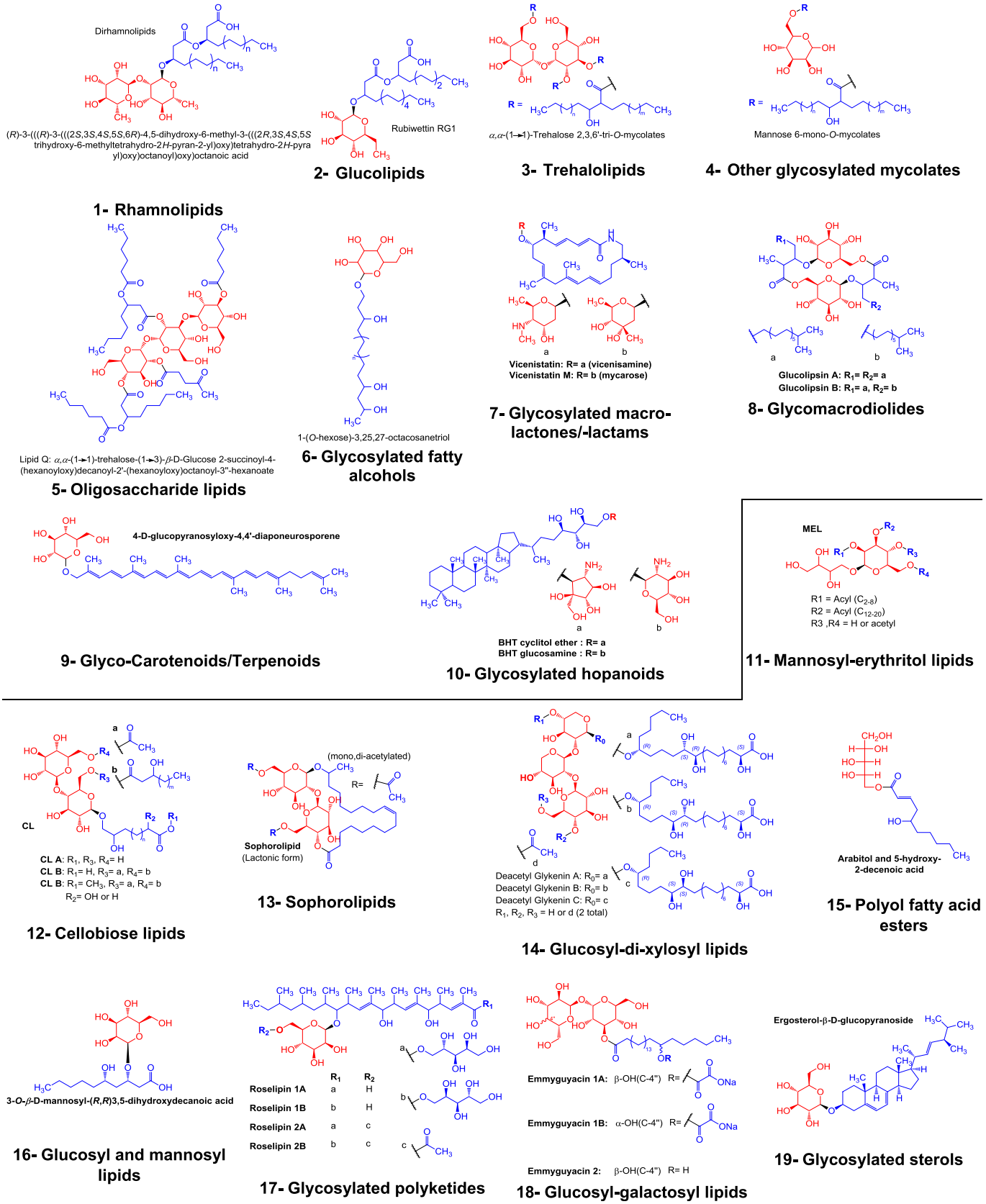

Fig. 3. Structures of prototypic members of bacterial and fungal simple glycolipid (SGL) groups. The glycosyl and lipid residues are colored in red and blue, respectively. Bacterial and fungal SGLs are represented in the upper and lower halves (separated by a line) of the figure, respectively. The representative structure of fungally produced glycosylated paraconic acids (20th group of SGLs) is not given as their structures have been debated [27,28].

Table 1
Chemical groups and members of bacterial simple glycolipids as well as names and phyla of native producers.

Common name: Chemical names (C_x: chain length of fatty acid chains)	Producer	Phylum
Bacteria		
1- Rhamnolipids		
Monorhamnolipids: α-L-rhamnopyranosyl-*R,R*-3-(3′-hydroxyalkanoyloxy) alkanoate ($C_{8\text{-}16}$)	Spp. of *Pseudomonas* and *Burkholderia* [29]	*Proteobacteria*
Dirhamnolipids: α-L-rhamnopyranosyl-(1−2)-α-L-rhamnopyranosyl-*R,R*-3-(3′-hydroxyalkanoyloxy)alkanoate ($C_{8\text{-}16}$)	Spp. of *Pseudomonas* and *Burkholderia* [29]	*Proteobacteria*
2- Glucolipids		
Rubiwettin RG1: β-D-glucopyranosyl 3-(3′-hydroxytetradecanoyloxy)decanoate	*Serratia rubidaea* [30].	*Proteobacteria*
3- Trehalolipids		
α,α-(1-1)-Trehalose 6-mono-*O*-mycolates	*Rhodococcus erythropolis* [31]	*Actinobacteria*
α,α-(1-1)-Trehalose 2,3-di-*O*-mycolates	*Tsukamurella* sp. [32]	*Actinobacteria*
Cord factor: α,α-(1-1)-Trehalose 6,6′-di-*O*-mycolates	Spp. of *Mycobacterium, Rhodococcus, Arthrobacter, Nocardia and Gordonia* [31,33]	*Actinobacteria*
α,α-(1-1)-Trehalose 2,3,6′-tri-*O*-mycolates	*Rhodococcus aurantiacus* [34]	*Actinobacteria*
STL-1, α,α-(1-1)-Trehalose 2,2′-di-*O*-succinoyl-3,4-di-*O*-alkanoates	*Rhodococcus erythropolis* [35]	*Actinobacteria*
STL-2, α,α-(1-1)-Trehalose 2,3,4-mono-*O*-succinoyl-di-*O*-alkanoates	*Rhodococcus erythropolis* [35]	*Actinobacteria*
STL-3, α,α-(1-1)-Trehalose 2,3,4,2′-mono-*O*-succinoyl-tri-*O*-alkanoates	Spp. of *Rhodococcus* [36,37], *Arthrobacter* [38]	*Actinobacteria*
4- Other glycosylated (non-trehalose containing) mycolates		
Sucrose 6-mono-*O*-mycolates	Spp. of *Arthrobacter, Corynebacterium, Nocardia, Brevibacterium* [39,40]	*Actinobacteria*
Fructose 6-mono-*O*-mycolates	Spp. of *Arthrobacter, Corynebacterium, Nocardia, Brevibacterium* [39,40]	*Actinobacteria*
Fructose 1,6-di-*O*-mycolates	Spp. of *Arthrobacter, Corynebacterium, Nocardia, Brevibacterium* [39,40]	*Actinobacteria*
Glucose-6-β-hydroxy-α-hexadecenoyl-eicosenoate	*Brevibacterium thiogenitalis* [41]	*Actinobacteria*
Mannose 6-mono-*O*-mycolates	*Arthrobacter* sp. [42]	*Actinobacteria*
Maltose 6-mono-*O*-mycolates	*Arthrobacter* sp. [42]	*Actinobacteria*
Maltose 6,6′-di-*O*-mycolates	*Arthrobacter* sp. [42]	*Actinobacteria*
Maltotriose 6,6′,6″-tri-*O*-mycolates	*Arthrobacter* sp. [42]	*Actinobacteria*
Cellobiose 6-mono-*O*-mycolates	*Arthrobacter* sp. [42]	*Actinobacteria*
5- Trehalose-containging Oligosaccharide lipids		
Lipid Q: β-D-glucose-(1−3)-α,α-(1-1)-trehalose hexanoyl-succinoyl-3-(hexanoyloxy)octanoate-3-(hexanoyloxy)decanoate	*Rhodococcus* sp. [43,44]	*Actinobacteria*
GL2: β-D-glucose-(1″-2′)-α,α-(1-1)-trehalose 4,6,2″,3″ tetra-*O*-alkanoates ($C_{8\text{-}10}$)	*Tsukamurella* sp. [32]	*Actinobacteria*
GL3: β-D-glucose-(1″-2′)-α,α-(1-1)-trehalose-(6′-1‴)-β-D-galactose-4,6,2″,3″-tetra-*O*-alkanoates ($C_{8\text{-}10}$)	*Tsukamurella* sp. [32]	*Actinobacteria*
β-D-glucose-(1−3)-α,α-(1-1)-trehalose-(6-1)-β-D-glucose-(6-1)-β-D-glucose mono-*O*-succinoyl-hepta-*O*-alkanoate ($C_{2\text{-}8}$)	*Nocardia corynebacteroides* [45–47]	*Actinobacteria*
4,6-(1-Carboxyethylidene)-3-*O*-Me-β-D-glucose-(1−3)-4,6-(1-carboxyethylidene)-β-D-glucose-(1−4)-β-D-glucose-(1−6)-α,α-(1″-1′)-trehalose-4″-*O*-alkanoyl-6′-*O*-alkenoate	*Mycobacterium smegmatis* [48,49]	*Actinobacteria*
6- Glycosylated fatty alcohols		
Alkane 1,2-diol glycoside; Hexose 1-(*O*-hexose)alk-2-yl alkanoate (Diol = $C_{19\text{-}20}$, alkanoate = $C_{14\text{-}16}$)	*Roseiflexus castenholzii* [50]	*Chloroflexi*
1-(*O*-hexose)-3,25-hexacosanediol and its homologue: 1-(*O*-hexose)-3,27-octacosanediol	Spp. of cyanobacteria e.g. *Anabaena, Nodularia, Calothrix, Synechococcus* [51]	*Cyanobacteria*
1-(*O*-hexose)-3-keto-25-hexacosanol and its homologue: 1-(*O*-hexose)-3-keto-27-octacosanol		*Cyanobacteria*
1-(*O*-hexose)-3,25,27-octacosanetriol		*Cyanobacteria*
1-(*O*-hexose)-3-keto-25,27-octacosanediol OR its isomer: 1-(*O*-hexose)-27-keto-3,25-octacosanediol		*Cyanobacteria*
7- Glycosylated macro-lactones/-lactams		
Barsilinolide A/B/C: 2-deoxy-α-L-fucopyranoside of C_{32}-membered macrolactone	*Nocardia brasiliensis* [52,53]	*Actinobacteria*
Fluvirucins: amino sugar glycosides of C_{14}-membered macrolactam	Spp. of *Actinomadura, Streptomyces, Microtetraspora and Saccharotrix mutabilis*	*Actinobacteria*
Vicenistatin: amino sugar (vicenisamine) glycoside of C_{20}-membered macrolactam	*Streptomyces* sp. [54,55]	*Actinobacteria*
Vicenistatin M: D-mycarose glycoside of C_{20}-membered macrolactam	*Streptomyces* sp. [54,55]	*Actinobacteria*
Erythromycins A, B, D, C, E, F and Erythromycin esters (C_{14}-membered macrolactam glycosides)	*Streptomyces erythreus* and *Nocardia* spp, and other *Streptomyces* spp. [56]	*Actinobacteria*
Oleandomycin (C_{14}-membered macrolactam glycosides)	*Streptomyces antibioticus* [57]	*Actinobacteria*
Pikromycin, Narbomycin, 5-*O*-mycaminosyl-narbonolide (C_{14}-membered macrolactam glycosides)	*Streptomyces felleus* and *S. narbonensis* [56]	*Actinobacteria*
10,11-Dihydropikromycin, Kayamicin (C_{14}-membered macrolactam glycosides)	*Streptomyces narbonensis* [56]	*Actinobacteria*
Spinosyns (Tetracyclic macrolide) containing forosamine (amino sugar) and tri-*O*-methyl rhamnose	*Saccharopolyspora spinosa* [56,58]	*Actinobacteria*
Lepicidin A	*Saccharopolyspora spinosa* [56]	*Actinobacteria*
Leucomycins, Josamycin, Platenomycins, Medicamycin, Espinomycins	*Streptomyces kitasatoensis* [56]	*Actinobacteria*
Carbomycin B, platenomycins W1/W2, Niddamycin, Midecamycin A3/A4	*Streptomyces platensis* [56]	*Actinobacteria*
Acumycin (cirramycin B), Cirramycin F and derivatives	*Streptomyces griseoflavus, S. fradiae, S. flocculus* [56]	*Actinobacteria*
Chalcomycin, Neutramycin	*Streptomyces bikiniensis, S. rimosus, S. hirsutus* [56]	*Actinobacteria*
Aldgamycin F, E and Swalpamycin	*Streptomyces lavendulae, S. avidinii, S amandii* (for swalpamycin) [56]	*Actinobacteria*

Table 1 (*continued*)

Common name: Chemical names (C_x: chain length of fatty acid chains)	Producer	Phylum
Spiramicins	*Streptomyces ambofaciens* [56,59]	*Actinobacteria*
Tylosins	*Streptomyces fradie*, *S. hygroscopicus* [56,60]	*Actinobacteria*
Concanamycins	*Streptomyces diastatochromogenes* [56]	*Actinobacteria*
Tetrins and related compounds, Maduralide	*Streptomyces* sp. [56]	*Actinobacteria*
Pimaricin	*Streptomyces natalensis* [56]	*Actinobacteria*
Colubricidin A	*Streptomyces* sp. [56,61]	*Actinobacteria*
Nystatin	*Streptomyces noursei* [62]	*Actinobacteria*
Amphotericin B	*Streptomyces nodosus* [63]	*Actinobacteria*
Oasomycins, Desertomycins	*Streptomyces cinnamoneus* (previsouly *Streptoverticillium baldacci*)	*Actinobacteria*
Rapamycin	*Streptomyces hygroscopicus* [64]	*Actinobacteria*
Avermectins	*Streptomyces avermetilis* [65]	*Actinobacteria*
PM100117 and PM100118	*Streptomyces caniferus*	*Actinobacteria*
8- Glycomacrodiolides (glycosylated macrocyclic dilactones)		
Glucolipsin A, B: dilactone of two glucosides of 3-hydroxy fatty acids C_{19}/C_{19}	*Streptomyces purpurogeniscleroticus, Nocardia vaccinii* [66]	*Actinobacteria*
Fattiviracin A1: dilactone of two glucosides of 3-,17-, ω-1-trihydroxy fatty acids C_{24}/C_{33}	*Kibdelosporangium albatum* [67]	*Actinobacteria*
Cycloviracin B1 and B2: dilactones glucosides of 3-,19-, ω-1-trihydroxy fatty acids (C_{22-28}) and of 3-,17-, ω-1-trihydroxy fatty acids (C_{22-24})	*Streptomyces microflavus* [67]	*Actinobacteria*
Elaiophylins, Efomycin G	*Streptomyces* spp. [68]	*Actinobacteria*
Halichoblelides A, B, C	*Streptomyces* spp. [69–71]	*Actinobacteria*
Bispolides A1, A2, A3, B1, B2a, B2b and B3	*Microbispora species* [72]	*Actinobacteria*
Macroviracins A-D: related to fattiviracin and cycolviracins	*Streptomyces* sp. [73]	*Actinobacteria*
9- Glyco-carotenoids/-terpenoids:		
9.1-Acyclic glycocarotenoids		
Rhodopsin glucoside	*Halorhodospira abdelmalekii*, *H. halochloris* [74]	*Proteobacteria*
Dihydroxylycopene mono-/di-glucosides and their acyl ($C_{12:0}$ or $C_{14:1}$) derivatives	*Halorhodospira abdelmalekii*, *H. halochloris* [74]	*Proteobacteria*
D-Glucosyl 4,4″-diapocarotene-6,6′-dioic acid	*Pseudomonas rhodos* [75], *Rhizobium lupini* [76,77]	*Proteobacteria*
1′-glucosyloxy-3′,4′-didehydro1′,2′-dihydro-ψ,ψ-carotene monoester	*Chondromyces apiculatus* [78], *Myxococcus fulvus* [79]	*Proteobacteria*
Staphyloxanthin: α-D-glucopyranosyl 1-*O*-(4,4′-diaponeurosporen-4-oate) 6-*O*-(12-methyltetradecanoate)	*Staphylococcus* spp. [80]	*Firmicutes*
4-D-glucopyranosyloxy-4,4′-diaponeurosporene	*Streptococcus faecium* [81]	*Firmicutes*
Hydroxy-diaponeurosporene glucoside esters	*Heliorestis* sp. [82]	*Firmicutes*
Rhodopin β-D-glucoside, Rhodopinal β-D-glucoside	*Rhodopseudomonas acidophila, Rhodospirillum tenue and Rhodocyclus purpureus* [83]	*Proteobacteria*
Oscillaxanthin: 1,1′-dihydroxy-2,2′-di-β-L-rhanmosyl- 1,2,1′,2′-tetrahydro-3,4,3′,4′-tetradehydrolycopene	*Oscillatoria rubescens* [84]	*Cyanobacteria*
Bacterioruberin mono- and di-glycosides	*Unidentified Halophilic bacterium* [85]	*Proteobacteria*
Diapolycopenedioic acid xylosyl esters A, B, and C	*Rubritalea squalenifaciens* [86]	*Verrucomicrobia*
Methyl 5-glucosyl-5,6-dihydro-apo-4,4′-lycopenoate	*Planococcus maritimus* [87]	*Firmicutes*
Vancoresmycin	*Amycolatopsis* [88]	*Actinobacteria*
9.2-Monocyclic glycocarotenoids		
Salinixanthin	*Salinibacter ruber* [89], *Rhodothermus marinus* [90]	*Bacteroidetes*
Phleixanthophyll, 4-ketophleixanthophyll	*Mycobacetrium phlei* [91]	*Actinobacteria*
Phleixanthophyll palmitate: (2′-*S*)-1′-[(6-*O*-palmityl-β-D-glucopyranosyl)oxy]-3′,4′-didehydro-1′,2′-dihydro-β,ψ-caroten-2′-ol	*Nocardia* sp. [92]	*Actinobacteria*
1′-[(6-*O*-acyl-β-D-glucopyranosyl)oxy]-1′,2′-dihydro-β,ψ-caroten-4-one	*Rhodococcus rhodochrous* [93,94]	*Actinobacteria*
Myxobactone	*Myxococcus fulvus* [79,95]	*Proteobacteria*
Myxobactin	*Myxococcus fulvus* [96]	*Proteobacteria*
Keto-myxocoxanthin glucoside ester (Myxobactone ester)	*Roseiflexus castenholzii* [97]	*Chloroflexi*
OH-γ-carotene glucoside laurate: 1'-[(6-*O*-lauryl-β-D-glucopyranosyl)oxy]-1′,2′-dihydro-β,ψ-carotene	*Chlorobium tepidum* [98]	*Chlorobi*
OH-chlorobactene glucoside laurate; 1'-[(6-*O*-lauryl-β-D-glucopyranosyl)oxy]-1′,2′-dihydro-ϕ,ψ-carotene	*Chlorobium tepidum* [98]	*Chlorobi*
OH-γ-carotene glucoside ester derivative	*Chloroflexus aurantiacus* [99]	*Chloroflexi*
1′-β-glucopyranosyl-3,4,3′,4′-tetradehydro-1′,2′-dihydro-β,ψ-caroten-2-one	*Meiothermus ruber* [100]	*Deinococcus-Thermus*
Myxoxanthophyll like glycocarotenoid: (3*R*,2′*S*)-myxol-2′-(2,4-di-*O*-methyl-α-L-fucoside)	*Synechocystis* sp. [101]	*Cyanobacteria*
Sioxanthin; (2′*S*)-1′-(β-D-glucopyranosyloxy)-3′,4′didehydro-1′,2′-dihydro-φ,ψ-caroten-2′-ol	*Salinispora* sp. [102]	*Actinobacteria*
9.3-Bicyclic glycocarotenoids		
Corynexanthin monoglycoside	*Corynebacterium* sp. [103]	*Actinobacteria*
Corynexanthin diglycoside	*Arthrobacter* sp [104]	*Actinobacteria*
Sarcixanthin monoglucosides	*Curtobacterium flaccumfaciens* [105], *Micrococcus luteus* [106], *M. yunnanensis* [107].	*Actinobacteria*
Sarcixanthin diglucosides	*Micrococcus luteus* [106], *M. yunnanensis* [107].	*Actinobacteria*
Zeaxanthin mono- and di-glucosides	*Erwinia herbicola, Rhodobacter sphaeroides* [108]	*Proteobacteria*
Zeaxanthin mono- and di-rhamnosides (mainly *Z*-isomers), Zeaxanthin di-glucoside	*Sulfolobus shibatae* [109]	*Archeabacteria*
Zeaxanthin mono- and di-rhamnosides	*Corynebacterium autotrophicum* (*Xanthobacter autotrophicus*) [110]	*Proteobacteria*
Aastaxanthin dirhamnoside	*Sphingomonas astaxanthinifaciens* [111]	*Proteobacteria*
Myxocoxanthin rhamnoside	*Sorangium compositum* [112]	*Proteobacteria*

(*continued on next page*)

Table 1 (*continued*)

Common name: Chemical names (C_x: chain length of fatty acid chains)	Producer	Phylum
Thermozeaxanthin-13, -15, and -17 (Zeaxanthin mono-β-D-glucoside-branched fatty acid esters)	*Thermus thermophilis* [113]	*Deinococcus-Thermus*
Thermobiszeaxanthin-13-13, -13-15, and -15-15 (Zeaxanthin di-β-D-glucoside-branched fatty acid esters)	*Thermus thermophilis* [113]	*Deinococcus-Thermus*
Adonixanthin and astaxanthin glucosides	*Agrobacterium aurantiacum* [114,115]	*Proteobacteria*
Decaprenoxanthin mono- and diglucoside (2*R*,6*S*,2′*R*,6′*S*)-(2,2′-bis(4-hydroxy-3-methyl-2-butenyl)ε,ε-carotene) di-β-D-glucoside	*Corynebacterium glutamicum* [116]	*Actinobacteria*
10- Glycosylated hopanoids/sterols		
Bacteriohopanetetrol cyclitol ether	*Chloracidobacterium thermophilum* [117,118]	*Acidobacteria*
BHT cyclitol	*Burkholderia cenocepacia* [119], *Zymomonas mobilis* [120] [121]	*Proteobacteria*
BHT glucosamine	*Burkholderia cenocepacia* [119], *Zymomonas mobilis* [120,121]	*Proteobacteria*
O-α-D-Glucuronopyranosyl BHT	*Rhodospirillum rubrum* [122]	*Proteobacteria*
Cholesteryl-α-D-glucopyranoside	*Helicobacter pylori* [123]	*Proteobacteria*
Cholesteryl-6-*O*-tetradecanoyl-α-D-glucopyranoside, Cholesteryl-6-*O*-dodecanoyl-α-D-glucopyranoside	*Helicobacter pylori* [123], *H. felis, H. muridarum, H. mustelae, H. fennelliae, and H. cinaedi* [124]	*Proteobacteria*
Cholesteryl-6-*O*-acyl-β-D-galactopyranoside	*Borrelia burgdorferi, B. garinii and B. afzelii* [125]	*Spirochaetes*
Cholesteryl-6-*O*-acyl-β-D-glucopyranoside	*Borrelia hermsii* [125]	*Spirochaetes*
Cholesteryl-β-D-glucopyranoside and its 3,4,6-triacyl derivatives	*Mycoplasma gallinarum* [126]	*Tenericutes*
Cholesteryl-α,α′-di-D-glucoside; α-D-glucopyranosyl-(1 → 3)-(*O*-acyl)-α-D-glucopyranosyl-(1 → 3)-cholesterol	*Acholeplasma axanthum* [127]	*Tenericutes*

lipids, mannosylerythritol lipids [221] and sophorolipids [222], whereas, many other SGLs are thought to passively diffuse out of the cell.

To give a general overview, we present the general biosynthetic map of SGLs showing the diversity of the immediate glycosyl and lipid precursors of SGLs and the pathways furnishing them (Fig. 5).

8. Metabolic engineering of simple glycolipids

Metabolic engineering can be employed to satisfy demand for simple glycolipids (SGLs) by offering solutions to the main challenges facing their production and commercialization. The most important challenge is the high cost of production of SGLs at high purities to qualify for medical or cosmeceutical applications (usually >90–95% purities are required) [21,22]. This high cost stems from a multiplicity of factors including the inherent low yield/productivity of microbial SGLs, costly raw nutritive materials, expensive biosafety containment measures when using pathogenic SGLs producers, expensive/laborious foam control and expensive downstream processing and purification. Futhermore, SGLs are in many cases naturally produced as mixture of homologues/congeners that are difficult to separate; this makes the study and attribution of a specific activity to a specific SGL homologue/congener unattainable. Lastly, there is accumulating evidence that SGLs biosynthesis is tightly regulated in native producers e.g. rhamnolipids production in *Pseudomonas aeruginosa* [226] and sophorolipids production in *Starmerella bombicola* [222]. These tight genetic and metabolic regulations possibly explain the limited improvement in SGL yields using simple optimization media components and process conditions in native SGL producers. One should not be misled, however, by the extraordinarily high GL yields reported in literature that are obtained through media optimization, particularly for rhamnolipids [231]. Such reports are questionable due to different quantification methods used that vary in their specificity and/or sensitivity. Standardized protocols for SGLs quantification were made recently available [232,233] and are expected to profoundly minimize discrepancies in quantification values in glycolipid research.

Although their cost-effectiveness is still unclear, chemical synthesis of GL could overcome many of the problems of SGLs production. Nonetheless, chemical synthesis of SGLs is confronted also by many other limitations and concerns. First, the difficult stereoselective synthesis of glycolipids which are mostly chiral molecules. An attempt to chemically synthesize monorhamnolipid, that is naturally produced as α-L-rhamnopyranosyl-*R*-β-hydroxydecanoyl-*R*-β-hydroxydecanoate, resulted in the inevitable co-production of three other diastereomers with different configurations of the β-hydroxyl groups (*R,R; R,S: S,S; S,R*) and different surface activities [234–236]. Second, certain ecological/health issues are associated with synthetic approaches that most probably involve the use of non-sustainable petrochemical substrates and generate toxic waste products [237]. Thirdly, the biodegradability and toxicity issues of co-produced new-to-nature SGLs diastereomers require attention and investigation.

Genetic engineering and synthetic biology could offer promising ecological solutions to current challenges facing SGLs production, particularly after recent advances in metabolic engineering and tools for cloning and heterologous expression of large biosynthetic pathways. The following sections discuss some of the metabolic engineering strategies for SGLs production.

8.1. Engineering heterotrophic carbon source utilization

Raw nutritive materials accounts for approximately more than 85% of the total estimated production/operation costs of SGLs [21]. A wide range of low-cost renewable raw materials were suggested for SGLs production [238,239], yet, the capacity of GL producers to utilize these raw materials should be investigated or genetically engineered in the selected production host. A successful example of the latter is the engineering of *P. aeruginosa* strain to utilize whey waste for RLs production via heterologous expression of *E. coli lac* genes [240]. Likewise, bacterial and fungal GL producers could be engineered to utilize cheap waste lignocellulosic wastes [241–243]. Although enhancing the utilization of waste oils by expression of lipid esterases seems a good strategy given the low cost and high GL yields [244,245] associated with these oily carbon sources, these carbon sources are, however, cumbersome during recovery of glycolipids as they necessitate extra steps for their removal adding to the net cost of glycolipids recovery [246].

8.2. Heterologous expression of GL biosynthetic pathway

Containment of biosafety level 2 organisms contribute remarkably in the operational costs of simple glycolipids (SGLs)

Table 2
Chemical groups and members of fungal simple glycolipids as well as names and phyla of their native producers.

Common name: Chemical names (C_x: chain length of fatty acid chains)	Producer	Phylum
Fungi		
1- Mannosyl-erythritol lipids (MEL, Ustilipids) and MEL congeners		
MEL	*Ustilago maydis, Pseudozyma (Candida) antarctica* [128,129], *Kurtzmanomyces* [130]	*Basidiomycota*
MEL	*Geotrichum candidum* [131]	*Ascomycota*
Mannosylmannitol lipids (MML), mannosylribitol lipids (MRL) and mannosylarabitol lipids (MAL)	*Pseudozyma parantarctica* [132]	*Basidiomycota*
2- Cellobiose lipids (CL, Ustilagic acids)		
Cellobiose (*β*-D-Glc-(1 → 4)-*β*-D-Glc) 2″-*O*-hexanoic acid 1-*O*-16-*ω*,*ω*-1-dihydroxyhexadecanoate or 1-*O*-16-*ω*,*ω*-1,*α*-trihydroxy hexadecanoate	*Ustilago maydis* [128]	*Basidiomycota*
Cellobiose 6′-*O*-acetyl-2″-*O*-*β*-hydroxyalkanoyl-1-*O*-16-*ω*,*ω*-1-dihydroxyhexadecanoate or 1-*O*-16-ω,ω-1,α-trihydroxy hexadecanoate methyl ester	*Ustilago maydis* [128], *Pseudozyma fusiformata* [133]	*Basidiomycota*
Cellobiose 16-*O*-*ω*,*ω*-1-dihydroxyhexadecanoate or 1-*O*-16-*ω*,*ω*-1,*α*-trihydroxy hexadecanoate	*Sympodiomycopsis paphiopedili* [134]	*Basidiomycota*
Microcin: cellobiose 2″,3″,4″,6″,6′-penta-*O*-acetyl-1-*O*-16-ω,α-dihydroxyhexadecanoate	*Cryptococcus humicola* [135]	*Basidiomycota*
Flocculosin: 2-(2′,4′-diacetoxy-5′-carboxy-pentanoyl) octadecyl cellobioside	*Anthracocystis (Pseudozyma)* flocculosa [136]	*Basidiomycota*
3- Sophorolipids		
Sophorose (*β*-D-Glc-(1 → 2)-*β*-D-Glc) -1-*O*-16-*ω*,*ω*-1-dihydroxyalkanoate or 1-*O*-16-*ω*,*ω*-1,*α*-trihydroxy alkanoate (C_{16}-$C_{18:0\text{-}2}$)	*Starmerella (Candida) bombicola, Candida apicola and other spp.* [137]	*Ascomycota*
Sophorose (*β*-D-Glc-(1 → 2)-*β*-D-Glc) -1-*O*-16-*ω*,*ω*-1-dihydroxyalkanoate or 1-*O*-16-*ω*,*ω*-1,*α*-trihydroxy alkanoate (C_{16}-$C_{18:0\text{-}2}$)	*Cryptococcus curvatus* [137]	*Basidiomycota*
Sophorose 6′-mono-*O*-acetyl or 6′,6″-di-*O*-acetyl -1-*O*-16-*ω*,*ω*-1-dihydroxyalkanoate or 1-*O*-16-*ω*,*ω*-1,*α*-trihydroxy alkanoate (C_{16}-$C_{20:1}$)	*Starmerella (Candida) bombicola, Candida apicola and other spp.* [137], Wickerhamiella domercqiae [138,139]	*Ascomycota*
Sophorose lipid lactonic/ring form, lactonization of free carboxyl group with C-4″ or C-6" (intramolecular ester bonds)	*Starmerella (Candida) bombicola, Candida apicola and other spp.* [137]	*Ascomycota*
Dimeric and trimeric sophorolipids (intermolecular ester bonds between carboxyl of one molecule to C-4″ of another molecule)	*Candida* spp. [140]	*Ascomycota*
4- Glucosyl-di-xylosyl lipids (Glykenins)		
Glykenins A, B, C: *O*-*β*-D-glucose-(1 → 2)-*O*-*β*-D-xylose-(1 → 2)-*O*-*β*-D-xylose tetrahydroxyhexacosanoic acids, mono-di or tri-acetylated	Basidiomycetous sp. [141]	*Basidiomycota*
5- Polyol fatty acid esters (Liamocins and their congeners)		
Liamocins	*Aureobasidium pullulans*	*Ascomycota*
Mannitol and pentitol esters of 3-D-hydroxypalmitic and 3-D-hydroxystearic acids	*Rhodotorula glutinis and Rhodotorula graminis*	*Basidiomycota*
6- Glucosyl and mannosyl lipids		
Monoglucosyloxyoctadecenoic acid	*Aspergillus niger* [142]	*Ascomycota*
Halymecin B: mannosylated tetramer of 3,5-dihydroxydecanoic acid	*Fusarium* sp. [143]	*Ascomycota*
Halymecins F: acetylated halemycin B, halymecin G: mannosylated trimer of 3,5-dihydroxydecanoic acid	*Simplicillium lamellicola* [144]	*Ascomycota*
(3*R*,5*R*)-3-*O*-*β*-D-mannosyl-3,5-dihydroxydecanoic acid	*Simplicillium lamellicola* [144]	*Ascomycota*
7- Glycosylated polyketides		
Roselipin 1, 2: 2,4,6,8,10,12,14,16,18-nonamethyl-5,9,13-trihydroxy-2*E*,6*E*,10*E*-icosenoic acid mannosylated (+-acetylated) at C-13, D-arabitol ester [145,146]	*Gliocladium* [145,146]	*Ascomycota*
TMC-151 A ~ F: 2,4,6,8,10,12,14,16,18-nonamethyl-5,9,13-trihydroxy-2*E*,6*E*,10*E*-icosenoic acid mannosylated (+-acetylated) at C-13, D-mannitol ester [145,146]	*Gliocladium catenulatum* [147]	*Ascomycota*
TMC-154: isolmeric form of roselipin 1 and TMC-171 A ~ C: as roselipin 3 but esterified to mannitol	*Gliocladium* [148]	*Ascomycota*
Roselipins 3A to 3E: 14,15-dehydro derivatives of roselipin 1A/B	*Clonostachys candelabrum* [149]	*Ascomycota*
Cladionol A: 15-mannosyl-2,4,6,8,10,12,14,16,18,20-decamethyl-3,7,11,15-tetrahydroxy-4*E*,8*E*,12*E*-docosenoic acid arabitol ester	*Gliocladium* [150]	*Ascomycota*
8- Glucosyl-galactosyl lipids		
Emmyguyacin 1A: *α*-D-glucopyranosyl-*α*-D-galactopyranose 3′-*O*-hydroxydocosanoate with 17-((carboxycarbonyl)oxy) group of oxalate ester at OH of C-17	*Fungal species* [151]	NA
Emmyguyacin 1B: Trehalose 3′-*O*-docosanoate with 17-((carboxycarbonyl)oxy) group of oxalate ester at OH of C-17	*Fungal species* [151]	NA
Emmyguyacin 2: as emmyguyacin 1A without the oxalate ester	*Fungal species* [151]	NA
9- Glycosylated sterols		
Ergosterol-*β*-D-glucopyranoside	*Pichia pastoris, Sordaria macrospora, Rhynchosporium secalis* [152]	*Ascomycota*
10- Glycosylated paraconic acids		
Gobienines A/B/C (non-confirmed structure [28])	*Acarospora gobiensis* (Lichen) [27]	*Ascomycota*

production. Moreover, working with pathogenic or opportunistic pathogens presents a health risk to manufacturing personnel as well as to public and environment. Heterologous expression of GL biosynthetic genes in hosts that are Generally Recognized As Safe (GRAS) is, therefore, a promising solution as it would require a less costly biosafety level 1 manufacturing facility.

Heterologous expression of rhamnolipids (RLs) in non-pathogenic hosts has received much attention because of the large commercial potential of RLs and because the main and best RLs producer is the opportunistic pathogenic bacterium

Table 3
Biological activities of different chemical groups of microbial simple glycolipids.

	Chemical group of simple glycolipids (SGLs)	A Antibacterial	B Antifungal	C Antiviral	D Antiparasitic	E Anticancer	F ↑ Cell differentiation[a]	G Immunomodulatory	H Antioxidant	I Antiadherent (Biofilm, wounds)	J Neuronal activity	K Sperm immobilizing activity	L ↓ Diacylglycerol acyl transferase 2[a]
	Bacterial SGLs												
1	Rhamnolipids	A1	B1	C1	D1			G1		I1			
2	Glycolipids (Rubiwettin)												
3	Trehalolipids			C3		E3	F3	G3					
4	Other glycosylated mycolates												
5	Oligosaccharide lipids					E5							
6	Glycosylated fatty alcohols												
7	Glycosylated macro-lactones/-lactams	A7	B7	C7	D7	E7		G7					
8	Glycomacrodiolides	A8	B8	C8	D8	E8		G8					
9	Glyco-carotenoids/-terpenoids							G9	H9				
10	Glycosylated hopanoids												
	Fungal SGLs												
11	Mannosyl-erythritol lipids	A11				E11	F11		H11		J11		
12	Cellobiose lipids	A12	B12										
13	Sophorolipids			C13			F13	G13		I13		K13	
14	Glucosyl-di-xylosyl lipids (Glykenins)	A14											
15	Polyol fatty acid esters	A15				E15							
16	Glucosyl and mannosyl lipids	A16				E16							
17	Glycosylated polyketides			C17	D17								L17
18	Glucosyl-galactosyl lipids			C18									
19	Glycosylated sterols												
20	Glycosylated paraconic acids												

References:

A1: [23]
A7: [56]
A8: [72,176,177]
A11: [182]
A12: [188–190]
A14: [194]
A15: [198–200]
A16: [144]
B1: [23,158,207–212].
B8: [161,162]
B12: [135,166–168]
C1: [178]
C3: [183]
C7: [191]
C8: [56,73]
C13: [197]
C17: [203]
C18: [151]
D7: [58,61,65]
D8: [169,170]
D17: [149]
E3: [47,184]
E5: [32,47]
E7: [195]
E8: [70,71,176]
E11: [204,205]
E15: [198]
F3: [36,163,164]
F11: [171–175]
F13: [171]
G1: [185].
G3: [192,193]
G7: [53,64,196]
G8: [201]
G9: [206]
G13: [213,214]
H9: [165]
H11: [129]
I1: [179–181]
I13: [186,187].
J11: [131]
K13: [197]
L17: [202]

[a] The signs ↑ and ↓ denotes for stimulation and inhibition, respectively.

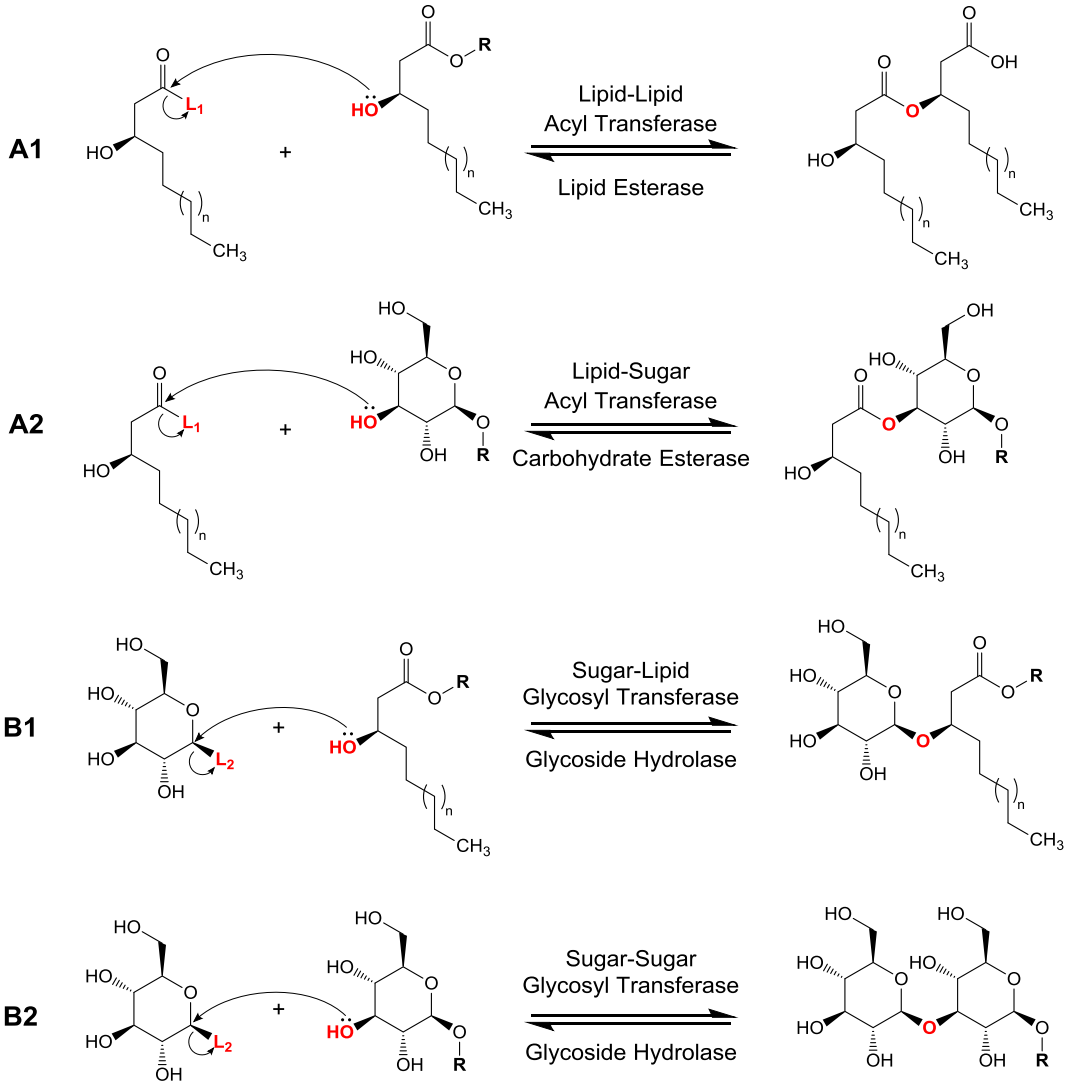

Fig. 4. Key enzymes of glycolipid biosynthesis and hydrolysis. Last steps of glycolipid biosynthesis involves linking of sugar and lipid moieties via either or both Acyl Transferases (AT) (**A1** and **A2**, forward reactions) and Glycosyl Transferases (GT) (**B1** and **B2**, forward reactions) which catalyze the ester and glycosidic bonds formation, respectively. Glycolipids are catabolized or broken down by Lipid Esterase (LE), Carbohydrate Esterases (CE) and Glycoside Hydrolases (GH) that hydrolyze the bond between alkyl-alkanoate ester, acyl-sugar ester and glycosidic bonds, respectively (reverse reactions). **L1**: Coenzyme A (CoA-S-) or Acyl Carrier Protein (ACP-S-) activating groups on acyl donors; **L2**: Nucleotides or phosphates activating groups on glycosyl donors. **R**: any substitution that could be glycosyl, lipid, or glycolipid units. Notes: *β*-glucose and *R*-3-hydroxyalkanoate are used as examples of any sugar and hydroxyl fatty acid of any chain length (*n*), respectively. Hydrolysis reactions do not generate activated products.

Pseudomonas aeruginosa [247]. One example is the successful expression of rhamnolipids biosynthetic genes of *P. aeruginosa* in non-pathogenic bacteria, namely *P. putida* [246] and *P. fluorescens* [248] as well as in *E. coli* [249,250], the best of which was recombinant *P. putida* [246], though, all recombinant strains produced RLs at much lower yields than the native producer. Interestingly, the non-pathogenic strain *P. chlororaphis* is naturally producing mono-rhamnolipids and not di-rhamnolipids as it lacks the gene coding for the second rhamnosyltransferase, *rhlC* [251]. Heterologous expression of *rhlC* from *P. aeruginosa* in *P. chlororaphis* resulted in production of di-RL at concentration more than twice that of mono-RL [251]. *Burkholderia kururiensis* is another nonpathogenic heterologous host that was successfully engineered for RLs production [252]. Further engineering strategies were reviewed for over-production of RL [253].

Several glycosylated carotenoids (C_{40} and C_{50}), e.g. glucosides of decaprenoxanthin and sarcinaxanthin as well as zeaxanthin, were successfully engineered in *Corynebacterium glutamicum* by overexpression of genes coding for lycopene cyclization, hydroxylation and glycosylation [254].

Interestingly, 17 genes were heterologously expressed in an *E. coli* strain that is already producing 6-deoxyerythronolide B precursor to produce the glycosylated macrolide, erythromycin C [255]. The cloned genes encoded the deoxysugar, desosamine, biosynthetic enzymes and the enzymes converting 6-deoxyerythronolide B to erythromycin C [255,256].

Selection criteria for candidate hosts for heterologous glycolipid production should include, in addition to being non-pathogenic, to be natively tolerant to high concentrations of the target SGLs if high productivities are sought [246]. This is particularly important for SGLs which mostly demonstrate antimicrobial activities.

Moreover, the candidate host should, preferably, abundantly produce the precursors required for SGLs biosynthesis. A good approach would be starting with analysis of the intracellular concentration of lipid and glycosyl precursors. One example is the evaluation of the *R*-specific enoyl-CoA hydratase-2 (ECH-2) activity

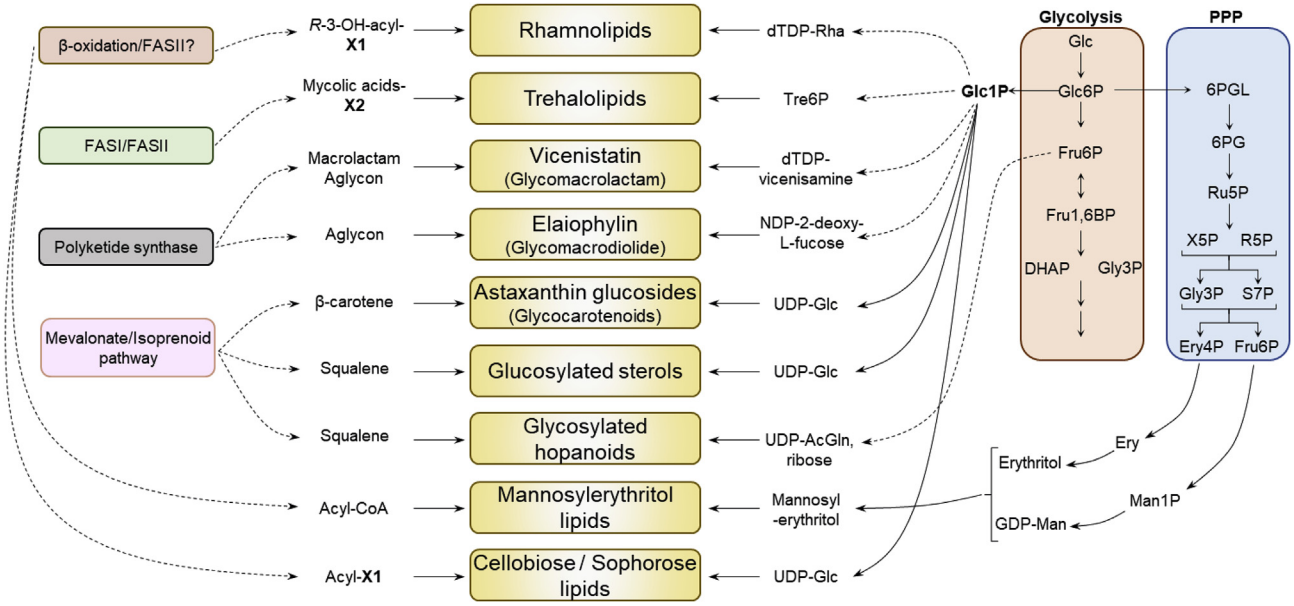

Fig. 5. Sugar and lipid precursors of prominent members of simple glycolipid groups and their furnishing pathways. Biosynthesis of simple glycolipids that harbor glucoside units, like glucosides of astaxanthin [115] and zeaxanthin [223] as well as glucosylated sterols [224], cellobiose and sophorose lipids [225], require UDP-glucose as glycosyl donor. Peculiar glycosyl donors that are activated in other ways than UDP are required in case of rhamnolipids [226], trehalolipids [227], vicenistatin [228] and elaiophylin [68]. All glycosyl donors are derived from glucose -1-phosphate except that of glycosylated hopanoids whose glycosyl donors is derived from β-D-fructofuranose-6-phosphate and ribose [119]. Mannosylerythritol lipids are expected to derive the glycosyl unit, mannosylerythritol, from erythritol and GDP-mannose which originate from D-erythrose-4-phosphate and β-D-fructofuranose-6-phosphate intermediates of the pentose phosphate pathway as described in the yeast *Yarrowia lipolytica* [229,230]. The lipid moiety originates mostly from fatty acid synthesis and/or β-oxidation except glycosylated macrolides, carotenoids and sterols/hopanoids whose lipid moiety is furnished from the polyketide for the former and from mevalonate/isoprenoid pathway for the latter two groups. **6PGL:** 6-phosphogluconolactone; **6PG**: 6-phosphogluconic acid; **Ru5P**: D-Ribulose-5-phosphate; **R5P**: D-Ribose-5-phosphate; **X5P**: D-Xylulose-5-phosphate; **S7P**: D-Sedoheptulose-7-phosphate; **Gly3P**: D-Glyceraldehyde-3-phosphate; **DHAP**: Dihydroxyacetone phosphate; **Ery4P**: D-Erythrose-4-phosphate; **Fru6P**: β-D-Fructofuranose-6-phosphate; **Fru1,6BP**: β-D-Fructofuranose-1,6-bisphosphate; **dTDP-Rha:** dTDP-rhamnose; **Tre6P**: Trehalose-6-phosphate; **Glc**: D-Glucopyranose; **Glc1P**: Glucose-1-phosphate; **Glc6P**: Glucose-6-phosphate; **UDP-glc**: Uridine diphosphate glucose; **UDP-AcGln**: Uridine diphosphate *N*-acetylglucosamine; **Man1P**: Mannose-1-phosphate; **GDP-Man**: GDP-mannose; ***R*-3-OH-acyl-X1**: R-3-hydroxy acyl-X1 (X1 = -CoA/-ACP); **X2** = Mycolyl Carrier Protein. **PPP**: Pentose Phosphate Pathway. Dashed lines means that multiple biosynthetic steps are involved.

in crude cell lysate of target host organism as this predicts the potential of this host to synthesize *R*-3-hydroxyalkanoate precursors [257] that form the lipid part of many *R*-3-hydroxyfatty acid-containing glycolipids, e.g. rhamnolipids and rubiwettins. The ECH-2 activity was recently reported to be significantly implicated in rhamnolipids biosynthesis [258].

Oleaginous yeasts, like *Yarrowia lipolytica* and *Rhodosporidium toruloides*, are potential candidates in view of their already high lipid flux [259,260]; therefore, they are supposed to have abundant lipid precursors for GLs biosynthesis.

8.3. Blocking competing pathways

Blocking competitive pathways is an important strategy that is expected to enhance GLs biosynthesis. One example is blocking polyhydroxyalkanoates (PHA) synthesis in *P. aeruginosa* that changed the distribution of produced rhamnolipids (RLs) congeners by doubling the amount of produced mono-rhamnolipids relative to di-rhamnolipids [258]. Also, PHA mutant of *P. putida* was used to enhance heterologous production of RLs [246].

8.4. Tailoring the GL pool composition

Most simple glycolipids (SGLs) are naturally produced in mixtures of congeners and homologues like rhamnolipids [29] and sophorolipids [222]. For studying the functions and properties of each GL species in these natural mixtures, engineered production of purified GL should be sought. This can be achieved by selectively knocking out the genes coding for biosynthesis of specific congeners or forms. A recent example is the production of sophorolipids pool enriched to 88% in the acidic non-lactonized free form by using a mutant strain of *Starmerella bombicola* [261] that is defective in lactone esterase [262].

9. Summary and perspectives

More than 5 decades of glycolipids research has led to the discovery of a huge number of simple microbial glycolipids, around 140 of which are cited in this review. In spite of the many publications demonstrating their great biomedical potential, the majority of discovered simple glycolipids are still unable to translate into commercial products because of their high cost of production mainly stemming from low biological yields. Metabolic engineering has the potential to overcome this cost problem particularly after the revolutionary developments in genetic engineering and synthetic biology techniques that were witnessed in the last 5 years.

This review is an attempt to structure the literature available on simple glycolipids aiming at providing metabolic engineers with an outlook on glycolipids and their biosynthesis. It highlights some of aspects and details that are still missing in the biosynthesis, transport and catabolism of glycolipids that need to be pursued and applied profitably in engineering cost-effective microbial glycolipid producers.

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References

[1] Schörken U, Kempers P. Lipid biotechnology: industrially relevant production processes. Eur J Lipid Sci Technol 2009;111(7):627–45.
[2] Herrera-González A, et al. Functionalization of natural compounds by enzymatic fructosylation. Appl Microbiol Biotechnol 2017;101(13):5223–34.
[3] Gantt RW, Peltier-Pain P, Thorson JS. Enzymatic methods for glyco(-diversification/randomization) of drugs and small molecules. Nat Product Rep 2011;28(11):1811–53.
[4] Zhang B, et al. De novo synthesis of trans-10, cis-12 conjugated linoleic acid in oleaginous yeast Yarrowia Lipolytica. Microb Cell Factories 2012;11(1):51.
[5] Beopoulos A, et al. Metabolic engineering for ricinoleic acid production in the oleaginous yeast Yarrowia lipolytica. Appl Microbiol Biotechnol 2014;98(1):251–62.
[6] Sabirova JS, et al. The 'LipoYeasts' project: using the oleaginous yeast Yarrowia lipolytica in combination with specific bacterial genes for the bioconversion of lipids, fats and oils into high-value products. Microb Biotechnol 2011;4(1):47–54.
[7] Varvaresou A, Iakovou K. Biosurfactants in cosmetics and biopharmaceuticals. Lett Appl Microbiol 2015;61(3):214–23.
[8] Lukic M, Pantelic I, Savic S. An overview of novel surfactants for formulation of cosmetics with certain emphasis on acidic active substances. Tenside Surfactants Deterg 2016;53(1):7–19.
[9] Holst O. Glycolipids: occurrence, significance, and properties. In: Fraser-Reid BO, Tatsuta K, Thiem J, editors. Glycoscience: chemistry and chemical biology. Berlin, Heidelberg: Springer Berlin Heidelberg; 2008. p. 1603–27.
[10] Chester MA. IUPAC-IUB joint commission on biochemical nomenclature (JCBN) nomenclature of glycolipids. J Mol Biol 1999;286(3):963–70.
[11] Leray C. Complex lipids. In: Introduction to lipidomics. CRC Press; 2012. p. 211–2.
[12] Leray C. Simple lipids with two different components. In: Introduction to lipidomics. CRC Press; 2012. p. 169–210.
[13] Leray C. Complex glycolipids. In: Introduction to lipidomics. CRC Press; 2012. p. 247–94.
[14] Merrill Jr AH, Vu MN. Glycolipids. In: Encyclopedia of cell biology. Waltham: Academic Press; 2016. p. 180–93.
[15] Hill K, Rhode O. Sugar-based surfactants for consumer products and technical applications. Lipid/Fett 1999;101(1):25–33.
[16] Poremba K, et al. Marine Biosurfactants, III. Toxicity testing with marine microorganisms and comparison with synthetic surfactants. Z für Naturforsch C 1991:210.
[17] Madsen JK, Kaspersen JD, Andersen CB, Nedergaard Pedersen J, Andersen KK, Pedersen JS, Otzen DE. Glycolipid biosurfactants activate, dimerize and stabilize *T. lanuginosus* lipase in a pH-dependent fashion. Biochemistry 2017;56:4256–68. https://doi.org/10.1021/acs.biochem.7b00420.
[18] Otzen DE. Biosurfactants and surfactants interacting with membranes and proteins: same but different? Biochim Biophys Acta (BBA) Biomembr 2017;1859(4):639–49.
[19] Madsen JK, et al. The anionic biosurfactant rhamnolipid does not denature industrial enzymes. Front Microbiol 2015;6.
[20] Sekhon KK, Rahman PKSM. Rhamnolipid biosurfactants – past, Present and future scenario of global market. Front Microbiol 2014;5.
[21] Ashby RD, et al. A process model for approximating the production costs of the fermentative synthesis of sophorolipids. J Surfactants Deterg 2013;16(5):683–91.
[22] Dhanarajan G, Sen R. Cost analysis of biosurfactant production from a scientist's perspective. In: Biosurfactants. CRC Press; 2014. p. 153–62.
[23] Lang S, Wullbrandt D. Rhamnose lipids – biosynthesis, microbial production and application potential. Appl Microbiol Biotechnol 1999;51(1):22–32.
[24] Singh A, Van Hamme JD, Ward OP. Surfactants in microbiology and biotechnology: Part 2. Application aspects. Biotechnol Adv 2007;25(1):99–121.
[25] Kitamoto D, Isoda H, Nakahara T. Functions and potential applications of glycolipid biosurfactants —from energy-saving materials to gene delivery carriers. J Biosci Bioeng 2002;94(3):187–201.
[26] Elshahawi SI, et al. A comprehensive review of glycosylated bacterial natural products. Chem Soc Rev 2015;44(21):7591–697.
[27] Řezanka T, Guschina IA. Macrolactone glycosides of three lichen acids from Acarospora gobiensis, a lichen of Central Asia. Phytochemistry 2001;58(8):1281–7.
[28] Kondoh A, et al. Total synthesis of nominal gobienine A. Chem A Eur J 2013;19(24):7731–8.
[29] Abdel-Mawgoud AM, Lépine F, Déziel E. Rhamnolipids: diversity of structures, microbial origins and roles. Appl Microbiol Biotechnol 2010;86(5):1323–36.
[30] Matsuyama T, et al. Surface-active novel glycolipid and linked 3-hydroxy fatty acids produced by Serratia rubidaea. J Bacteriol 1990;172(6):3015–22.
[31] Kretschmer A, Wagner F. Characterization of biosynthetic intermediates of trehalose dicorynomycolates from Rhodococcus erythropolis grown on n-alkanes. Biochim Biophys Acta (BBA) Lipids Lipid Metab 1983;753(3):306–13.
[32] Vollbrecht E, et al. Production and structure elucidation of di- and oligosaccharide lipids (biosurfactants) from Tsukamurella sp. nov. Appl Microbiol Biotechnol 1998;50(5):530–7.
[33] Rapp P, et al. Formation, isolation and characterization of trehalose dimycolates from Rhodococcus erythropolis grown on n-alkanes. Microbiology 1979;115(2):491–503.
[34] Tomiyasu I, et al. Occurrence of a novel glycolipid, 'trehalose 2,3,6'-trimycolate' in a psychrophilic, acid-fast bacterium, Rhodococcus aurantiacus (Gordona aurantiaca). FEBS Lett 1986;203(2):239–42.
[35] Uchida Y, et al. Extracellular accumulation of mono- and di-succinoyl trehalose lipids by a strain of Rhodococcus erythropolis grown on n-alkanes. Agric Biol Chem 1989;53(3):757–63.
[36] Sudo T, et al. Induction of the differentiation of human HL-60 promyelocytic leukemia cell line by succinoyl trehalose lipids. Cytotechnology 2000;33(1):259–64.
[37] Espuny MJ, et al. Nutritional requirements of a biosurfactant producing strain Rhodococcus sp 51T7. Biotechnol Lett 1996;18(5):521–6.
[38] Passeri A, et al. Marine Biosurfactants, II. Production and characterization of an anionic trehalose tetraester from the marine bacterium Arthrobacter sp. EK 1. Z für Naturforsch C 1991:204.
[39] Suzuki T, Tanaka H, Itoh S. Sucrose lipids of arthrobacteria, corynebacteria and nocardia grown on sucrose. Agric Biol Chem 1974;38(3):557–63.
[40] Itoh S, Suzuki T. Fructose-lipids of arthrobacter, corynebacteria, nocardia and mycobacteria grown on fructose. Agric Biol Chem 1974;38(8):1443–9.
[41] Okazaki H, et al. L-Glutamic acid fermentation; Part VI. Structure of a sugar lipid produced by Brevibacterium thiogenitalis. Agric Biol Chem 1969;33(5):764–70.
[42] Li Z-Y, et al. Formation and identification of interfacial-active glycolipids from resting microbial cells. Appl Environ Microbiol 1984;48(3):610–7.
[43] Esch SW, et al. A novel trisaccharide glycolipid biosurfactant containing trehalose bears ester-linked hexanoate, succinate, and acyloxyacyl moieties: NMR and MS characterization of the underivatized structure. Carbohydr Res 1999;319(1):112–23.
[44] Konishi M, et al. *Deep-sea Rhodococcus sp.* BS-15, lacking the phytopathogenic fas genes, produces a novel glucotriose lipid biosurfactant. Mar Biotechnol 2014;16(4):484–93.
[45] Kim J-S, et al. Microbial glycolipid production under nitrogen limitation and resting cell conditions. J Biotechnol 1990;13(4):257–66.
[46] Powalla M, Lang S, Wray V. Penta- and disaccharide lipid formation by Nocardia corynebacteroides grown on n-alkanes. Appl Microbiol Biotechnol 1989;31(5):473–9.
[47] Palme O, et al. Selected microbial glycolipids: production, modification and characterization. In: Sen R, editor. Biosurfactants. New York, NY: Springer New York; 2010. p. 185–202.
[48] Kamisango K, et al. Pyruvylated glycolipids from Mycobacterium smegmatis. Nature and location of the lipid components. J Biol Chem 1985;260(7):4117–21.
[49] Saadat S, Ballou CE. Pyruvylated glycolipids from Mycobacterium smegmatis. Structures of two oligosaccharide components. J Biol Chem 1983;258(3):1813–8.
[50] van ver Meer MT, et al. Alkane-1,2-diol-based glycosides and fatty glycosides and wax esters in *Roseiflexus castenholzii* and hot spring microbial mats. Arch Microbiol 2002;178(3):229–37.
[51] Bauersachs T, et al. Distribution of heterocyst glycolipids in cyanobacteria. Phytochemistry 2009;70(17):2034–9.
[52] Mikami Y, et al. A new antifungal macrolide component, brasilinolide B, produced by *Nocardia brasiliensis*. J Antibiot (Tokyo) 2000;53(1):70–4.
[53] Komatsu K, et al. Absolute stereochemistry of immunosuppressive macrolide brasilinolide A and its new congener brasilinolide C. J Org Chem 2004;69(5):1535–41.
[54] Shindo K, et al. Vicenistatin, a novel 20-membered macrocyclic lactam antitumor antibiotic. J Antibiot (Tokyo) 1993;46(7):1076–81.
[55] Matsushima Y, et al. Isolation and structure elucidation of vicenistatin M, and importance of the vicenisamine aminosugar for exerting cytotoxicity of vicenistatin. J Antibiot (Tokyo) 2001;54(3):211–9.
[56] Dembitsky VM. Astonishing diversity of natural surfactants: 2. Polyether glycosidic ionophores and macrocyclic glycosides. Lipids 2005;40(3):219–48.
[57] Vilches C, et al. Biosynthesis of oleandomycin by Streptomyces antibioticus: influence of nutritional conditions and development of resistance. Microbiology 1990;136(8):1447–54.
[58] Mertz FP, Yao RC. Saccharopolyspora spinosa sp. nov. isolated from soil collected in a sugar mill rum still. Int J Syst Evol Microbiol 1990;40(1):34–9.
[59] Hara O, Hutchinson CR. A macrolide 3-O-acyltransferase gene from the midecamycin-producing species *Streptomyces* mycarofaciens. J Bacteriol 1992;174(15):5141–4.
[60] Baltz RH, et al. Biosynthesis of the macrolide antibiotic tylosin. A preferred pathway from tylactone to tylosin. J Antibiot (Tokyo) 1983;36(2):131–41.
[61] Kong F, et al. Colubricidin A, a novel macrolide antibiotic from a *Streptomyces* sp. Tetrahedron Lett 1999;40(52):9219–23.

[62] Brautaset T, et al. Biosynthesis of the polyene antifungal antibiotic nystatin in Streptomyces noursei ATCC 11455: analysis of the gene cluster and deduction of the biosynthetic pathway. Chem Biol 2000;7(6):395–403.
[63] Gallis HA, Drew RH, Pickard WW. Amphotericin B: 30 years of clinical experience. Rev Infect Dis 1990;12(2):308–29.
[64] Park SR, et al. Biosynthesis of rapamycin and its regulation: past achievements and recent progress. J Antibiot 2010;63(8):434–41.
[65] Ikeda H, Ōmura S. Avermectin biosynthesis. Chem Rev 1997;97(7):2591–610.
[66] Qian-Cutrone J, et al. Glucolipsin A and B, two new glucokinase activators produced by *Streptomyces purpurogeniscleroticus* and *Nocardia vaccinii*. J Antibiot (Tokyo) 1999;52(3):245–55.
[67] Yokomizo K, et al. Fattiviracin A1, a novel antiviral agent produced by *Streptomyces microflavus* strain No. 2445. II. Biological properties. J Antibiot (Tokyo) 1998;51(11):1035–9.
[68] Haydock SF, et al. The putative elaiophylin biosynthetic gene cluster in Streptomyces sp. DSM4137 is adjacent to genes encoding adenosylcobalamin-dependent methylmalonyl CoA mutase and to genes for synthesis of cobalamin. J Biotechnol 2004;113(1):55–68.
[69] Yamada T, Minoura K, Numata A. Halichoblelide, a potent cytotoxic macrolide from a Streptomyces species separated from a marine fish. Tetrahedron Lett 2002;43(9):1721–4.
[70] Yamada T, et al. Halichoblelides B and C, potent cytotoxic macrolides from a Streptomyces species separated from a marine fish. Tetrahedron Lett 2012;53(23):2842–6.
[71] Han Y, et al. Halichoblelide D, a new elaiophylin derivative with potent cytotoxic activity from Mangrove-derived *Streptomyces* sp. 219807. Molecules 2016;21(8):970.
[72] Okujo N, et al. Bispolides, novel 20-membered ring macrodiolide antibiotics from Microbispora. J Antibiot 2007;60(3):216–9.
[73] Takahashi S, et al. Determination of absolute structure of macroviracins by chemical synthesis. Org Lett 2003;5(9):1555–8.
[74] Takaichi S, et al. Dihydroxylycopene diglucoside diesters: a novel class of carotenoids from the phototrophic purple sulfur bacteria Halorhodospira abdelmalekii and Halorhodospira halochloris. Arch Microbiol 2001;175(3):161–7.
[75] Kleinig H, et al. New C30-carotenoic acid glucosyl esters from *Pseudomonas rhodos*. Z für Naturforsch C 1979:181.
[76] Kleinig H, et al. Carotenoids of rhizobia. I. *New Carotenoids from Rhizobium lupini*. Helvetica Chim Acta 1977;60(1):254–8.
[77] Kleinig H, Broughton WJ. Carotenoid pigments in a red strain of Rhizobium from Lotononis bainesii Baker. Arch Microbiol 1982;133(2). 164–164.
[78] Kleinig H, Reichenbach H. A new carotenoid glucoside ester from *Chondromyces apiculatus*. Phytochemistry 1973;12(10):2483–5.
[79] Kleinig H, Reichenbach H. Biosynthesis of carotenoid glucoside esters in Myxococcus fulvus (Myxobacterales): inhibition by nicotine and carotenoid turnover. Biochim Biophys Acta (BBA) Lipids Lipid Metab 1973;306(2):249–56.
[80] Marshall JH, Wilmoth GJ. Pigments of Staphylococcus aureus, a series of triterpenoid carotenoids. J Bacteriol 1981;147(3):900–13.
[81] Taylor RF, Davies BH. Triterpenoid carotenoids and related lipids. Triterpenoid monohydroxy- and monoglucosyloxy-carotenoids from Streptococcus faecium UNH 564P. Biochem J 1974;139(3):761–9.
[82] Takaichi S, et al. Novel carotenoid glucoside esters from alkaliphilic heliobacteria. Arch Microbiol 2003;179(2):95–100.
[83] Schmidt K. Carotenoids of purple nonsulfur bacteria. Arch für Mikrobiol 1971;77(3):231–8.
[84] Hertzberg S, Liaaen-Jensen S. The structure of oscillaxanthin. Phytochemistry 1969;8(7):1281–92.
[85] Arpin N, Fiasson JL, Liaaen-Jensen A. Bacterial carotenoids. XXXIX. C 50 -carotenoids. 10. Bacterioruberin mono- and diglucoside. Acta Chem Scand 1972;26(6):2526–8.
[86] Shindo K, et al. Diapolycopenedioic acid xylosyl esters A, B, and C, novel antioxidative glyco-C30-carotenoic acids produced by a new marine bacterium Rubritalea squalenifaciens. J Antibiot 2008;61(3):185–91.
[87] Shindo K, Misawa N. New and rare carotenoids isolated from marine bacteria and their antioxidant activities. Mar Drugs 2014;12(3):1690.
[88] Hopmann C, et al. Isolation and structure elucidation of vancoresmycin—a new antibiotic from Amycolatopsis sp. ST 101170. Tetrahedron Lett 2002;43(3):435–8.
[89] Lutnaes BF, Oren A, Liaaen-Jensen S. New C_{40}-carotenoid acyl glycoside as principal carotenoid in Salinibacter ruber, an extremely halophilic eubacterium. J Nat Prod 2002;65(9):1340–3.
[90] Lutnaes BF, et al. Carotenoids of thermophilic bacteria—rhodothermus marinus from submarine Icelandic hot springs. Biochem Syst Ecol 2004;32(5):455–68.
[91] Hertzberg S, Liaaen Jensen S. Bacterial carotenoids XX. The carotenoids of Mycobacterium phlei strain Vera. 2. The structures of the phlei-xanthophylls—two novel tertiary glucosides. Acta Chem Scand 1967;21(1):15–41.
[92] Vacheron M-J, et al. Structural Investigations on cell walls of Nocardia sp. Eur J Biochem 1972;29(1). p. 156–106.
[93] Takaichi S, Ishidsu J-i. Carotenoid glycoside ester from *Rhodococcus rhodochrous*. Methods Enzym 1992;213:366–74.
[94] Takaichi S, et al. Carotenoid glucoside mycolic acid esters from the nocardioform actinomycetes, Rhodococcus rhodochrous. Phytochemistry 1997;45(3):505–8.
[95] Kleinig H. Membranes from Myxococcus fulvus (myxobacterales) containing carotenoid glucosides. Biochim Biophys Acta (BBA) Biomembr 1972;274(2):489–98.
[96] Kleinig H, Reichenbach H, Achenbach H. Carotenoid pigments of stigmatella aurantiaca (Myxobacterales). Arch für Mikrobiol 1970;74(3):223–34.
[97] Takaichi S, et al. Absence of carotenes and presence of a tertiary methoxy group in a carotenoid from a thermophilic filamentous photosynthetic bacterium huncastenholzii. Plant Cell Physiol 2001;42(12):1355–62.
[98] Takaichi S, et al. New carotenoids from the thermophilic green sulfur bacterium Chlorobium tepidum: 1′,2′-dihydro-γ-carotene, 1′,2′-dihydrochlorobactene, and OH-chlorobactene glucoside ester, and the carotenoid composition of different strains. Arch Microbiol 1997;168(4):270–6.
[99] Takaichi S, et al. A monocyclic carotenoid glucoside ester is a major carotenoid in the green filamentous bacterium *Chloroflexus aurantiacus*. Plant Cell Physiol 1995;36(5):773–8.
[100] Burgess ML, et al. Carotenoid glycoside esters from the thermophilic bacterium Meiothermus ruber. J Nat Prod 1999;62(6):859–63.
[101] Takaichi S, Maoka T, Masamoto K. Myxoxanthophyll in Synechocystis sp. PCC 6803 is myxol 2'-dimethyl-fucoside, (3R,2'S)-myxol 2'-(2,4-di-O-methyl-alpha-L-fucoside), not rhamnoside. Plant Cell Physiol 2001;42(7):756–62.
[102] Richter TKS, Hughes CC, Moore BS. Sioxanthin, a novel glycosylated carotenoid, reveals an unusual subclustered biosynthetic pathway. Environ Microbiol 2015;17(6):2158–71.
[103] Dembitsky VM. Astonishing diversity of natural surfactants: 3. Carotenoid glycosides and isoprenoid glycolipids. Lipids 2005;40(6):535–57.
[104] Arpin N, Liaaen-Jensen S, Trouilloud M. *Bacterial carotenoids. 38. C 50 -carotenoids. 9*. Isolation of decaprenoxanthin mono- and diglucoside from an Arthrobacter sp. Acta Chem Scand 1972;26(6):2524–6.
[105] Häberli A, Bircher C, Pfander H. Isolation of a new carotenoid and two new carotenoid glycosides from Curtobacterium flaccumfaciens pvar poinsettiae. Helvetica Chim Acta 2000;83(2):328–35.
[106] Osawa A, et al. Characterization and antioxidative activities of rare C_{50} carotenoids-sarcinaxanthin, sarcinaxanthin monoglucoside, and sarcinaxanthin diglucoside obtained from Micrococcus yunnanensis. J Oleo Sci 2010;59(12):653–9.
[107] Osawa A, et al. Characterization and antioxidative activities of rare C(50) carotenoids-sarcinaxanthin, sarcinaxanthin monoglucoside, and sarcinaxanthin diglucoside-obtained from Micrococcus yunnanensis. J Oleo Sci 2010;59(12):653–9.
[108] Hunter CN, et al. Introduction of new carotenoids into the bacterial photosynthetic apparatus by combining the carotenoid biosynthetic pathways of Erwinia herbicola and Rhodobacter sphaeroides. J Bacteriol 1994;176(12):3692–7.
[109] Kull DR, Pfander H. Isolation and structure elucidation of carotenoid glycosides from the thermoacidophilic archaea Sulfolobus shibatae. J Nat Prod 1997;60(4):371–4.
[110] Hertzberg S, Borch G, Liaaen-Jensen S. Bacterial carotenoids. L. absolute configuration of zeaxanthin dirhamnoside. Arch Microbiol 1976;110(1):95–9.
[111] Asker D, et al. Astaxanthin dirhamnoside, a new astaxanthin derivative produced by a radio-tolerant bacterium, *Sphingomonas astaxanthinifaciens*. J Antibiot 2009;62(7):397–9.
[112] Kleinig H, et al. Carotenoid pigments of Sorangium compositum (Myxobacterales) including two new carotenoid glucoside esters and two new carotenoid rhamnosides. Arch für Mikrobiol 1971;78(3):224–33.
[113] Yokoyama A, et al. Thermozeaxanthins, new carotenoid-glycoside-esters from thermophilic eubacterium Thermus thermophilus. Tetrahedron Lett 1995;36(27):4901–4.
[114] Yokoyama A, Adachi K, Shizuri Y. New carotenoid glucosides, astaxanthin glucoside and adonixanthin glucoside, isolated from the astaxanthin-producing marine bacterium, Agrobacterium aurantiacum. J Nat Prod 1995;58(12):1929–33.
[115] Yokoyama A, Shizuri Y, Misawa N. Production of new carotenoids, astaxanthin glucosides, by Escherichia coli transformants carrying carotenoid biosynthesis genes. Tetrahedron Lett 1998;39(22):3709–12.
[116] Krubasik P, et al. Detailed biosynthetic pathway to decaprenoxanthin diglucoside in Corynebacterium glutamicum and identification of novel intermediates. Arch Microbiol 2001;176(3):217–23.
[117] Garcia Costas AM, et al. Identification of the bacteriochlorophylls, carotenoids, quinones, lipids, and hopanoids of "Candidatus Chloracidobacterium thermophilum". J Bacteriol 2012;194(5):1158–68.
[118] Hallenbeck PC, et al. Draft genome sequence of the photoheterotrophic Chloracidobacterium thermophilum strain OC1 found in a Mat at Ojo Caliente. Genome Announc 2016;4(1).
[119] Schmerk CL, et al. Elucidation of the Burkholderia cenocepacia hopanoid biosynthesis pathway uncovers functions for conserved proteins in hopanoid-producing bacteria. Environ Microbiol 2015;17(3):735–50.
[120] Moreau RA, et al. Analysis of intact hopanoids and other lipids from the bacterium Zymomonas mobilis by high-performance liquid chromatography. Anal Biochem 1995;224(1):293–301.
[121] Wu CH, et al. Quantitative hopanoid analysis enables robust pattern detection and comparison between laboratories. Geobiology 2015;13(4):391–407.
[122] Llopiz P, Neunlist S, Rohmer M. Prokaryotic triterpenoids: O-α-D-

glucuronopyranosyl bacteriohopanetetrol, a novel hopanoid from the bacterium *Rhodospirillum rubrum*. Biochem J 1992;287(1):159–61.
[123] Hirai Y, et al. Unique cholesteryl glucosides in Helicobacter pylori: composition and structural analysis. J Bacteriol 1995;177(18):5327–33.
[124] Haque M, et al. Steryl glycosides: a characteristic feature of the Helicobacter spp.? J Bacteriol 1995;177(18):5334–7.
[125] Stübs G, et al. Acylated cholesteryl galactosides are specific antigens of Borrelia causing lyme disease and frequently induce antibodies in late stages of disease. J Biol Chem 2009;284(20):13326–34.
[126] Smith PF. Biosynthesis of cholesteryl glucoside by Mycoplasma gallinarum. J Bacteriol 1971;108(3):986–91.
[127] Mayberry WR, Smith PF. Structures and properties of acyl diglucosylcholesterol and galactofuranosyl diacylglycerol from Acholeplasma axanthum. Biochim Biophys Acta 1983;752(3):434–43.
[128] Spoeckner S, et al. Glycolipids of the smut fungus Ustilago maydis from cultivation on renewable resources. Appl Microbiol Biotechnol 1999;51(1):33–9.
[129] Morita T, et al. Production of mannosylerythritol lipids and their application in cosmetics. Appl Microbiol Biotechnol 2013;97(11):4691–700.
[130] Kakugawa K, et al. Isolation of yeast Kurtzmanomyces sp. I-11, novel producer of mannosylerythritol lipid. Biosci Biotechnol Biochem 2002;66(1):188–91.
[131] Kurz M, et al. Ustilipids, acylated β-D-mannopyranosyl D-erythritols from Ustilago maydis and Geotrichum candidum. J Antibiot (Tokyo) 2003;56(2):91–101.
[132] Morita T, et al. Formation of the two novel glycolipid biosurfactants, mannosylribitol lipid and mannosylarabitol lipid, by Pseudozyma parantarctica JCM 11752T. Appl Microbiol Biotechnol 2012;96(4):931–8.
[133] Kulakovskaya TV, et al. Ustilagic acid secretion by Pseudozyma fusiformata strains. FEMS Yeast Res 2005;5(10):919–23.
[134] Kulakovskaya TV, et al. Characterization of an antifungal glycolipid secreted by the yeast Sympodiomycopsis paphiopedili. FEMS Yeast Res 2004;5(3):247–52.
[135] Puchkov EO, et al. The mycocidal, membrane-active complex of Cryptococcus humicola is a new type of cellobiose lipid with detergent features. Biochim Biophys Acta (BBA) Biomembr 2002;1558(2):161–70.
[136] Cheng Y, et al. Insertional mutagenesis of a fungal biocontrol agent led to discovery of a rare cellobiose lipid with antifungal activity. Appl Environ Microbiol 2003;69(5):2595–602.
[137] Daniel H-J, et al. Production of sophorolipids from whey: development of a two-stage process with Cryptococcus curvatus ATCC 20509 and Candida bombicola ATCC 22214 using deproteinized whey concentrates as substrates. Appl Microbiol Biotechnol 1999;51(1):40–5.
[138] Ma X, Li H, Song X. Surface and biological activity of sophorolipid molecules produced by Wickerhamiella domercqiae var. sophorolipid CGMCC 1576. J Colloid Interface Sci 2012;376(1):165–72.
[139] Chen J, et al. Production, structure elucidation and anticancer properties of sophorolipid from Wickerhamiella domercqiae. Enzyme Microb Technol 2006;39(3):501–6.
[140] Price NPJ, et al. Structural characterization of novel sophorolipid biosurfactants from a newly identified species of Candida yeast. Carbohydr Res 2012;348:33–41.
[141] Nishida F, et al. Structure elucidation of glykenin, glycosidic antibiotics from Basidiomycetes sp. VII. Structure elucidation of the GK components using tandem mass spectrometry. J Mass Spectrom Soc Jpn 1995;43(1):37–44.
[142] Laine RA, et al. Monoglucosyloxyoctadecenoic acid, a glycolipid from *Aspergillus Niger*. Biochemistry 1972;11(12):2267–71.
[143] Chen C, et al. Halymecins, new antimicroalgal substances produced by fungi isolated from marine algae. J Antibiot (Tokyo) 1996;49(10):998–1005.
[144] Le Dang Q, et al. Antimicrobial activities of novel mannosyl lipids isolated from the biocontrol fungus Simplicillium lamellicola BCP against phytopathogenic bacteria. J Agric Food Chem 2014;62(15):3363–70.
[145] Tabata N, et al. Structure elucidation of roselipins, inhibitors of diacylglycerol acyltransferase produced by Gliocladium roseum KF-1040. J Antibiot (Tokyo) 1999;52(9):815–26.
[146] Tomoda H, et al. Roselipins, inhibitors of diacylglycerol acyltransferase, produced by Gliocladium roseum KF-1040. J Antibiot (Tokyo) 1999;52(8):689–94.
[147] Kohno J, et al. Isolation and structure determination of TMC-151s: novel polyketide antibiotics from Gliocladium catenulatum Gilman & Abbott TC 1280. Tetrahedron 1999;55(25):7771–86.
[148] Kohno J, et al. TMC-171A, B,C and TMC-154, novel polyketide antibiotics produced by Gliocladium sp. TC 1304 and TC 1282. J Antibiot (Tokyo) 1999;52(12):1114–23.
[149] Ayers S, et al. Anthelmintic constituents of Clonostachys candelabrum. J Antibiot 2010;63(3):119–22.
[150] Kasai Y, et al. Cladionol A, a polyketide glycoside from marine-derived fungus Gliocladium species. J Nat Prod 2005;68(5):777–9.
[151] Boros C, et al. Emmyguyacins A and B: Unusual glycolipids from a sterile fungus species that inhibit the low-pH conformational change of hemagglutinin A during replication of influenza virus. J Nat Prod 2002;65(2):108–14.
[152] Sakaki T, et al. Sterol glycosides and cerebrosides accumulate in Pichia pastoris, Rhynchosporium secalis and other fungi under normal conditions or under heat shock and ethanol stress. Yeast 2001;18(8):679–95.
[153] Mimee B, Labbé C, Bélanger RR. Catabolism of flocculosin, an antimicrobial metabolite produced by Pseudozyma flocculosa. Glycobiology 2009;19(9):995–1001.
[154] Van Bogaert INA, et al. The biosynthetic gene cluster for sophorolipids: a biotechnological interesting biosurfactant produced by Starmerella bombicola. Mol Microbiol 2013;88(3):501–9.
[155] Van Hamme JD, Singh A, Ward OP. Physiological aspects: Part 1 in a series of papers devoted to surfactants in microbiology and biotechnology. Biotechnol Adv 2006;24(6):604–20.
[156] Ron EZ, Rosenberg E. Natural roles of biosurfactants. Environ Microbiol 2001;3(4):229–36.
[157] Kannenberg EL, Poralla K. Hopanoid biosynthesis and function in bacteria. Naturwissenschaften 1999;86(4):168–76.
[158] Cortés-Sánchez AdJ, Hernández-Sánchez H, Jaramillo-Flores ME. Biological activity of glycolipids produced by microorganisms: new trends and possible therapeutic alternatives. Microbiol Res 2013;168(1):22–32.
[159] Rodrigues L, et al. Biosurfactants: potential applications in medicine. J Antimicrob Chemother 2006;57(4):609–18.
[160] Gudiña EJ, et al. Potential therapeutic applications of biosurfactants. Trends Pharmacol Sci 2013;34(12):667–75.
[161] Arcamone FM, et al. Melanosporin and elaiophylin, new antibiotics from Streptomyces melanosporus (sive melonsporofaciens) n. sp. G Microbiol 1959;7(3):207–16.
[162] Fang A, Wong GK, Demain AL. Enhancement of the antifungal activity of rapamycin by the coproduced elaiophylin and nigericin. J Antibiot (Tokyo) 2000;53(2):158–62.
[163] Isoda H, et al. Succinoyl trehalose lipid induced differentiation of human monocytoid leukemic cell line U937 into monocyte-macrophages. Cytotechnology 1995;19(1):79–88.
[164] Saita N, et al. Trehalose 6,6′-dimycolate (cord factor) of Mycobacterium tuberculosis induces corneal angiogenesis in rats. Infect Immun 2000;68(10):5991–7.
[165] Matsuno T, Miki W. Biological functions and activities of animal carotenoids. Pure Appl Chem 1991;63(1):141–6.
[166] Mimee B, et al. Antifungal activity of flocculosin, a novel glycolipid isolated from Pseudozyma flocculosa. Antimicrob Agents Chemother 2005;49(4):1597–9.
[167] Puchkov EO, et al. Cytoplasmic membrane of a sensitive yeast is a primary target for Cryptococcus humicola mycocidal compound (microcin). Biochim Biophys Acta (BBA) Biomembr 2001;1512(2):239–50.
[168] Kulakovskaya TV, Kulakovskaya EV, Golubev WI. ATP leakage from yeast cells treated by extracellular glycolipids of Pseudozyma fusiformata. FEMS Yeast Res 2003;3(4):401–4.
[169] Hammann P, Kretzschmar G, Seibert G. Secondary metabolites by chemical screening. 7: I. Elaiophylin derivatives and their biological activities. J Antibiot 1990;43(11):1431–40.
[170] Otoguro K, et al. In vitro and in vivo antiprotozoal activities of bispolides and their derivatives. J Antibiot 2010;63(5):275–7.
[171] Isoda H, et al. The neurite-initiating effect of microbial extracellular glycolipids in PC12 cells. Cytotechnology 1999;31(1):165.
[172] Shibahara M, et al. Mannosylerythritol lipid increases levels of galactocerамide in and neurite outgrowth from PC12 pheochromocytoma cells. Cytotechnology 2000;33(1):247–51.
[173] Wakamatsu Y, et al. Mannosylerythritol lipid induces characteristics of neuronal differentiation in PC12 cells through an ERK-related signal cascade. Eur J Biochem 2001;268(2):374–83.
[174] Isoda H, et al. Differentiation of human promyelocytic leukemia cell line HL60 by microbial extracellular glycolipids. Lipids 1997;32(3):263–71.
[175] Isoda H, et al. Microbial extracellular glycolipid induction of differentiation and inhibition of the protein kinase C activity of human promyelocytic leukemia cell line HL60. Biosci Biotechnol Biochem 1997;61(4):609–14.
[176] Lee S-Y, et al. Structure determination and biological activities of elaiophylin produced by Streptomyces sp. MCY-846. J Microbiol Biotechnol 1996;6:245–9.
[177] Ritzau M, et al. New macrodiolide antibiotics, 11-O-monomethyl- and 11,11'-O-dimethylelaiophylins, from Streptomyces sp. HKI-0113 and HKI-0114. J Nat Prod 1998;61(11):1337–9.
[178] Haferburg D, et al. Antiphytovirale Aktivität von rhamnolipid aus Pseudomonas aeruginosa. Acta Biotechnol 1987;7(4):353–6.
[179] Janek T, Lukaszewicz M, Krasowska A. Identification and characterization of biosurfactants produced by the Arctic bacterium Pseudomonas putida BD2. Colloids Surf B Biointerfaces 2013;110:379–86.
[180] Rodrigues LR, et al. Interference in adhesion of bacteria and yeasts isolated from explanted voice prostheses to silicone rubber by rhamnolipid biosurfactants. J Appl Microbiol 2006;100(3):470–80.
[181] Stipcevic T, Pijac A, Pijac G. Enhanced healing of full-thickness burn wounds using di-rhamnolipid. Burns 2006;32(1):24–34.
[182] Kitamoto D, et al. Surface active properties and antimicrobial activities of mannosylerythritol lipids as biosurfactants produced by Candida Antarctica. J Biotechnol 1993;29(1):91–6.
[183] Azuma M, et al. Role of interferon in the augmented resistance of trehalose-6,6′-dimycolate-treated mice to influenza virus infection. J General Virol 1987;68(3):835–43.
[184] Watanabe R, et al. Inhibitory effect of trehalose dimycolate (TDM) and its stereoisometric derivatives, trehalose dicorynomycolates (TDCMs), with low

toxicity on lung metastasis of tumour cells in mice. Vaccine 1999;17(11):1484–92.
[185] Andra J, et al. Endotoxin-like properties of a rhamnolipid exotoxin from Burkholderia (Pseudomonas) plantarii: immune cell stimulation and biophysical characterization. Biol Chem 2006;387(3):301–10.
[186] Díaz De Rienzo MA, et al. Sophorolipid biosurfactants: possible uses as antibacterial and antibiofilm agent. New Biotechnol 2015;32(6):720–6.
[187] Lydon HL, Baccile N, Callaghan B, Marchant R, Mitchell CA, Banat IM. Adjuvant antibiotic activity of acidic sophorolipids with potential for facilitating wound healing. Antimicrob Agents Chemother 2017. https://doi.org/10.1128/aac.02547-16.
[188] Haskins RH, Thorn JA. Biochemistry of the ustilaginales: VII. Antibiotic activity of ustilagic acid. Can J Bot 1951;29(6):585–92.
[189] Mimee B, Pelletier R, Bélanger RR. In vitro antibacterial activity and antifungal mode of action of flocculosin, a membrane-active cellobiose lipid. J Appl Microbiol 2009;107(3):989–96.
[190] Tulloch AP. Glycosides of hydroxy fatty acids. In: Kates M, editor. Glycolipids, phosphoglycolipids, and sulfoglycolipids. Boston, MA: Springer US; 1990. p. 463–87.
[191] Naruse N, et al. Fluvirucins A1, A2, B1, B2, B3, B4 and B5, new antibiotics active against influenza A virus. I. Production, isolation, chemical properties and biological activities. J Antibiot 1991;44(7):733–40.
[192] Besra GS, et al. Structural elucidation of a novel family of acyltrehaloses from Mycobacterium tuberculosis. Biochemistry 1992;31(40):9832–7.
[193] Saavedra R, et al. Mycobacterial di-O-acyl-trehalose inhibits mitogen- and antigen-induced proliferation of murine T cells in vitro. Clin Diagn Lab Immunol 2001;8(6):1081–8.
[194] Nishida F, et al. Structures of deacetyl glykenins-a, b, and c, glycosidic antibiotics from basidiomycetes sp. Tetrahedron Lett 1988;29(41):5287–90.
[195] Perez M, et al. PM100117 and PM100118, new antitumor macrolides produced by a marine Streptomyces caniferus GUA-06-05-006A. J Antibiot 2016;69(5):388–94.
[196] Tanaka Y, et al. Brasilinolide A, a new macrolide antibiotic produced by Nocardia brasiliensis: producing strain, isolation and biological activity. J Antibiot (Tokyo) 1997;50(12):1036–41.
[197] Shah V, et al. Sophorolipids, microbial glycolipids with anti-human immunodeficiency virus and sperm-immobilizing activities. Antimicrob Agents Chemother 2005;49(10):4093–100.
[198] Manitchotpisit P, et al. Aureobasidium pullulans as a source of liamocins (heavy oils) with anticancer activity. World J Microbiol Biotechnol 2014;30(8):2199–204.
[199] Bischoff KM, et al. Liamocin oil from Aureobasidium pullulans has antibacterial activity with specificity for species of Streptococcus. J Antibiot 2015;68(10):642–5.
[200] Price NPJ, et al. Polyols, not sugars, determine the structural diversity of antistreptococcal liamocins produced by Aureobasidium pullulans strain NRRL 50380. J Antibiot 2017;70(2):136–41.
[201] Lee S-Y, et al. Immunosuppressive activity of elaiophylins. J Microbiol Biotechnol 1997;7(4):272–7.
[202] Inokoshi J, et al. Expression of two human acyl-CoA:diacylglycerol acyltransferase isozymes in yeast and selectivity of microbial inhibitors toward the isozymes. J Antibiot 2009;62(1):51–4.
[203] Singh SB, et al. Isolation, structure, and HIV-1-integrase inhibitory activity of structurally diverse fungal metabolites. J Ind Microbiol Biotechnol 2003;30(12):721–31.
[204] Zhao X, et al. Treatment of mouse melanoma cells with phorbol 12-myristate 13-acetate counteracts mannosylerythritol lipid-induced growth arrest and apoptosis. Cytotechnology 2000;33(1):123–30.
[205] Zhao X, et al. Mannosylerythritol lipid is a potent inducer of apoptosis and differentiation of mouse melanoma cells in culture. Cancer Res 1999;59(2):482–6.
[206] Jyonouchi H, Zhang L, Tomita Y. Studies of immunomodulating actions of carotenoids. II. Astaxanthin enhances in vitro antibody production to T-dependent antigens without facilitating polyclonal B-cell activation. Nutr Cancer 1993;19(3):269–80.
[207] Haba E, et al. Physicochemical characterization and antimicrobial properties of rhamnolipids produced by Pseudomonas aeruginosa 47T2 NCBIM 40044. Biotechnol Bioeng 2003;81(3):316–22.
[208] Benincasa M, et al. Chemical structure, surface properties and biological activities of the biosurfactant produced by Pseudomonas aeruginosa LBI from soapstock. Antonie Leeuwenhoek 2004;85(1):1–8.
[209] Miao S, et al. Rhamnolipids as platform molecules for production of potential anti-zoospore agrochemicals. J Agric Food Chem 2015;63(13):3367–76.
[210] Stanghellini ME, Miller RM. Biosurfactants: Their identity and potential efficacy in the biological control of zoosporic plant pathogens. Plant Dis 1997;81(1):4–12.
[211] Yoo DS, Lee BS, Kim EK. Characteristics of microbial biosurfactant as an antifungal agent against plant pathogenic fungus. J Microbiol Biotechnol 2005;15(6):1164–9.
[212] Sanchez L, et al. Rhamnolipids elicit defense responses and induce disease resistance against biotrophic, hemibiotrophic, and necrotrophic pathogens that require different signaling pathways in Arabidopsis and highlight a central role for salicylic acid. Plant Physiol 2012;160(3):1630–41.
[213] Fu SL, et al. Sophorolipid treatment decreases LPS induced inflammatory responses and NO production in macrophages. J Am Coll Surg 2007;205(3):S44.
[214] Hagler M, et al. Sophorolipids decrease IgE production in U266 cells by downregulation of BSAP (Pax5), TLR-2, STAT3 and IL-6. J Allergy Clin Immunol 2007;119(1):S263.
[215] Williams GJ, Thorson JS. Natural product glycosyltransferases: properties and applications. In: Advances in enzymology and related areas of molecular biology. John Wiley & Sons, Inc; 2009. p. 55–119.
[216] Röttig A, Steinbüchel A. Acyltransferases in bacteria. Microbiol Mol Biol Rev 2013;77(2):277–321.
[217] Davies G, Henrissat B. Structures and mechanisms of glycosyl hydrolases. Structure 1995;3(9):853–9.
[218] Mukai K, et al. Substrate specificities in hydrolysis of polyhydroxyalkanoates by microbial esterases. Biotechnol Lett 1993;15(6):601–4.
[219] Jaeger KE, Steinbüchel A, Jendrossek D. Substrate specificities of bacterial polyhydroxyalkanoate depolymerases and lipases: bacterial lipases hydrolyze poly(ω-hydroxyalkanoates). Appl Environ Microbiol 1995;61(8):3113–8.
[220] Ali YB, Verger R, Abousalham A. Lipases or esterases: does it really matter? Toward a new bio-physico-chemical classification. In: Sandoval G, editor. Lipases and phospholipases: methods and protocols. Totowa, NJ: Humana Press; 2012. p. 31–51.
[221] Kulakovskaya E, Kulakovskaya T. Chapter 5-Metabolism of yeast extracellular glycolipids. In: Extracellular glycolipids of yeasts. Academic Press; 2014. p. 65–74.
[222] Van Bogaert INA, et al. Sophorolipids: microbial synthesis and application. In: Biosurfactants. CRC Press; 2014. p. 19–36.
[223] Hundle BS, et al. Functional expression of zeaxanthin glucosyltransferase from Erwinia herbicola and a proposed uridine diphosphate binding site. Proc Natl Acad Sci 1992;89(19):9321–5.
[224] Grille S, et al. The functions of steryl glycosides come to those who wait: recent advances in plants, fungi, bacteria and animals. Prog Lipid Res 2010;49(3):262–88.
[225] Teichmann B, et al. A biosynthetic gene cluster for a secreted cellobiose lipid with antifungal activity from Ustilago maydis. Mol Microbiol 2007;66(2):525–33.
[226] Abdel-Mawgoud AM, et al. Rhamnolipids: detection, analysis, biosynthesis, genetic regulation & bioengineering of production. In: Soberón-Chávez G, editor. Biosurfactants: from genes to applications (microbiology monographs). Münster, Germany: Springer; 2011. p. 13–55.
[227] Marrakchi H, Lanéelle M-A, Daffé M. Mycolic acids: structures, biosynthesis, and beyond. Chem Biol 2014;21(1):67–85.
[228] Ogasawara Y, et al. Cloning, sequencing, and functional analysis of the biosynthetic gene cluster of macrolactam antibiotic vicenistatin in Streptomyces halstedii. Chem Biol 2004;11(1):79–86.
[229] Morita T, et al. Genome and transcriptome analysis of the basidiomycetous yeast Pseudozyma Antarctica producing extracellular glycolipids, mannosylerythritol lipids. PLOS ONE 2014;9(2), e86490.
[230] Mirończuk AM, Biegalska A, Dobrowolski A. Functional overexpression of genes involved in erythritol synthesis in the yeast Yarrowia lipolytica. Biotechnol Biofuels 2017;10(1):77.
[231] Irorere VU, et al. Microbial rhamnolipid production: a critical re-evaluation of published data and suggested future publication criteria. Appl Microbiol Biotechnol 2017;101(10):3941–51.
[232] Smyth TJP, et al. Protocols for the detection and chemical characterisation of microbial glycolipids. In: McGenity TJ, Timmis KN, Nogales B, editors. Hydrocarbon and lipid microbiology protocols : biochemical methods. Berlin, Heidelberg: Springer Berlin Heidelberg; 2016. p. 29–60.
[233] Abdel-Mawgoud AM, Lépine F, Déziel E. Liquid chromatography/mass spectrometry for the identification and quantification of rhamnolipids. In: Filloux A, Ramos J-L, editors. Pseudomonas methods and protocols. Springer New York; 2014. p. 359–73.
[234] Palos Pacheco R, et al. Synthesis and characterization of four diastereomers of monorhamnolipids. J Am Chem Soc 2017;139(14):5125–32.
[235] Menhour B, et al. A stereocontrolled synthesis of the hydrophobic moiety of rhamnolipids. Tetrahedron Lett 2015;56(9):1159–61.
[236] Bauer J, et al. Chemical synthesis of a glycolipid library by a solid-phase strategy allows elucidation of the structural specificity of immunostimulation by rhamnolipids. Chem – A Eur J 2006;12(27):7116–24.
[237] Fleurackers S. Biosurfactants versus chemically synthesized surface-active agents. In: Biosurfactants. CRC Press; 2014. p. 37–48.
[238] Makkar RS, Cameotra SS, Banat IM. Advances in utilization of renewable substrates for biosurfactant production. AMB Express 2011;1(1):5.
[239] Banat IM, et al. Cost effective technologies and renewable substrates for biosurfactants' production. Front Microbiol 2014;5(697).
[240] Koch AK, et al. Genetic construction of lactose-utilizing strains of Pseudomonas aeruginosa and their application in biosurfactant production. Nat Biotech 1988;6(11):1335–9.
[241] Meijnen J-P, de Winde JH, Ruijssenaars HJ. Engineering Pseudomonas putida S12 for efficient utilization of D-xylose and L-arabinose. Appl Environ Microbiol 2008;74(16):5031–7.
[242] Zahoor A, Lindner SN, Wendisch VF. Metabolic engineering of Corynebacterium glutamicum aimed at alternative carbon sources and new products. Computational and Structural Biotechnology Journal 2012;3(4), e201210004.
[243] Wei N, et al. Simultaneous utilization of cellobiose, xylose, and acetic acid from lignocellulosic biomass for biofuel production by an engineered yeast

platform. ACS Synth Biol 2015;4(6):707–13.
[244] Shah V, Jurjevic M, Badia D. Utilization of restaurant waste oil as a precursor for sophorolipid production. Biotechnol Prog 2007;23(2):512–5.
[245] Ozdal M, Gurkok S, Ozdal OG. Optimization of rhamnolipid production by Pseudomonas aeruginosa OG1 using waste frying oil and chicken feather peptone. 3 Biotech 2017;7(2):117.
[246] Wittgens A, et al. Growth independent rhamnolipid production from glucose using the non-pathogenic Pseudomonas putida KT2440. Microb Cell Factories 2011;10(1):80.
[247] Dobler L, et al. Rhamnolipids in perspective: gene regulatory pathways, metabolic engineering, production and technological forecasting. New Biotechnol 2016;33(1):123–35.
[248] Ochsner UA, et al. Production of Pseudomonas aeruginosa rhamnolipid biosurfactants in heterologous hosts. Appl Environ Microbiol 1995;61(9):3503–6.
[249] Wang QH, et al. Engineering bacteria for production of rhamnolipid as an agent for enhanced oil recovery. Biotechnol Bioeng 2007;98(4):842–53.
[250] Cabrera-Valladares N, et al. Monorhamnolipids and 3-(3-hydroxyalkanoyloxy)alkanoic acids (HAAs) production using Escherichia coli as a heterologous host. Appl Microbiol Biotechnol 2006;73(1):187–94.
[251] Solaiman DY, et al. Dirhamnose-lipid production by recombinant nonpathogenic bacterium Pseudomonas chlororaphis. Appl Microbiol Biotechnol 2015;99(10):4333–42.
[252] Tavares LFD, et al. Characterization of rhamnolipids produced by wild-type and engineered Burkholderia kururiensis. Appl Microbiol Biotechnol 2013;97(5):1909–21.
[253] Müller MM, Hausmann R. Regulatory and metabolic network of rhamnolipid biosynthesis: traditional and advanced engineering towards biotechnological production. Appl Microbiol Biotechnol 2011;91(2):251–64.
[254] Heider SAE, et al. Production and glucosylation of C_{50} and C_{40} carotenoids by metabolically engineered Corynebacterium glutamicum. Appl Microbiol Biotechnol 2014;98(3):1223–35.
[255] Peirú S, et al. Production of the potent antibacterial polyketide erythromycin C in Escherichia coli. Appl Environ Microbiol 2005;71(5):2539–47.
[256] Baltz RH. Molecular engineering approaches to peptide, polyketide and other antibiotics. Nat Biotech 2006;24(12):1533–40.
[257] Abdel-Mawgoud AM, Lépine F, Déziel E. A chiral high-performance liquid chromatography–tandem mass spectrometry method for the stereospecific analysis of enoyl-coenzyme A hydratases/isomerases. J Chromatogr A 2013;1306:37–43.
[258] Abdel-Mawgoud AM, Lépine F, Déziel E. A stereospecific pathway diverts β-oxidation intermediates to the biosynthesis of rhamnolipid biosurfactants. Chem Biol 2014;21(1):156–64.
[259] Adrio JL. Oleaginous yeasts: promising platforms for the production of oleochemicals and biofuels. Biotechnol Bioeng 2017;114:1915–20. https://doi.org/10.1002/bit.26337.
[260] Ageitos JM, et al. Oily yeasts as oleaginous cell factories. Appl Microbiol Biotechnol 2011;90(4):1219–27.
[261] Baccile N, et al. Development of a cradle-to-grave approach for acetylated acidic sophorolipid biosurfactants. ACS Sustain Chem Eng 2017;5(1):1186–98.
[262] Ciesielska K, et al. Exoproteome analysis of Starmerella bombicola results in the discovery of an esterase required for lactonization of sophorolipids. J Proteomics 2014;98(Supplement C):159–74.