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Review

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Diversity and origin of carotenoid biosynthesis: its history of coevolution towards plant photosynthesis

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Summary

The development of photosynthesis was a highlight in the progression of bacteria. In addition to the photosystems with their structural proteins, the photosynthesis apparatus consists of different cofactors including essential carotenoids. Thus, the evolution of the carotenoid pathways in relation to the functionality of the resulting structures in photosynthesis is the focus of this review. Analysis of carotenoid pathway genes indicates early evolutionary roots in prokaryotes. The pathway complexity leading to a multitude of structures is a result of gene acquisition, including their functional modifications, emergence of novel genes and gene exchange between species. Along with the progression of photosynthesis, carotenoid pathways coevolved with photosynthesis according to their advancing functionality. Cyanobacteria, with their oxygenic photosynthesis, became a landmark for evolutionary events including carotenogenesis. Concurrent with endosymbiosis, the cyanobacterial carotenoid pathways were inherited into algal plastids. In the lineage leading to Chlorophyta and plants, carotenoids evolved to their prominent role in protection and regulation of light energy input as constituents of a highly efficient light-harvesting complex.

I. Overview of carotenoid structures and function

The photosynthesis apparatus is a complex system of proteins and different types of cofactors. This includes the synthesis of customized functional pigments including carotenoids. The chain length of these terpenoids varies from C_{30} to C_{40} and C_{50} and they possess various modifications at the end groups. A prominent structural feature of carotenoids is an extended conjugated double-bond system. More than 700 different natural carotenoid structures have been identified (Britton *et al.*, 2004), many of them present in bacteria. An outline on basic carotenoid structures and their modifications is presented in Fig. 1(a). Carotenoids are functional pigments and their diverse structures determine their multiple functions. Due to their lipophilic nature

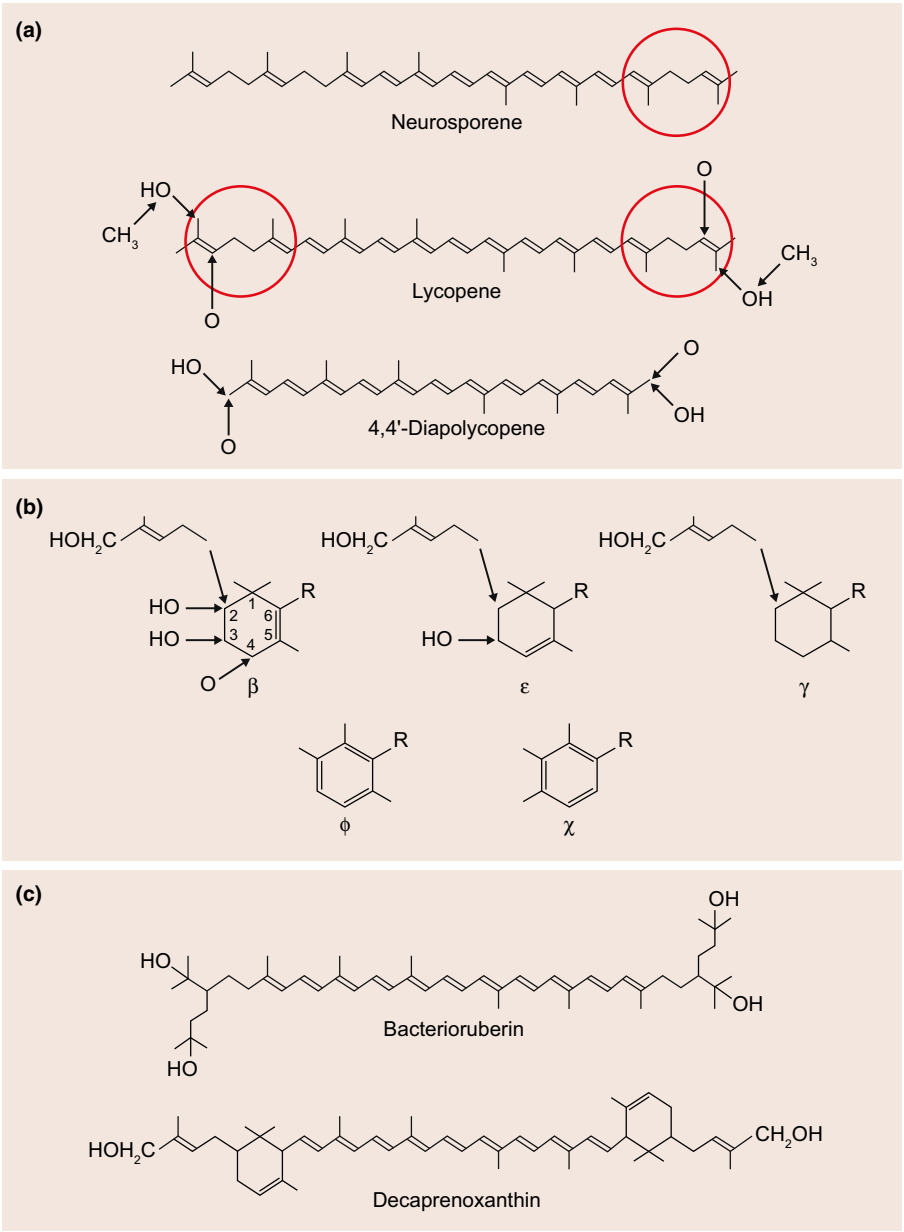

Fig. 1 Carotenoid structures: (a) modifications of basic C_{30} and C_{40} acyclic carbon skeletons; (b) cyclic end groups of bacterial carotenoids; (c) C_{50} carotenoids present in Archaea and Bacteria. Arrows indicate the position of potential substituents; circled regions can be cyclized to ionone rings; R indicates residual carotenoid structure.

and their terminal polar substituents, they are membrane stabilizers by spanning the membrane with the lipophilic central part of the molecule and anchoring in the polar outer membrane region with their polar head groups (Gruszecki & Strzalka, 2005). This is one function of carotenoids, especially in extremophilic Archaea and Bacteria such as halophilic *Halobacterium* (Lazrak *et al.*, 1988) or thermophilic *Thermus* (Yokoyama *et al.*, 1995). The polyene system enables the carotenoids to quench photoexcited and photosensitized molecules including singlet oxygen and to dissipate the radiation energy as heat. It also accounts for the ability of carotenoids to absorb light in energy-conservation processes (Britton, 1995). Both photofunctions make carotenoids essential components for photosynthesis (Ruban, 2015). There, they are an integral part of functional pigment protein complexes. By endosymbiosis, the ability of carotenoid formation was transferred from Cyanobacteria to algae and further on to plants.

In coevolution with photosynthesis, carotenoid structures were optimized to play their protective role and their light-harvesting role. Given this central process for life on our planet, this review is focused on the evolutionary adaptation of specialized carotenoid structures and pathways to photosynthesis. In this context, the ultimate aspect is how carotenoid synthesis was inherited via cyanobacteria into algal plastids in the lineage leading to plants to finally play their efficient protective and light-harvesting role in plant oxygenic photosynthesis.

II. Carotenoid genes and enzymes of Archaea

The basic part of the carotenoid pathway comprises the synthesis of C_{40} phytoene, its desaturation and in many cases cyclization of both ends of the linear hydrocarbon. The genes for the basic C_{40} pathway are well conserved in Archaea and Bacteria, indicating a common carotenogenic progenitor. However, the major carotenoid synthesized by Euryarchaeota is the C_{50} bacterioruberin (Fig. 1c) acting as a membrane stabilizer (Lazrak *et al.*, 1988). Genes involved in its biosynthesis are *crtB* encoding a universal phytoene synthase that is conserved in almost all carotenogenic species. Its function was first elucidated by heterologous expression of a bacterial gene (Sandmann & Misawa, 1992). The following reaction is catalysed by the product of the phytoene desaturase gene *crtI*. This enzyme inserts four double bonds extending the polyene structure to form all-E lycopene. The resulting lycopene molecule is extended by addition of an isoprenoid cation from dimethylallyl pyrophosphate to the double bond both at C-2 and C-2′ by a prenyl transferase. In the next step, double bonds are inserted at C-3,4 and C-3′,4′ by the product of the gene referred to in this review as $crtD_A$, which is evolutionally related to *crtI* (Yang *et al.*, 2015). Finally, both added prenyl groups at each end of the molecule become hydroxylated by addition of water to their C-2″,3″ and C-2‴,3‴ double bonds by the *cruF*-encoded enzyme. All genes of the pathway have been cloned from *Haloarcula japonica* (Yang *et al.*, 2015). In the class Halobacteria, lycopene is cyclized to β-carotene as a branch parallel to the C_{50} pathway. The gene for this cyclase from *Halobacterium salinarum* referred in this review as $crtY_A$ represents the ancient type of a group of multiple cyclases (Peck *et al.*, 2002). In these halophilic species, β-carotene is cleaved to retinal. The apocarotenoid retinal is a component of bacteriorhodopsin where it is protein-bound and functions as a light receptor. Thus, retinal is the earliest example of light absorption by carotenoids in an energy-conservation processes. This proton pumping membrane-bound bacteriorhodopsin of halophilic Archaea drives the generation of ATP (Oesterhelt, 1976) in association with the C_{50} carotenoid bacterioruberin (Fig. 1c) (Henderson, 1977). Similar rhodopsin-driven ion pumps can also be found in Bacteria (Kandori, 2015).

In the phylum Crenarchaeota, β-carotene is synthesized by the same lycopene cyclase and is then hydroxylated at C-3 and C-3′ to zeaxanthin by a CrtZ-type β-carotene hydroxylase (Klassen, 2010). After glycosylation of the hydroxyl groups, this resulting glucoside and rhamnoside (Kull & Pfander, 1997) replaces bacterioruberin as alternative membrane enforcers by spanning the membrane and clamping the bilayer by interaction of the glucose residues with the polar lipid heads (Gruszecki & Strzalka, 2005), with the added advantage of better photoprotection (Sandmann, 2019).

Looking at the lineage from Archaea to Eukarya, only Mucoromycota acquired a biosynthesis pathway to β-carotene resembling the situation in Halobacteria. This pathway occurs in several higher fungi including further modifications of carotenoid structure. A prominent feature of fungi is the fusion gene of a bifunctional lycopene cyclase-phytoene synthase *crtYB* with both individual domains (Verdoes *et al.*, 1999). It originated from the cotranscribed overlapping genes of $crtY_A$ and *crtB* similar to the organization found in the gene cluster of *Sulfolobus solfaticus* (Hemmi *et al.*, 2003). The protein resulting from *crtYB* expression undergoes posttranslational cleavage into independently functional phytoene synthase and lycopene cyclase (Breitenbach *et al.*, 2012).

III. Diversity of pathways and genes in Bacteria

Carotenoid structures formed in Bacteria and their biosynthesis pathways are more diverse compared to Archaea. According to a phylogenetic tree of *crtB* (Klassen, 2010), carotenogenesis of species of the Gram positive Actinobacteria with C_{50} carotenoids is most closest related to the archaeal pathway. Instead of the acyclic C_{50} bacterioruberin, the intermediate formed by extension of lycopene is cyclized to decaprenoxanthin (Fig. 1c) in species of the orders Corynebacteriales and Micrococcales in concerted action by a heterodimeric cyclase encoded by *crtYe* and *crtYf* (Krubasik & Sandmann, 2001). Both genes are adjacent in a carotenogenic gene cluster in *Corynebacterium*-related species (Sandmann & Yukawa, 2005). Catalytic activity was found only when both genes were expressed simultaneously. Both genes are related to archaeal $crtY_A$. The difference to $CrtY_A$ is the utilization of a C_{50} instead of a C_{40} substrate and the formation of the ε-ionone rings of decaprenoxanthin. Variations of this type of heterodimeric cyclase to *crtYg* with *crtYh* and *lytA,B*, respectively, are able to form β- (Netzer *et al.*, 2010) or γ-rings (Tao *et al.*, 2007) with a C_{50} substrate (ring structures in Fig. 1b). Most other bacterial species are unable to synthesize C_{50} carotenoids as membrane enforcers. Instead, this function has been replaced by hopanoids and sterols (Rohmer *et al.*, 1979; Wei *et al.*, 2016). Since this evolutionary pressure to keep C_{50} carotenoids was lost, they played no further role in other bacterial groups. The phylogenetic tree of *crtI* discriminates between species of the Actinobacteria with either a C_{50} or those with a C_{40} carotenoid pathway (Klassen, 2010). Within the orders Actinomycetales and Streptomycetales, a C_{40} pathway generates arylic isorenieratene with β-carotene as an intermediate. In the final step, the aromatic desaturase encoded by *crtU* desaturates both β-rings of β-carotene to the ϕ-rings (Krügel *et al.*, 1999). The only other bacterial group synthesizing arylic carotenoids are Chlorobi.

A modification of the initial reaction of carotenogenesis, the condensation of two molecules of C_{20} prenyl pyrophosphates in the C_{40} pathway, is the utilization of C_{15} prenyl pyrophosphates instead by diapophytoene synthase CrtM, resulting in the formation of a C_{30} diapophytoene. This diapocarotenoid resembles the starting point of the acyclic C_{30} pathway in Fimicutes. Acyclic C40 carotenoids are present in different orders of photosynthetic and nonphotosynthetic Proteobacteria. Acyclic carotenoid synthesis will be addressed below in the context of photosynthetic bacteria.

IV. Principles of carotenoid pathway evolution

1. Gene duplication and divergence of the *crtI* gene family

Novel protein-encoding genes may evolve by gene duplication. An advantageous duplicant can acquire a modified or novel function enabling the modification of an existing pathway. Even if the novel function is completely different to the original one, the encoded enzyme should have a related reaction mechanism. A prominent

example of gene duplication is the presence of two paralogous lycopene cyclase genes of *Prochlorococcus*. Both encoded cyclases convert lycopene but the original CrtL-b forms β-ionone rings whereas CrtL-e converts one half of lycopene into an ε-ring in the synthesis of α-carotene. However, CrtL-e has still retained some cyclase function for a β-ring (Stickforth *et al.*, 2003). In another α-carotene-synthesizing cyanobacterium, *Acaryochloris*, only one *crtL* and the *cruA/cruP* genes are present (Sugiyama & Takaichi, 2020). In *Bacillus firmus*, two orthologous genes *crtNb* and *crtNc* (Steiger *et al.*, 2015) are located next to each other in the genome and both encode oxidase which sequentially oxidize the same terminal carbon atoms in a C_{30} pathway to 4,4′-diapolycopen-4,4′-dial and 4,4′-diapolycopen-4,4′-dionic acid, respectively (Fig. 2b).

Several genes descended from a common ancestor can form a family of functionally different orthologous genes. A well-studied carotenogenic example is the *crtI* gene family. The phytoene desaturase gene in the majority of bacterial groups is *crtI* (Sandmann, 2021b). This gene represents a family of heterofunctional homologues with divergent biological functions (Sandmann, 2002). A genealogical tree of CrtI family genes can be found in Klassen (2010). They encode three different groups of enzymes such as desaturases, isomerases and hydroxylases. Their modified catalytic functions are illustrated in Fig. 2. As indicated there, their catalysis proceeds through a common step, the formation of an allylic carbocation by hydride transfer to FAD as cosubstrate in the reaction or by protonation of a double bond (Fraser *et al.*, 1992; Schaub *et al.*, 2012). The final step determines the type of reaction. CrtI from *Pantoea ananatis* (formerly *Erwinia uredovora*) was characterized as a four-step phytoene desaturase (Fig. 2a) which inserts four double bonds into the 15Z-phytoene molecule leading to the formation of all-*trans* lycopene by extension of the polyene system (Misawa *et al.*, 1990). Other phytoene desaturases from the purple bacterium *Rhodobacter* (Raisig *et al.*, 1996) or the fungus *Neurospora* (Hausmann & Sandmann, 2000) vary in their product specificity by carrying out only three or even five desaturation steps producing neurosporene or 3,4-dehydrolycopene, respectively. The phytoene desaturase from *Rubrivivax* exhibits the particular feature of synthesizing neurosporene in addition to lycopene (Harada *et al.*, 2001). Mutations in the carotene binding site (Schaub *et al.*, 2012) generated neurosporene producers by decreasing the specificity of neurosporene binding for the last desaturation step (Stickforth & Sandmann, 2011). Other biochemical factors determining the neurosporene to lycopene conversion are high amounts of enzyme vs low amounts of phytoene (Stickforth & Sandmann, 2007). Under these conditions, the three-step desaturase of *Rhodobacter* can be forced to synthesize lycopene *in vitro*. The same effect was observed in a *Rhodobacter* mutant with increased desaturase formation. In addition, a four-step desaturase can be forced to catalyse up to six desaturation steps by enzyme overexpression in *Escherichia coli* (Fraser *et al.*, 1992).

The diapophytoene desaturase CrtN, which uses a C_{30} substrate, is otherwise very similar to CrtI. It exhibits the same reaction mechanism and cofactor dependence and also exists as a three- and a four-step desaturase in *Staphylococcus* or *Methylomonas*, respectively (Raisig & Sandmann, 1999; Tao *et al.*, 2005). The enzyme is substrate-specific for C_{30} carotenes with the exception of C_{40} ζ-carotene which is also be converted by CrtN (Raisig & Sandmann, 2001). In return, CrtI is able to desaturate C_{30} diapo-ζ-carotene. CrtD from purple bacteria is a 1-hydroxycarotene 3,4-desaturase (Fig. 2a) which extends the polyene system generated by the products of *crtI* or *crtN*. Its alternative substrates are either 1-HO-neurosporene or 1-HO-lycopene (Steiger *et al.*, 2000). In a similar way to CrtD, the evolutionarily related product of $crtD_A$ of Archaea carries out the same desaturation reaction at C-3,4 but at the acyclic C_{50} substrate in the pathway to bacterioruberin.

The CrtI-related hydroxylases and isomerase of Fig. 2(b,c) diverged from desaturases (Fig. 2a) having acquired new catalytic properties. The hydroxylases form a hydroxyl group by reaction of the carbocation mentioned above with a hydroxyl ion. Two hydroxylation steps carried out at the same carbon cause the elimination of water leading to a keto (Breitenbach *et al.*, 2013a) or aldehyde group (Steiger *et al.*, 2015). CrtO, in general a two-step hydroxylase, inserts only one keto group at C4 but not at C4′ of β-carotene. However in *Nostoc*, CrtO works as a canthaxanthin producing diketolase (Schöpf *et al.*, 2013). The *crtO* gene is widespread in cyanobacteria and Chloroflexi. It was first identified from the cyanobacterium *Synechocystis* (Fernandez-Gonzalez *et al.*, 1997).

A C_{30} carotenoid pathway is typical for species of the order Bacillales of the phylum Firmicutes starting this pathway by a *crtM*-encoded diapophytoene synthase. A final product is 4,4′-diapolycopen-4,4′-dionic acid (Steiger *et al.*, 2015). The carboxyl group is formed by two reactions catalysed by the products of the homologous genes *crtNb* and *crtNc*. CrtNb hydroxylates a terminal methyl group at each end of diapolycopene twice leading to the 4,4′-dialdehyde by water elimination (Fig. 2b). The following hydroxylation step by CrtNc generates the 4,4′-diacid. Among bacteria other than bacilli, *Methylomonas* is the only known species possessing the genes for the pathway to 4,4′-diapolycopen-4,4′-dionic acid (Tao *et al.*, 2005). The gene *crtO* listed mistakenly for Bacilli (Klassen, 2010) in fact is a *crtNc* gene due to its phylogeny and the pathway in these species (Steiger *et al.*, 2015).

The isomerization mechanism of CrtH (Fig. 2c) involves carbocation formation by proton addition to the *cis* double bond, representing a reversed desaturation reaction. The next step is proton abstraction resulting in the formation of a thermodynamically favourable *trans* double bond. For the rearrangement of all original *cis* double bonds of 7,9,7′,9′-*cis* to *all-trans* lycopene, four catalytic reactions are necessary.

2. Gene diversity and displacement

Different types of lycopene cyclases with limited similarity but with a common NAD(P)H-binding domain exist (Krubasik & Sandmann, 2000a; Sugiyama & Takaichi, 2020). Nevertheless, their mechanism (including those of the C_{50} cyclases) is the same (Hornero-Mendez & Britton, 2002). It proceeds by protonation of C2 followed by addition of the resulting carbocation at C1 to carbon C6. Finally, proton abstraction from either C6, C4 or C18 determines the type of ring formation mentioned above (Fig. 1b). In the catalytic process, NAD(P)H plays an indirect role as an

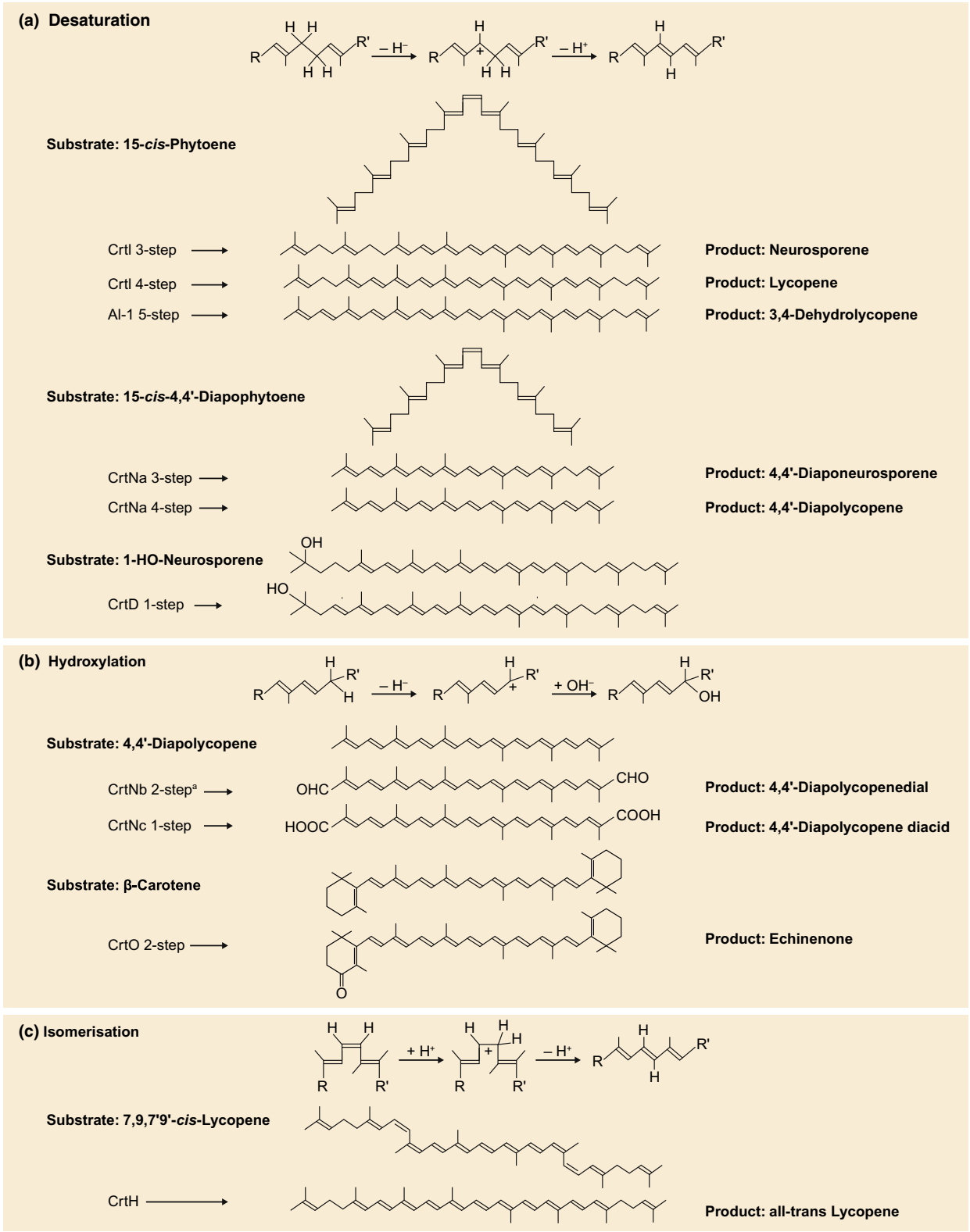

Fig. 2 The CrtI family and their development from desaturases to hydroxylases and isomerase with their enzymatic reactions. The common reactions of all these enzyme types with different substrates and the mechanisms leading to their final product. (a) Different substrate-specificities of desaturases; (b) hydroxylations resulting in hydroxy, keto and carboxylic derivatives; (c) isomerization mechanism of 7,9,7′,9′-*cis* lycopene. [a]Formation of an aldehyde group by two hydroxylations at the same carbon and water elimination.

activator (Schnurr *et al.*, 1996; Hornero-Mendez & Britton, 2002). In Actinomycetales, the β-carotene forming lycopene cyclase is encoded by the $crtY_A$-related genes *crtYc* in combination with *crtYd* (Krubasik & Sandmann, 2000b). Alternatively, the lycopene cyclase gene of the Streptomycetales is *crtY*, a novel nonhomologous type first described for a proteobacterium (Misawa *et al.*, 1990). This CrtY lycopene cyclase dominates in all bacterial groups, with the formation of cyclic carotenoids with the exception of cyanobacteria. Alternatively, species from the phylum Sphingobacteria, for example, use either *crtY* or the *crtYc*/*crtYd* pair for cyclization or possess both gene types simultaneously (Klassen, 2010). A third type of unrelated lycopene cyclase gene is *crtL*, exhibiting a distinct conserved pattern with *crtY* implying a common phylogenetic origin (Krubasik & Sandmann, 2000a). The two genes coexist in different isorenieratene-synthesizing *Mycobacterium* species (Klassen, 2010). *crtY*- and *crtL*-encoded enzymes can also function asymmetrically as monocyclases forming only a single β-ionone ring (Teramoto *et al.*, 2003; Tao *et al.*, 2004). In addition to the lycopene cyclase genes described above, a fourth type of lycopene cyclase gene, *cruA*, present in species of the phylum Chlorobi (Frigaard *et al.*, 2004) and in Cyanobacteria in which it is accompanied by a functional paralogous *cruP* gene that increases diversification. CruP may primarily be a mono cyclase (Maresca *et al.*, 2007; Sugiyama & Takaichi, 2020).

The most noticeable gene replacement occured in an ancestor of the Chlorobi–Cyanobacteria lineage. Instead of the *crtI* phytoene desaturase gene, a novel set of genes for this desaturation chain was acquired. In Cyanobacteria, this replacement was investigated in detail (Sandmann, 2009) and will be addressed below.

3. Horizontal gene transfer

Bacteria can obtain genes from other species by horizontal gene transfer, distributing genes among different species. Previous studies have showm that horizontal transfer of genes for carotenoid biosynthesis played a major role in the distribution of carotenoid pathways in unrelated phylogenetic lineages (Phadwal, 2005; Klassen, 2010). For example, phylogenetic analysis indicates for the *crtA* gene was transferred from Bacteriodetes to purple bacteria accompanied by a modification of its function (Klassen, 2009). Originally, the gene *crtA-OH* of *Flavobacterium* P99-3 encoded a 2′-hydroxylase in myxol biosynthesis (Rählert & Sandmann, 2009). In *Rhodobacter* and *Rubrivivax* this enzyme became a 2-ketolase for spheroidene and spirilloxanthin (structures in Fig. 3a) with broad substrate specificity (Gerjets *et al.*, 2009). Since the oxygen-dependent monooxygenase mechanism is retained, the keto derivatives can only be synthesized under aerobic growth conditions. This formation of spheroidenone (structure in Fig. 3a) is regarded as an induced photoprotective mechanism against oxygen (Slouf *et al.*, 2012). Another example is the *crtO* gene, which was acquired by horizontal transfer from *Nocardia* and other species of Corynebacterineae (Klassen, 2010). Apart from single gene transfer, a whole photosynthetic gene cluster with genes for structural photosynthesis genes, bacteriochlorophyll synthesis and all *crt* genes for the carotenoid pathway to 2,2′-diketospirilloxanthin was transferred from an α-subclass bacterium to the β-subclass purple bacterium *Rubrivivax* (Igarashi *et al.*, 2001). Plasmids play an important role in gene mobilization. In *Roseobacter*, plasmids have been detected which contain a photosynthesis gene cluster including the genes of the carotenoid pathway (Petersen *et al.*, 2012).

V. Carotenoids in prokaryotic photosynthesis

1. Anoxygenic phototrophs

Bacteria from four different phyla are able to carry out anoxygenic photosynthesis. The purple Proteobactera and Chloroflexi use a type 2 reaction centre (RC) for light energy utilization. This drives a cyclical redox chain via quinone, which develops a proton gradient. The proton motive force is used to generate ATP from ADP (Blankenship, 2002). The photoactive pigment of the reaction centre is bacteriochlorophyll, which in purple bacteria is associated either with the neurosporene-derived spheroidene or the lycopene-derived spirilloxanthin (Ermler *et al.*, 1994). These carotenoids protect the reaction centre bacteriochlorophyll against photooxidation by energy quenching from its triplet state and participate in light-harvesting in the surrounding antenna. A carotenoid biosynthesis gene cluster has been cloned from *Rhodobacter*, which allowed the first identification of most genes for the synthesis of acyclic spheroidene (Armstrong *et al.*, 1989). This pathway involves the desaturation of phytoene to neurosporene by the three-step *crtI* gene product, water addition to the C-1,2 double bond by the hydratase CrtC, 3,4-desaturation by the CrtI-related CrtD and methylation of the resulting hydroxyl group by CrtF. The reaction sequence leading to spheroidene has been demonstrated by pathway reconstruction in a heterologous host (Steiger *et al.*, 2000). In spirilloxanthin-synthesizing species from α-Proteobacteria the reaction product of a four-step CrtI desaturase is lycopene (Takaichi, 1999), which is then modified in the same way as described for spheroidene synthesis but at each ends of the molecule. In *Chloroflexus*, carotenoids are bicyclic β-carotene together with monocyclic γ-carotene (Fig. 3b) and its 1-hydroxyderivatives (Takaichi, 1999). In the genus *Chromatium* with a type 2 reaction centre, some species synthesize okenone (Fig. 3a) with genes similar to *Chloroflexus* including an additional *crtU* gene and instead of *crtD* the *cruO* gene, which encodes a C4 ketolase (Vogl & Bryant, 2011). In this anaerobic species, okenone function is primarily in energy transfer to bacteriochlorophyll (Andersson *et al.*, 1996).

Heliobacteria from the phylum Firmicutes and green sulphur bacteria species such as *Chlorobium* use a type 1 RC. Compared to a type 2 RC, its redox potential is more negative, resulting in the formation of the reductant ferredoxin in a linear electron transport reaction from H_2S as an electron source (Blankenship, 2002). Light-harvesting peripheral antenna with bacteriochlorophyll and carotenoids, the chlorosome, transfer radiation energy by excitonic coupling to the RC bacteriochlorophylls, which are also associated with carotenoids. The carotenoid pathway of *Chlorobium* produces chlorobactene (Fig. 3a), a monocyclic carotenoid with one ϕ-ionone ring whereas some species additionally synthesize isorenieratene with two ϕ-ionone rings (Fig. 3a). They are accompanied by a

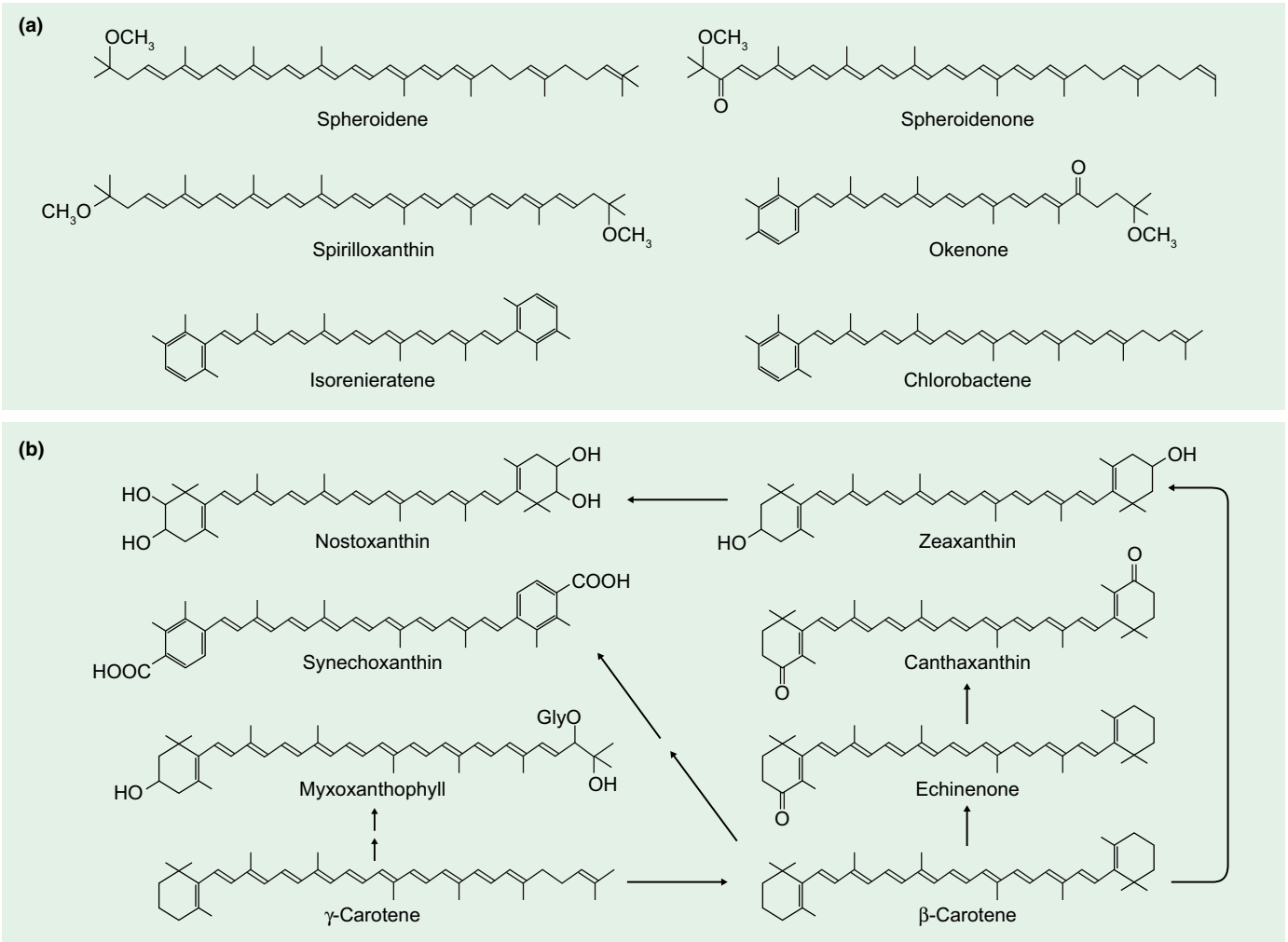

Fig. 3 Carotenoids from photosynthetic bacteria: (a) from anoxygenic phototrophs; (b) biosynthesis pathways in cyanobacteria stating from γ-carotene.

glycosylated 1′-OH derivative of chlorobactene, which together are constituents of the chlorosome and the photosynthetic RC (Takaichi, 1999; Chen *et al.*, 2020). The gene of phytoene synthase *crtB* and the gene forming the ϕ-ionone ring *crtU* are homologous to those of Actinobacteria (Sandmann, 2021a). However, a new lycopene cyclase CruA and a new way of phytoene desaturation is present in Chlorobi. The desaturases encoded by *crtP* and *crtQb* genes (Frigaard *et al.*, 2004) are different from *crtI*-desaturases in structure and catalytic action. Their evolution and function will be addressed in the section on cyanobacteria. Species of *Heliobacterium* with type I RC synthesize the completely different C_{30} carotenoid diaponeurosporene (structure in Fig. 2) (Takaichi *et al.*, 1997). Its synthesis starts with diapophytoene synthase encoded by *crtM*, which is highly homologous to the phytoene synthase gene *crtB*. This synthase condenses two molecules of C_{15} farnesyl pyrophosphate instead of two molecules of C_{20} geranylgeranyl pyrophosphate (Wieland *et al.*, 1994). The *crtM* gene further evolved to a squalene synthase gene (Sandmann, 2002), which is involved in the initiation of sterol synthesis in Proteobacteria (Wei *et al.*, 2016). Desaturation to diaponeurosporene is carried out by the *crtNa* gene product (Raisig *et al.*, 1996).

The type 1 and type 2 RCs are regarded as of a common origin (Sánchez-Baracaldo & Cardona, 2020). Nevertheless, phylogenetic analyses of the species above show that they are polyphyletic. A similar lack of relatedness to specific phylogenetic groups also holds for different carotenoid pathways, as pointed out in the section on lateral transfer of carotenoid biosynthesis genes.

2. Cyanobacterial oxygenic photosynthesis

In cyanobacteria, photosynthesis combines photosystem (PS) II with PSI, both similar to the different RCs of anoxygenic phototrophs. This culminated in the development of oxygenic photosynthesis as the most effective light-capturing process. The lower redox potential reached by the coupled photosystems drives a linear electron transport chain for the reduction of $NADP^+$ to NADPH in addition to formation of ATP. This generates both coenzymes necessary for sugar synthesis in the secondary reaction of photosynthesis. Electrons fed into this redox chain originate from water oxidation. The invention of this oxygen-evolving complex in Cyanobacteria using water instead of H_2S for photosynthesis created our oxygen atmosphere. Cyanobacterial photosynthesis

utilizes phycobiliproteins as light-harvesting peripheral antenna. As covalently protein-bound chromophores, linear tetrapyrroles transfer light energy to the reaction center chlorophylls. These water-soluble antenna are photoprotected by a family of orange carotenoid proteins that preferentially bind mono or diketo carotenoids (Kerfeld *et al.*, 2017). In contrast to the situation in anoxygenic photosynthesis, the carotenoids in both RCs are replaced by β-carotene, which accompanies Chl*a*, a modification of bacteriochlorophyll. Other carotenoids play no photoactive role in cyanobacterial photosynthesis.

VI. Carotenoid pattern and pathways in Cyanobacteria

As a generator of oxygen, cyanobacteria were forced to protect themselves from oxygenic conditions. An effective way of protection against reactive oxygen species is through carotenoids. These pigments have the potential to transfer radiation energy from pigments in the exited state and to quench $^1O_2$, transferring both molecules to the harmless ground state. Moreover, carotenoids are efficient scavengers of oxygen radicals formed in photosynthetic redox reactions (Sandmann, 2019). Depending on the carotenoid composition of cyanobacteria, there is a grouping between clades of filamentous cyanobacteria, in which in particular echinenone (ketolated at C4) together with canthaxanthin (diketolated at C4,4′) dominate, whereas zeaxanthin with two hydroxyl groups at C3,3′ is more typical for unicellular species (Takaichi & Mochimaru, 2007; Maresca *et al.*, 2008). Several other carotenoids (Fig. 3b) are zeaxanthin-derived nostoxanthin (2,3,2′,3′-tetrahydroxy-β-carotene), the monocyclic glycoside myxoxanthophyl and synechoxanthin with two χ-rings (Fig. 3b).

For carotenoid synthesis, cyanobacteria use genes already present in other bacteria but also acquired new more appropriate genes. Whereas phytoene synthesis in cyanobacteria is catalysed by the *crtB* gene product, which is ubiquitous for C_{40} and C_{50} carotenogenesis in all species, Cyanobacteria no longer possess *crtI* for desaturation but used a novel set of genes in contrast to all other bacteria other than Chlorobi. This includes two desaturases encoded by two genes, *crtP* for phytoene desaturase and *crtQb* for ζ-carotene desaturase (Sandmann & Vioque, 1999), in a poly-*cis* desaturation pathway (Breitenbach & Sandmann, 2005). The product of 15Z-phytoene desaturation is 9Z,15Z,9′Z-ζ-carotene (Fig. 4, left part). Before its further desaturation by CrtQb, the central double bond has to be isomerized to di-*cis* 9Z,9′Z-ζ-carotene by the gene product of *Z-ISO* (Sugiyama *et al.*, 2020). The resulting carotenoid of the second desaturation step is tetra-*cis* prolycopene (7Z,9Z,7Z′9Z′-lycopene). Before it can be cyclized it has to be isomerized into the all-*trans* form by CrtH (Breitenbach *et al.*, 2001; Masamoto *et al.*, 2001). The *crtP* and *crtQb* genes are highly homologous and may be a result of gene duplication. The *crtU* gene of β-carotene desaturase of Actinobacteria and Chlorobi shares similar regions especially at the C-terminus with them (Sandmann, 2002). The evolution of these novel desaturases can be comprehended from extant cyanobacteria (Fig. 4). *Gloeobacter*, a genus near the root of the Cyanobacteria lineage, is a relict in still using *crtI* (Steiger *et al.*, 2005; Tsuchiya *et al.*, 2005). In one strain, *Nostoc* 7120 (formerly *Anabaena*), *crtQa*, a modified *crtI* gene cloned by genetic complementation (Linden *et al.*, 1993), still retains the coding region and functionality for a desaturase, which can use different *cis* isomers of ζ-carotene as substrate (Albrecht *et al.*, 1996; Breitenbach *et al.*, 2013b). This gene can be regarded as a link to the prolycopene isomerase gene *crtH*. This gene resembles a *crtI*-related gene which lost both desaturase functions, solely maintaining the isomerase feature. Including *Z-ISO*, which is unrelated to *crtH*, four novel genes are necessary to replace the functionality of *crtI* in total. This raises the question of the functional relevance and the selective advantage of this complicated *crtI* gene replacement.

A decisive feature of CrtP and CrtQb is their use of oxidized plastoquinone (PQ) as a cofactor in the desaturation reactions (Schneider *et al.*, 1997; Breitenbach *et al.*, 2013). This connects phytoene and ζ-carotene desaturations to photosynthetic electron transport, as illustrated in Fig. 5. PSII is very sensitive to excess light and more susceptible to photodestruction than the other components of the photosynthetic electron transport chain including PSI. Therefore, under light-damaging conditions, the PQ pool becomes less reduced and as a consequence a better provision of oxidized PQ is available for carotene desaturation. Thus, the newly acquired set of genes for desaturation can be regarded as support for the transcriptionally upregulated carotenogenesis (Götz *et al.*, 1999; Albrecht *et al.*, 2001), as a part of short-term adaptation mechanisms necessary to cover the increased demand for carotenoid photoprotectants under high light.

According to the formation of individual combinations of carotenoids and the usage of different genes, three groups of cyanobacteria can be distinguished (Fig. 6): a filamentous group represented by *Nostoc/Anabaena*-related species, the *Synechococcus/Prochlorococcus*-clade of the phylogenetic tree (Moore *et al.*, 2019), which corresponds to the CrtL-clade of Sugiyama & Takaichi (2020), and a group of unicellular cyanobacteria of the CruA-clade. The next step in the pathway after desaturation is cyclization of lycopene to β-carotene, which is catalysed by the product of the genes *cruA* and *cruP*. An exception are species of the Crt-L clade in which *cruA* is missing (Sugiyama & Takaichi, 2020). Their CruP cyclase is responsible for the formation of γ-carotene with only one β-ionone ring which is the precursor of myxol and its glycoside myxoxanthophyll (Fig. 3b). The first specific modification in its synthesis is addition of water to the C1′,2′ double bond. This step is carried out by a novel hydratase encoded by *cruF*, which is unrelated to the hydratase gene *crtC* (Graham & Bryant, 2008). The next reaction is extension of the double bond system at C3′,4′ and C2′ hydroxylation. The *crtR*-dependent addition of a hydroxyl group to C3 of the β-ionone ring completes the synthesis of myxol. Remarkably, CrtR is unable to hydroxylate β-carotene simultaneously in species with canthaxanthin instead of zeaxanthin synthesis. Glycosylation at C2′ to myxoxanthophyll is carried out by the *cruG* protein (Graham & Bryant, 2008).

The carotenoid derived from β-carotene in the filamentous group is canthaxanthin produced by interaction of the *crtO*- with the *crtW*-encoded enzymes (Mochimaru *et al.*, 2005; Schöpf *et al.*, 2013). CrtW can also ketolate myxoxanthophyll. In addition to Cyanobacteria, *crtW* (Misawa *et al.*, 1995) is also common in

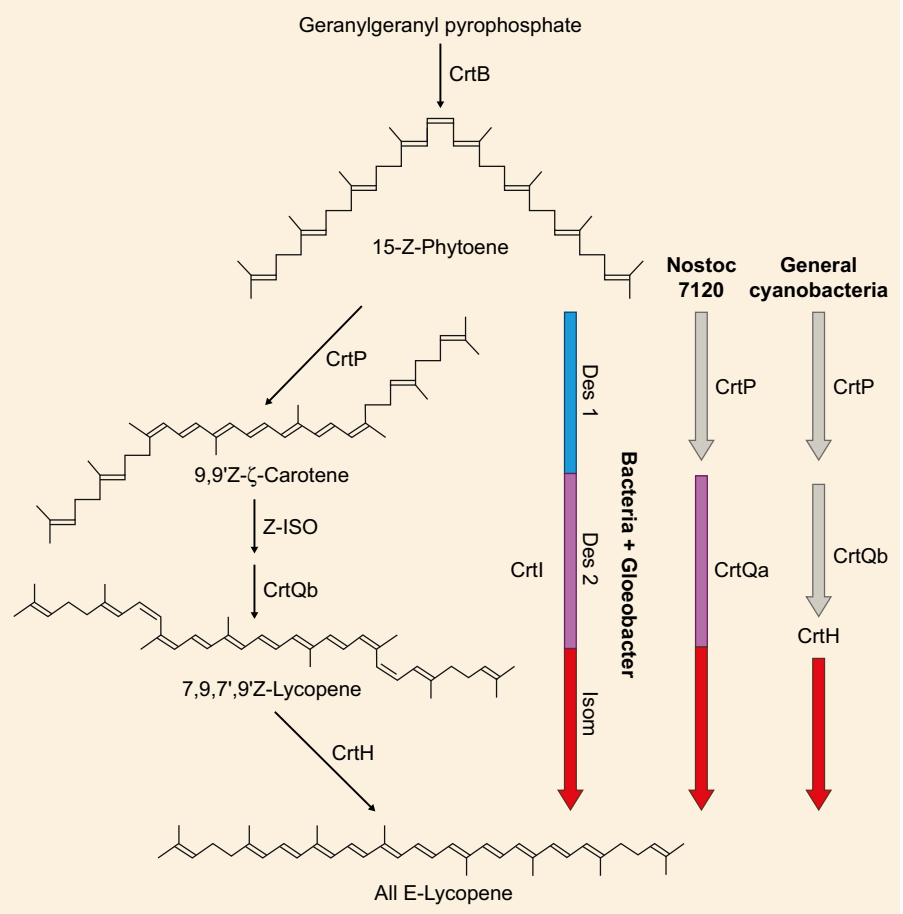

Fig. 4 Evolutionary replacement of CrtI-catalysed desaturation by a poly-*cis* desaturation pathway with *crtP* and *crtQb* desaturase genes in Cyanobacteria (left part). Different stages of *crtI* gene evolution to a ζ-carotene desaturase gene *crtQa* and further on to an isomerase gene *crtH*, and the stepwise takeover by *crtP* and *crtQb* monitored in different species (right part).

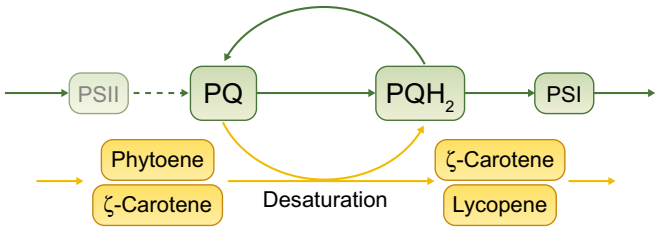

Fig. 5 Connection between photosynthetic electron transport and carotene desaturation. Competition for oxidized plastoquinone (PQ) as an electron acceptor for photosystem (PS) II and carotenoid desaturation, and its generation by reoxidation through PSI. Desaturation of phytoene (Phy) to ζ-carotene (ζ-Car) and further on to lycopene (Lyc). Lower PSII activity enhances the desaturation of carotenes.

different groups of Actinobacteria. Synechoxanthin (Fig. 3b), with two χ-rings and carboxylic C18 and C18′, is found in a range of filamentous and unicellular cyanobacteria except for species from the CrtL-clade (Cui *et al.*, 2020) The genes for synechoxanthin synthesis have been identified from *Synechococcus* 7002 and involve *cruE* for the formation of both aromatic χ-rings from β-carotene, and *cruH* encoding a C18 hydroxylase (Graham & Bryant, 2008). The CruE desaturase is homologous to CrtU. Functionally, they differ in the formation of aromatic rings with different positions of the methyl groups (compare ring structures in Fig. 1b). By analogy to phytoene desaturation via carbo cations (Fig. 2), the first desaturation yields a C3,4 double bond. Upon catalysis by CrtU, the second desaturation via a C2 carbo cation involves a charge stabilization by a C1,2 methyl rearrangement, which is also driven by complete ring desaturation. The difference in CruE catalysis is an additional C4,3 methyl rearrangement during the first desaturation step. Interestingly, small amounts of isorenieratene are formed in a reaction omitting the C4,3 methyl rearrangement during catalysis of CruE (Cui *et al.*, 2020).

In cyanobacteria that lack formation of canthaxanthin, CrtO catalyses mono ketolation of β-carotene to echinenone (Fernandez-Gonzalez *et al.*, 1997) and CrtR hydroxylation of β-carotene to zeaxanthin (Fig. 3b) (Masamoto *et al.*, 1998). A carotenoid present in a few cyanobacterial species is nostoxanthin (Takaichi & Mochimaru, 2007). Responsible for its synthesis is the *crtG* gene also found in Proteobacteria (Nishida *et al.*, 2005). Among different groups of bacteria, diverse β-ionone ring oxygenase genes exist. These are the C3 hydroxylases CrtY and CrtR, the C2 hydroxylase CrtG, and the C4 ketolase CrtW. They all are dioxygenases using oxygen in combination with a reductant in their oxygenation reaction (Fraser *et al.*, 1997) and possess in common three histidine-rich motifs for binding of a nonhaem di-iron cluster. In this respect, they all are evolutionarily related to membrane-bound fatty acid desaturases (Shanklin & Cahoon, 1998). In contrast to the CrtI-related hydroxylases that modify an allylic carbon, the fatty acid desaturase-related hydroxylases need

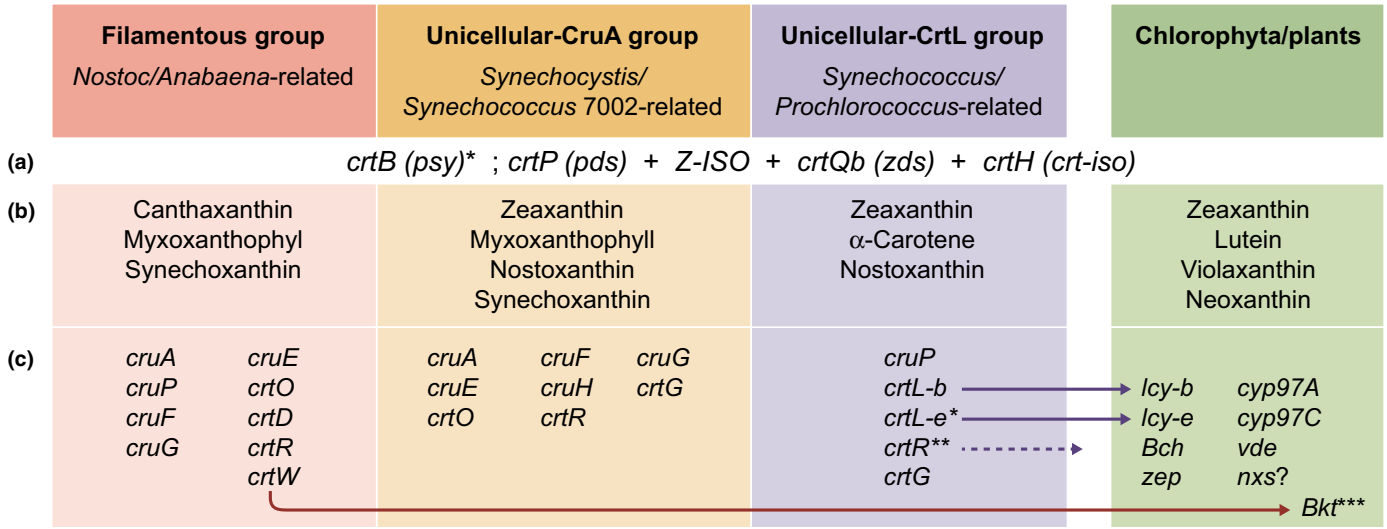

Fig. 6 Groupings of cyanobacteria according to their carotenoid composition and pathway gene and progression from *Synechococcus*/*Prochlorococcus*-type of the CrtL-clade to plants. (a) Common carotenogenic genes – the consensus nomenclature of prokaryotic carotenogenic genes starts 'crt' plus one letter and continuous with 'cru' plus one letter, eukaryotic genes were assigned by a three-letter code; (b) carotenoids; (c) individual pathway genes. *Only for *Prochlococcus*; **inherited to algal chloroplasts but lost in Chlorophytes; ***present only in a few unicellular chlorophytes.

reductants to drive the hydroxylation of a nonallylic carbon. The genes *crtW* and *crtR* share a common evolutionary relationship. However, they are much more distant to *crtZ*, as indicated by limited sequence similarities (Masamoto *et al.*, 1998). The whole pathway of *Synechocystis* (formerly *Aphanocapsa*) was established by *in vitro* labelling kinetics (Bramley & Sandmann, 1985).

The major carotenoid of the CrtL-clade cyanobacteria is zeaxanthin. For its synthesis, *crtL-b*, also present in Actinobacteria although *crtYb–crtYc* are the dominant lycopene cyclase genes in this phylum (Klassen, 2010), replaces the *cruA* cyclase gene in the cyclization to β-carotene (Maresca *et al.*, 2008). A second *crtL*-related cyclase gene, *crtL-b*, which is involved in the formation of α-carotene, coexists in *Prochlorococcus* species (Stickforth *et al.*, 2003).

In addition to cyanobacteria, zeaxanthin is synthesized in Archaea and in several groups of bacteria. It is notable that in Cyanobacteria, with the exception of *crtB*, the whole gene set for the zeaxanthin pathway is completely replaced by genes newly acquired or selected from established pathways in Protobacteria and Bacteroidetes. Comparing carotenoid composition and biosynthesis of Cyanobacteria with algae and especially green plant tissue, species from the CrtL-clade such as *Synechococcus* and *Prochlorococcus* are the best-matching extant examples.

VII. Carotenoids in algae and plants: from cyanobacteria to photosynthetic eukaryotes

1. Carotenoids in the plastid lineage

In photosynthetic eukaryotes, carotenoid biosynthesis is located in plastids which are of endosymbiotic origin (Keeling, 2013). A single-cell eukaryote engulfed an ancient unicellular cyanobacterium which was not digested but survived and developed endosymbiotic to a chloroplast. In the course of this transformation to a chloroplast as the photosynthesis factory, most genes were transferred to the host nucleus including all genes for carotenogenesis. Since carotenoid biosynthesis is carried out in the chloroplast, the nuclear encoded enzymes were extended with an N-terminal transit sequence for plastid import. It is generally accepted that all algae originated from an initial common endosymbiotic event (Reyes-Prieto *et al.*, 2007).

The evolutionary history of algal carotenoid biosynthesis reveals a complexity similar to Bacteria. Starting from the ancestral background, algae developed novel pathway modifications, most of them oxygenation reactions. Secondary endosymbiosis, by which algae originating from primary endosymbiosis were engulfed by a heterotroph, led to new groups of algae which are only distantly related to those from primary endosymbiosis and plants (Archibald & Keeling, 2002). The carotenoid biosynthesis of these algae will not be addressed here but details on their pathways to novel carotenoid structures is covered elsewhere (Takaichi, 2011; Sandmann, 2021b).

Three extant major algal lineages originating from primary endosymbiosis exist, the Glaucophyta, the Rhodophyta and the Chlorophyta. Closest to the root of the phylogeny tree of algae are the Glaucophyta (Keeling, 2004). Their carotenoid biosynthesis resembles the situation of a cyanobacterium with zeaxanthin as the major carotenoid along with its precursor β-carotene (Takaichi *et al.*, 2016). To date, carotenoid pathway genes from Glaucophyta have not been investigated. In addition to the carotenoids, these algae still retain the cyanobacterial phycobilisomes as peripheral antennae. Next in the phylogenetic lineage are the Rhodophyta with still existing phycobilisomes. Their carotenoid content varies within the different groups. Unicellular species, including the primordial Cyanidiophyceae, possess the same carotenoid composition of zeaxanthin with minor amounts of β-carotene as the Glaucophyta. An extended carotenoid distribution is found in filamentous and thalloid Rhodophyta. These groups synthesize α-carotene and in addition its 3,3′-dihydroxy derivative lutein. In the class Compsopogonophyceae, formation of violaxanthin, the

C5,6,5′,6′-diepoxide of zeaxanthin, is found (Schubert *et al.*, 2006; Takaichi *et al.*, 2016). However, the corresponding gene for epoxidation has not yet been identified. All genes involved in the pathway to zeaxanthin were identified in *Cyanidioschyzon merolae* (Cunningham *et al.*, 2007). They all reflect the ancestry of the pathway genes from CrtL-clade cyanobacteria. The gene for lutein-accumulating *Porphyra umbilicalis*, a cytochrome P450-type carotene hydroxylase from the CYP97B subfamily, was cloned and its function as a β-carotene hydroxylase confirmed (Yang *et al.*, 2014). Although new for the plastid lineage, another P450 hydroxylase involved in zeaxanthin emerged sporadically in the bacterium *Thermus thermophilus* but was not detected in any other species (Blasco *et al.*, 2004).

In Chlorophyta, at the end of the algal lineage, the cyanobacterial phycobilisome antenna system has been lost. It is replaced by a light-harvesting complex (LHC) that is evolutionarily novel but related to a zeaxanthin and β-carotene-containing LHC from the rhodophyte *Porphyridium cruentum* (Wolfe *et al.*, 1994). In this peripheral antenna system, carotenoids play a major role, switching reversibly either from light transmission to photo protection by energy dissipation or vice versa.

With the exception of the minor carotenoids loroxanthin and siphonaxanthin derived from lutein by Chlorophyta, carotenoid composition and the biosynthesis of Chlorophyta and plant chloroplasts is highly conserved (Takaichi, 2011). Therefore, biosynthesis of chloroplast carotenoids will be addressed in one context. An exclusive feature of chlorophytes from the class Chlorophyceae is the formation of highly photo protective astaxanthin (4,4′-diketozeaxanthin) under extreme stress conditions (Lemoine & Schoefs, 2010). This involves upregulation of the *bkt* gene (Huang *et al.*, 2006), which is an orthologue of *crtW* from cyanobacteria (Fig. 6c). However, this gene does not fit in an evolutionary line from cyanobacteria along the chloroplast lineage to Chlorophytes.

2. Customized carotenoids for light-harvesting

As shown in Fig. 7(a), the cyanobacterial pathway to zeaxanthin and α-carotene is extended in plants and their direct predecessors, the members of the Chlorophyta, by hydroxylation of α-carotene to lutein and epoxidation of zeaxanthin via antheraxanthin to violaxanthin and further modification to neoxanthin. All genes of the pathway to α-carotene and β-carotene evolved directly from the carotenogenic genes of a CrtL-clade cyanobacterium, including *lcy-e* which is derived from *crtL-e* of *Prochlorococcus*. This has been demonstrated for several chlorophytes (Lohr *et al.*, 2005; Wang *et al.*, 2018) and plants (Nisar *et al.*, 2015). Surprisingly, the β-carotene hydroxylase gene *bch* of Chlorophyta (Sun *et al.*, 1996) and plants is very poorly related to *crtR* although orthologues to this cyanobacterial gene exist in Rhodophyta. A possible explanation is the loss of the cyanobacterial-type zeaxanthin hydroxylase gene in the lineage from Rhodophyta to Chlorophyta and replacement by a more closely related bacterial *crtZ* gene by horizontal transfer (Cui *et al.*, 2013), which is regarded as an important evolutionary factor in chloroplast development (Keeling & Palmer, 2008). Two other hydroxylase genes are P450 monooxygenases, which are necessary for the synthesis of lutein (Kim *et al.*, 2009). The gene *cyp97b* is preferentially involved in hydroxylation of the β-ionone ring of α-carotene and resembles the P450 gene from Rhodophyta (Yang *et al.*, 2014). The second P450 hydroxylase gene, *cyp97c*, is new in the chloroplast lineage and specific for ε-ionone ring hydroxylation. It appears to have evolved from an ancestral gene of the rhodophycean type by duplication and functional divergence. Other newly acquired genes are *zep* for the epoxidation of zeaxanthin to violaxanthin (Marin *et al.*, 1996) and *vde* (Bugos & Yamamoto, 1996) for the back deepoxidation reaction from violaxanthin to zeaxanthin (Fig. 7a). Both enzymes are members of the lipocalin family, which bind small lipophilic molecules (Bugos *et al.*, 1998). The nature of the *nsy* gene for the conversion of violaxanthin to neoxanthin is still open and has not be conclusively identified (Neuman *et al.*, 2014). Apart from chloroplasts, plants may contain other types of nonphotosynthetic plastids including chromoplasts (Sun *et al.*, 2018). They are present in flowers and fruit and sequester carotenoids. The modifications to these so-called secondary carotenoids will not be covered here.

All carotenoids mentioned above play a specific functional role in plant photosynthesis. The cyanobacterial principle of associating chlorophylls of photo systems with β-carotene for photo protection is retained. Similar to cyanobacteria and algae, β-carotene is a structural element of the core photosynthesis complexes of plants (Gao *et al.*, 2018). By contrast, the newly available oxygenated carotenoids, the xanthophylls, are used for the improvement of light-harvesting. Apart from the minor carotenoid containing LHCs, the major novel LHC-II of PSII possess two molecules of lutein integrated into its centre for stabilization of the antenna proteins and quenching of ^{3}Chl (Jahns & Holzwarth, 2012) in addition to one 9′-*cis* neoxanthin and one xanthophyll cycle carotenoid (Liu *et al.*, 2004).

Zeaxanthin and violaxanthin together with antheraxanthin are the xanthophyll cycle carotenoids with different S1 states (Young & Frank, 1996). From violaxanthin with a higher energy level, absorbed light energy can be transferred to Chl (Fig. 7b). Under light-saturated conditions with high proton flux into the lumen, violaxanthin de-epoxidase is activated at pH < 6.2 (Jahns *et al.*, 2009). Then, the generated de-epoxidized zeaxanthin (Fig. 7a) with its lower S1 state participates in energy absorption from ^{1}Chl. This prevents the formation of harmful triplet Chl and subsequently the formation highly oxidative $^1O_2$.

For the replacement of the phycobilisomes for light-harvesting at the end of the plastid lineages, a pigment-protein complex evolved in which the violaxanthin–zeaxanthin pair acts as a valve to regulate the radiation energy flow to the photosystem by feedback control from the electron transport chain. This new structure of the LHCs was possible by coevolution of genes for novel carotenoids with structural proteins of this complex (Dall'Osto *et al.*, 2015).

VIII. Conclusion

Photosynthesis progressed to oxygenic photosynthesis with two coupled photo systems resulting in higher energy conservation. This relates not only to the evolution of structural proteins but also to metabolic pathways of essential cofactors that were

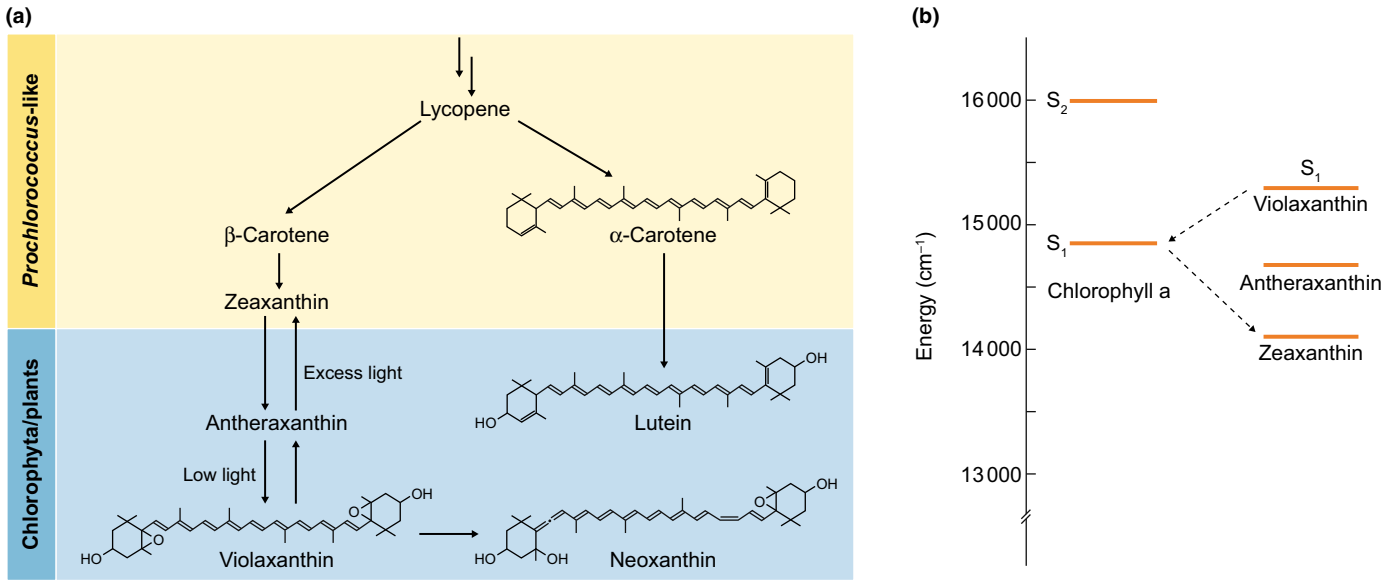

Fig. 7 From zeaxanthin and α-carotene of cyanobacteria to the carotenoids in the light harvesting complex of green chloroplasts. (a) Regulation of photoprotection vs light-harvesting; (b) S1-state energy levels and transfer between xanthophyll cycle carotenoids and Chla.

necessary for a functional photosynthetic apparatus. Indispensable for a steady improvement of photosynthesis was the coevolution of carotenoid synthesis to provide appropriate carotenoids at each stage of the evolution of photosynthesis. In particular, bacteria offered a pool of diverse carotenoid structures that could be selected according to demand. A crucial factor for pathway modifications was horizontal gene transfer and the evolution of a gene family of heterofunctional homologues from a common ancient origin.

It is generally difficult to understand the full history of evolutionary lineages of a pathway from extant species representing the tip of an evolutionary process which is mostly hidden in the past. Looking at the phylogeny of the *crtB* gene with orthologues in all carotenogenic organisms, it is evident that there is no conformity with the phylogeny of species (Klassen, 2010). Furthermore, gene transfer between distantly related species complicates our understanding of ancestor–descendent relationships.

Ancient cyanobacteria generated our oxygen atmosphere as a side effect of water splitting in the process of photosynthesis. Therefore, the individual cyanobacterial groups had to adapt their carotenoid inventory to the aerobic environment in order to protect their photosystems from oxidation by selection of genes from other bacteria and evolution of novel genes. This optimization of favourable carotenoids for photosynthesis continued after endosymbiosis along the chloroplast lineage. It finally culminated in the synthesis of specialized carotenoids for the light-harvesting systems in plants as highly effective antennae which replaced the phycobilisomes.

During the evolution of carotenoids, their functionality changed fundamentally. In Archaea, they act as a membrane reinforcer, but in bacteria they are replaced by other terpenoids in this function and changed to cellular photoprotectants. With the emerging photosynthetic organisms, carotenoids became important for light-harvesting and essential for the protection of chlorophylls by quenching their triplet state. Looking at the carotenoids associated with the antenna systems and RCs of different photosynthetic bacteria, it can be concluded that there is no selectivity for a particular structure among species. The importance of carotenoids grew with the emerging oxygen atmosphere and drove the evolution towards the sophisticated LHCs of plants with a novel functional set of carotenoids. Among them, the reversibly interchanging violaxanthin–zeaxanthin pair resembles either an antenna by increasing the rate of energy input via singlet state energy transfer to the RC or an energy drainage by deexcitation of singlet Chl, respectively, depending on the light conditions.

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References

Albrecht M, Linden H, Sandmann G. 1996. Biochemical characterization of purified ζ-carotene desaturase from *Anabaena* PCC 7120 after expression in *Escherichia coli*. *European Journal of Biochemistry* **236**: 115–120.

Albrecht M, Steiger S, Sandmann G. 2001. Expression of a ketolase gene mediates the synthesis of canthaxanthin in *Synechococcus* leading to resistance against pigment photodegradation and UV-B sensitivity of photosynthesis. *Photochemistry and Photobiology* **73**: 551–555.

Andersson O, Cogdell RJ, Gillbro T. 1996. Femtosecond dynamics of carotenoid-to-bacteriochlorophyll a energy transfer in the light-harvesting antenna complexes from the purple bacterium *Chromatium purpuratum*. *Chemical Physics* **210**: 195–217.

Archibald M, Keeling PJ. 2002. Recycled plastids: a 'green movement' in eukaryotic evolution. *Trends in Genetics* **18**: 577–584.

Armstrong GA, Alberti M, Leach F, Hearst JE. 1989. Nucleotide sequence, organization, and nature of the protein products of the carotenoid biosynthesis gene cluster of *Rhodobacter capsulatus*. *Molecular and General Genetics* **216**: 254–268.

Blankenship R. 2002. *Molecular mechanisms of photosynthesis*. London, UK: Blackwell Science.

Blasco F, Kauffmann I, Schmid RD. 2004. CYP175A1 from *Thermus thermophilus* HB27, the first beta-carotene hydroxylase of the P450 superfamily. *Applied and Microbial Biotechnology* **64**: 671–674.

Bramley PM, Sandmann G. 1985. *In vitro* and *in vivo* biosynthesis of xanthophylls by the cyanobacterium *Aphanocapsa*. *Phytochemistry* **24**: 2919–2922.

Breitenbach J, Bruns M, Sandmann G. 2013b. Contribution of two ζ-carotene desaturases to the poly-*cis* desaturation pathway in the cyanobacterium *Nostoc* PCC 7120. *Archives of Microbiology* **195**: 491–498.

Breitenbach J, Fraser PD, Sandmann G. 2012. Carotenoid synthesis and phytoene synthase activity during mating of *Blakeslea trispora*. *Phytochemistry* **76**: 40–45.

Breitenbach J, Gerjets T, Sandmann G. 2013a. Catalytic properties and reaction mechanism of the CrtO carotenoid ketolase from the cyanobacterium *Synechocystis* sp. PCC 6803. *Archives of Biochemistry and Biophysics* **529**: 86–91.

Breitenbach J, Sandmann G. 2005. ζ-Carotene *cis* isomers as products and substrates in the plant poly-cis carotenoid biosynthetic pathway to lycopene. *Planta* **220**: 785–793.

Breitenbach J, Vioque A, Sandmann G. 2001. Gene sll0033 from *Synechocystis* 6803 encodes a carotene isomerase involved in the biosynthesis of all-E lycopene. *Zeitschrift für Naturforschung C* **56**: 915–917.

Britton G. 1995. Structures and properties of carotenoids in relation to function. *FASEB Journal* **9**: 1551–1558.

Britton G, Liaaen-Jensen S, Pfander H. 2004. *Carotenoids Handbook*. Basel, Switzerland: Birkhäuser Verlag.

Bugos RC, Hieber AD, Yamamoto HY. 1998. Xanthophyll cycles enzymes are members of the lipocalin family, the first identified from plants. *Journal of Biological Chemistry* **273**: 15321–15324.

Bugos RC, Yamamoto HY. 1996. Molecular cloning of violaxanthin de-epoxidase from romaine lettuce and expression in *Escherichia coli*. *Proceedings of the National Academy of Science, USA* **93**: 6320–6325.

Chen JH, Wu H, Xu C, Liu XC, Huang Z, Chang S, Wang W, Han G, Kuang T, Shen JR *et al.* 2020. Architecture of the photosynthetic complex from a green sulfur bacterium. *Science* **370**: 6350.

Cui H, Wang Y, Qin S. 2013. Molecular evolution of lycopene cyclases involved in the formation of carotenoids in eukaryotic algae. *Plant Molecular Biology Reporter* **29**: 1013–1020.

Cui X, Liu XL, Shen G, Ma J, Husain F, Rocher D, Zumberg JE, Bryant DA, Summons RE. 2020. Niche expansion for phototrophic sulfur bacteria at the Proterozoic–Phanerozoic transition. *Proceedings of the National Academy of Science, USA* **117**: 17599–17606.

Cunningham FX, Lee H, Gantt E. 2007. Carotenoid biosynthesis in the primitive red alga *Cyanidioschyzon merolae*. *Eukaryotic Cell* **6**: 533–545.

Dall'Osto L, Bressan M, Bassi R. 2015. Biogenesis of light harvesting proteins. *Biochimica et Biophysica Acta* **1847**: 861–871.

Ermler U, Michel H, Schiffer M. 1994. Structure and function of the photosynthetic reaction center from *Rhodobacter sphaeroides*. *Journal of Bioenergetics and Biomembranes* **26**: 5–15.

Fernandez-Gonzalez B, Sandmann G, Vioque A. 1997. A new type of asymmetrically acting β-carotene ketolase is required for the synthesis of echinenone in the cyanobacterium *Synechocystis* sp. PCC6803. *Journal of Biological Chemistry* **272**: 9728–9733.

Fraser PD, Misawa N, Linden H, Yamano S, Kobayashi K, Sandmann G. 1992. Expression in *Escherichia coli*, purification and reactivation of the recombinant *Erwinia uredovora* phytoene desaturase. *Journal of Biological Chemistry* **267**: 19891–19895.

Fraser PD, Miura Y, Misawa N. 1997. *In vitro* characterization of astaxanthin biosynthetic enzymes. *Journal of Biological Chemistry* **272**: 6128–6135.

Frigaard NU, Maresca JA, Yunker CE, Jones AD, Bryant DA. 2004. Genetic manipulation of carotenoid biosynthesis in the green sulfur bacterium *Chlorobium tepidum*. *Journal of Bacteriology* **186**: 5210–5220.

Gao J, Wang H, Yuan Q, Feng Y. 2018. Structure and function of the photosystem supercomplexes. *Frontiers in Plant Science* **9**: 357.

Gerjets T, Steiger S, Sandmann G. 2009. Catalytic properties of the expressed acyclic carotenoid 2-ketolases from *Rhodobacter capsulatus* and *Rubrivivax gelatinosus*. *Biochimica et Biophysica Acta* **1791**: 125–131.

Götz T, Windhövel U, Böger P, Sandmann G. 1999. Protection of photosynthesis against UV-B radiation by carotenoids in transformants of the cyanobacterium *Synechococcus* PCC7942. *Plant Physiology* **120**: 599–604.

Graham JE, Bryant DA. 2008. The biosynthetic pathway for synechoxanthin, an aromatic carotenoid synthesized by the euryhaline, unicellular cyanobacterium *Synechococcus* sp. strain PCC 7002. *Journal of Bacteriology* **190**: 7966–7974.

Gruszeckia WI, Strzayka K. 2005. Carotenoids as modulators of lipid membrane physical properties. *Biochimica et Biophysica Acta* **1740**: 108–115.

Harada J, Nagashima KV, Takaichi S, Misawa N, Matsuura K, Shimada K. 2001. Phytoene desaturase, CrtI, of the purple photosynthetic bacterium, *Rubrivivax gelatinosus*, produces both neurosporene and lycopene. *Plant and Cell Physiology* **42**: 1112–1118.

Hausmann A, Sandmann G. 2000. A single 5-step desaturase is involved in the carotenoid biosynthesis pathway to β-carotene and torulene in *Neurospora crassa*. *Fungal General Biology* **30**: 147–153.

Hemmi H, Ikejiri S, Nakayama T, Nishino T. 2003. Fusion-type lycopene β-cyclase from a thermoacidophilic archaeon *Sulfolobus solfataricus*. *Biochemical and Biophysical Research Communications* **305**: 586–591.

Henderson R. 1977. The purple membrane from *Halobacterium halobium*. *Annual Reviews of Biophysics and Bioenergetics* **6**: 87–109.

Hornero-Mendez D, Britton G. 2002. Involvement of NADPH in the cyclization reaction of carotenoid biosynthesis. *FEBS Letters* **515**: 133–136.

Huang JC, Chen F, Sandmann G. 2006. Stress-related differential expression of multiple β-carotene ketolase genes in the unicellular green alga *Haematococcus pluvialis*. *Journal of Biotechnology* **122**: 176–185.

Igarashi N, Harada J, Nagashima S, Matsuura K, Shimada K, Nagashima KVP. 2001. Horizontal transfer of the photosynthesis gene cluster and operon rearrangement in purple bacteria. *Journal of Molecular Evolution* **52**: 333–341.

Jahns P, Holzwarth AR. 2012. The role of the xanthophyll cycle and of lutein in photoprotection of photosystem II. *Biochimica et Biophysica Acta* **1817**: 182–193.

Jahns P, Latowski D, Strzalka K. 2009. Mechanism and regulation of the violaxanthin cycle: the role of antenna proteins and membrane lipids. *Biochimica et Biophysica Acta* **1787**: 3–14.

Kandori H. 2015. Ion-pumping microbial rhodopsins. *Frontiers in Molecular Biosciences* **2**: 52.

Keeling PJ. 2004. Diversity and evolutionary history of plastids and their hosts. *American Journal of Botany* **91**: 1481–1493.

Keeling PJ. 2013. The number, speed, and impact of plastid endosymbioses in eukaryotic evolution. *Annual Reviews of Plant Biology* **64**: 583–607.

Keeling PJ, Palmer JD. 2008. Horizontal gene transfer in eukaryotic evolution. *Nature Reviews Genetics* **9**: 605–618.

Kerfeld CA, Melnicki MR, SutterM D-M. 2017. Structure, function and evolution of the cyanobacterial orange carotenoid protein and its homologs. *New Phytologist* **215**: 937–951.

Kim J, Smith JJ, Tian L, DellaPenna D. 2009. The evolution and function of carotenoid hydroxylases in *Arabidopsis*. *Plant and Cell Physiology* **50**: 463–479.

Klassen JL. 2009. Pathway evolution by horizontal transfer and positive selection is accommodated by relaxed negative selection upon upstream pathway genes in purple bacterial carotenoid biosynthesis. *Journal of Bacteriology* **191**: 7500–7508.

Klassen JL. 2010. Phylogenetic and evolutionary patterns in microbial carotenoid biosynthesis are revealed by comparative genomics. *PLoS ONE* **5**: e11257.

Krubasik P, Kobayashi M, Sandmann G. 2001. Expression and functional analysis of a gene cluster involved in the synthesis of decaprenoxanthin reveals the mechanisms for C50 carotenoid formation. *European Journal of Biochemistry* **268**: 3702–3708.

Krubasik P, Sandmann G. 2000a. Molecular evolution of lycopene cyclases involved in the formation of carotenoids with ionone end groups. *Biochemical Society Transactions* **28**: 806–810.

Krubasik P, Sandmann G. 2000b. A carotenogenic gene cluster from *Brevibacterium linens* with novel lycopene cyclase genes involved in the synthesis of aromatic carotenoids. *Molecular and General Genetics* **263**: 423–432.

Krügel H, Krubasik P, Weber K, Saluz HP, Sandmann G. 1999. Functional analysis of genes from *Streptomyces griseus* involved in the synthesis of isorenieratene, a carotenoid with aromatic end groups, revealed a novel type of carotenoid desaturase. *Biochimica et Biophysica Acta* **1439**: 57–64.

Kull DR, Pfander H. 1997. Isolation and structure elucidation of carotenoid glycosides from the thermoacidophilic Archaea *Sulfolobus shibatae*. *Journal of Natural Products* **60**: 371–374.

Lazrak T, Wolff G, Albrecht M, Nakatani Y, Ourisson G, Kates M. 1988. Bacterioruberins reinforce reconstituted *Halobacterium* lipid memebranes. *Biochimica et Biophysica Acta* **939**: 160–162.

Lemoine Y, Schoefs B. 2010. Secondary ketocarotenoid astaxanthin biosynthesis in algae: a multifunctional response to stress. *Photosynthesis Research* **106**: 155–177.

Linden H, Vioque A, Sandmann G. 1993. Isolation of a carotenoid biosynthesis gene coding for a ζ-carotene desaturase from *Anabaena* PCC7120 by heterologous complementation. *FEMS Microbiology Letters* **106**: 99–104.

Liu Z, Yan H, Wang K, Kuang T, Zhang J, Gui L, An X, Chang W. 2004. Crystal structure of spinach major light-harvesting complex at 2.72 Å resolution. *Nature* **428**: 287–292.

Lohr M, Im C-S, Grossman AR. 2005. Genome-based examination of chlorophyll and carotenoid biosynthesis in *Chlamydomonas reinhardtii*. *Plant Physiology* **138**: 490–515.

Maresca JA, Graham JE, Bryant DA. 2008. Carotenoid biosynthesis in chlorophototrophs: the biochemical and genetic basis for structural diversity. *Photosynthesis Research* **97**: 121–140.

Maresca JA, Graham JE, Wu M, Eisen JA, Bryant DA. 2007. Identification of a fourth family of lycopene cyclases in photosynthetic bacteria. *Proceedings of the National Academy of Science, USA* **104**: 11784–11789.

Marin E, Nussaume L, Quesada A, Gonneau M, Sotta B, Hugueney P, Frey A, Marion-Poll A. 1996. Molecular identification of zeaxanthin epoxidase of *Nicotiana plumbaginifolia*, a gene involved in abscisic acid biosynthesis and corresponding to the ABA locus of *Arabidopsis thaliana*. *EMBO Journal* **15**: 2331–2342.

Masamoto K, Misawa N, Kaneko T, Kikuno R, Toh H. 1998. Beta-carotene hydroxylase gene from the cyanobacterium *Synechocystis* sp. PCC6803. *Plant and Cell Physiology* **39**: 560–564.

Masamoto K, Wada H, Kaneko T, Takaichi S. 2001. Identification of a gene required for *cis*-to-*trans* carotene isomerization in carotenogenesis of the cyanobacterium *Synechocystis* sp PCC 6803. *Plant and Cell Physiology* **42**: 1398–1402.

Misawa N, Nakagawa M, Kobayashi K, Yamano S, Izawa Y, Nakamura K, Harashima K. 1990. Elucidation of the *Erwinia uredovora* carotenoid biosynthetic pathway by functional analysis of gene products expressed in *Escherichia coli*. *Journal of Bacteriology* **172**: 6704–6712.

Misawa N, Satomi Y, Kondo K, Yokoyama A, Kajiwara S, Saito T, Ohtani T, Miki W. 1995. Structure and functional analysis of a marine bacterial carotenoid biosynthesis gene cluster and astaxanthin biosynthetic pathway proposed at the gene level. *Journal of Bacteriology* **177**: 6575–6584.

Mochimaru M, Masukawa H, Takaichi S. 2005. The cyanobacterium *Anabaena* sp. PCC 7120 has two distinct β-carotene ketolases: CrtO for echinenone and CrtW for ketomyxol synthesis. *FEBS Letters* **579**: 6111–6114.

Moore KR, Magnabosco C, Momper L, Gold DA, Bosak T, Fournier GP. 2019. An expanded ribosomal phylogeny of cyanobacteria supports a deep placement of plastids. *Frontiers of Microbiology* **10**: 1612.

Netzer R, Stafsnes MH, Andreassen T, Goksoyr A, Bruheim P, Brautaset T. 2010. Biosynthetic pathway for gamma-cyclic sarcinaxanthin in Micrococcus *luteus*: heterologous expression and evidence for diverse and multiple catalytic functions of C_{50} carotenoid cyclases. *Journal of Bacteriology* **192**: 5688–5699.

Neuman H, Galpaz N, Cunningham FX, Zamir D, Hirschberg J. 2014. The tomato mutation nxd1 reveals a gene necessary for neoxanthin biosynthesis and demonstrates that violaxanthin is a sufficient precursor for abscisic acid biosynthesis. *The Plant Journal* **78**: 80–93.

Nisar N, Li L, Lu S, Khin NC, Pogson BJ. 2015. Carotenoid metabolism in pants. *Molecular Plant* **8**: 68–82.

Nishida Y, Adachi K, Kasai H, Shizuri Y, Shindo K, Sawabe A, Komemushi S, Miki W, Misawa N. 2005. Elucidation of a carotenoid biosynthesis gene cluster encoding a novel enzyme, 2,2'-beta-hydroxylase, from *Brevundimonas* sp. strain SD212 and combinatorial biosynthesis of new or rare xanthophylls. *Applied and Environmental Microbiology* **71**: 4286–4296.

Oesterhelt D. 1976. Bacteriorhodopsin as an example of a light-driven proton pump. *Angewandte Chemie International Edition* **15**: 17–24.

Peck RF, Johnson EA, Krebs MP. 2002. Identification of a lycopene beta-cyclase required for bacteriorhodopsin biogenesis in the archaeon *Halobacterium salinarum*. *Journal of Bacteriology* **184**: 2889–2897.

Petersen J, Brinkmann H, Bunk B, Michael V, Päuker O, Pradella S. 2012. Think pink: photosynthesis, plasmids and the *Roseobacter* clade. *Environmental Microbiology* **14**: 2661–2672.

Phadwal K. 2005. Carotenoid biosynthetic pathway: molecular phylogenies and evolutionary behavior of *crt* genes in eubacteria. *Gene* **345**: 35–43.

Rählert N, Sandmann G. 2009. A *crtA*-related gene from *Flavobacterium* P99–3 encodes a novel carotenoid 2-hydroxylase involved in myxol biosynthesis. *FEBS Letters* **583**: 1605–1610.

Raisig A, Bartley G, Scolnik P, Sandmann G. 1996. Purification in an active state and properties of the 3-step phytoene desaturase from *Rhodobacter capsulatus* after overexpression in *Escherichia coli*. *Journal of Biochemistry* **119**: 559–564.

Raisig A, Sandmann G. 1999. 4,4′-Diapophytoene desaturase: catalytic properties of an enzyme from the C-30 carotenoid pathway of *Staphylococcus aureaus*. *Journal of Bacteriology* **181**: 6184–6187.

Raisig A, Sandmann G. 2001. Functional properties of diapophytoene and related desaturases of C_{30} and C_{40} carotenoid biosynthetic pathways. *Biochimica et Biophysica Acta* **1533**: 164–170.

Reyes-Prieto A, Weber APM, Bhattacharya D. 2007. The origin and establishment of the plastid in algae and plants. *Annual Reviews of Genetics* **41**: 147–168.

Rohmer M, Bouvier P, Ourisson G. 1979. Molecular evolution of biomembranes: structural equivalents and phylogenetic precursors of sterols. *Proceedings of the National Academy of Science, USA* **76**: 847–851.

Ruban AV. 2015. Evolution under the sun: optimizing light harvesting in photosynthesis. *Journal of Experimental Botany* **66**: 7–23.

Sánchez-Baracaldo P, Cardona T. 2020. On the origin of oxygenic photosynthesis and cyanobacteria. *New Phytologist* **225**: 1440–1446.

Sandmann G. 2002. Molecular evolution of carotenoid biosynthesis from bacteria to plants. *Physiologia Plantarum* **116**: 431–440.

Sandmann G. 2009. Evolution of carotene desaturation: the complication of a simple pathway. *Archives of Biochemistry and Biophysics* **483**: 169–174.

Sandmann G. 2019. Antioxidant protection from UV- and light-stress related to carotenoid structures. *Antioxidants* **8**: E219.

Sandmann G. 2021a. Carotenoid biosynthesis in the phylum Actinobacteria. In: Misawa N, ed. *Carotenoids, advances in experimental medicine and biology*. Singapore: Springer Nature, 175–181.

Sandmann G. 2021b. Diversity and evolution of carotenoid biosynthesis from prokaryotes to plants. In: Misawa N, ed. *Carotenoids, advances in experimental medicine and biology*. Singapore: Springer Nature, 79–94.

Sandmann G, Misawa N. 1992. New functional assignment of the carotenogenic genes *crtB* and *crtE* with constructs of these genes from *Erwinia* species. *FEMS Microbiology Letters* **90**: 253–258.

Sandmann G, Vioque A. 1999. Phylogeny of carotenogenic genes and enzymes in cyanobacteria. In: Pescheck GA, Löffelhardt W, Schmetterer G, eds. *The phototrophic porkaryotes*. New York, NY, USA: Kluwer Academic, 799–803.

Sandmann G, Yukawa H. 2005. Vitamin synthesis: carotenoids, biotin and pantothenate. In: Eggeling J, Bott M, eds. *Handbook of Corynebacterium glutamicum*. Boca Raton, FL, USA: Taylor & Francis, 397–415.

Schaub P, Yu Q, Gemmecker S, Poussin-Courmontagne P, Mailliot J, McEwen AG, Ghisla S, Al-Babili S, Cavarelli J, Beyer P. 2012. On the structure and function of the phytoene desaturase CRTI from *Pantoea ananatis*, a membrane-peripheral and FAD-dependent oxidase/isomerase. *PLoS ONE* 7: e39550.

Schneider C, Böger P, Sandmann G. 1997. Phytoene desaturase: heterologous expression in an active state, purification, and biochemical properties. *Protein Expression and Purification* **10**: 175–179.

Schnurr G, Misawa N, Sandmann G. 1996. Expression, purification and properties of lycopene cyclase from *Erwinia uredovora*. *Biochemical Journal* **315**: 869–874.

Schöpf L, Mautz J, Sandmann G. 2013. Multiple ketolases involved in light regulation of canthaxanthin biosynthesis in *Nostoc punctiforme* PCC73102. *Planta* **237**: 1279–1285.

Schubert N, Garcia-Mendoza E, Pacheco-Ruiz I. 2006. Carotenoid composition of marine red algae. *Journal of Phycology* **42**: 1208–1216.

Shanklin J, Cahoon EB. 1998. Desaturation and related modifications of fatty acids. *Annual Reviews of Plant Physiology and Plant Molecular Biology* **49**: 611–641.

Šlouf V, Chábera P, Olsen JD, Martin EC, Qian P, Hunter CN, Polívka T. 2012. Photoprotection in a purple phototrophic bacterium mediated by oxygen-

dependent alteration of carotenoid excited-state properties. *Proceedings of the National Academy of Science, USA* **109**: 8570–8575.

Steiger S, Astier C, Sandmann G. 2000. Substrate specificity of the expressed 3,4-carotenoid desaturase from *Rubrivivax gelatinosus* reveals the detailed reaction sequence to spheroidene and spirilloxanthin. *Biochemical Journal* **349**: 635–640.

Steiger S, Jackisch Y, Sandmann G. 2005. Carotenoid biosynthesis in *Gloeobacter violaceus* PCC4721 involves a single *crtI*-type phytoene desaturase instead of typical cyanobacterial enzymes. *Archives of Microbiology* **103**: 207–214.

Steiger S, Perez-Fons L, Cutting SM, Fraser PD, Sandmann G. 2015. Annotation and functional assignment of the genes for the C_{30} carotenoid pathways from the genomes of two bacteria *Bacillus indicus* and *Bacillus firmus*. *Microbiology* **161**: 194–202.

Stickforth P, Sandmann G. 2007. Kinetic variations determine the product pattern of phytoene desaturase from *Rubrivivax gelatinosus*. *Archives of Biochemistry and Biophysics* **461**: 235–241.

Stickforth P, Sandmann G. 2011. Structural and kinetics properties of a mutated phytoene desaturase from *Rubrivivax gelatinosus* with modified product specificity. *Archives of Biochemistry and Biophysics* **505**: 118–122.

Stickforth P, Steiger S, Hess WR, Sandmann G. 2003. A novel type of lycopene ε-cyclase in the marine cyanobacterium *Prochlorococcus marinus* MED4. *Archives of Microbiology* **179**: 409–415.

Sugiyama K, Takahashi K, Nakazawa K, Yamada M, Kato S, Shinomura T, Nagashima Y, Suzuki H, Ara T, Harada J *et al.* 2020. Oxygenic phototrophs need ζ-carotene isomerase (Z-ISO) for carotene synthesis: functional analysis in *Arthrospira* and *Euglena*. *Plant and Cell Physiology* **61**: 276–282.

Sugiyama K, Takaichi S. 2020. Carotenogenesis in cyanobacteria: CruA/CruP-type and CrtL-type lycopene cyclases. *Journal of General and Applied Microbiology* **66**: 53–58.

Sun T, Yuan H, Cao H, Yazdani M, Tadmor Y, Li L. 2018. Carotenoid metabolism in plants: the role of plastids. *Molecular Plant* **11**: 58–74.

Sun Z, Gantt E, Cunningham FX. 1996. Cloning and functional analysis of the β-carotene hydroxylase of *Arabidopsis thaliana*. *Journal of Biological Chemistry* **271**: 24349–24352.

Takaichi S. 1999. Carotenoids and carotenogenesis in anoxygenic photosynthetic bacteria. In: Frank HA, Young AJ, Britton G, Cogdell RJ, eds. *The photochemistry of carotenoids*. New York, NY, USA: Kluwer Academic, 39–69.

Takaichi S. 2011. Carotenoids in algae: distributions, biosynthesis and functions. *Marine Drugs* **9**: 1101–1118.

Takaichi S, Inoue K, Akaike M, Kobayashi M, Oh-oka H, Madigan MT. 1997. The major carotenoid in all known species of heliobacteria is the C_{30} carotenoid 4,4′-diaponeurosporene, not neurosporene. *Archives of Microbiology* **168**: 277–281.

Takaichi S, Mochimaru M. 2007. Carotenoids and carotenogenesis in cyanobacteria: unique ketocarotenoids and carotenoid glycosides. *Cellular and Molecular Life Sciences* **64**: 2607–2619.

Takaichi S, Yokoyama A, Mochimaru M, Uchida H, Murakami A. 2016. Carotenogenesis diversification in phylogenetic lineages of Rhodophyta. *Journal of Phycology* **52**: 329–338.

Tao L, Picataggio S, RouviérePE CQ. 2004. Asymmetrically acting lycopene cyclases (CrtLm) from non-photosynthetic bacteria. *Molecular and General Genomics* **271**: 180–188.

Tao L, Schenzl A, Odom JM, Cheng Q. 2005. Novel carotenoid oxidase involved in biosynthesis of 4,4′-diapolycopene dialdehyde. *Applied and Environmental Microbiology* **71**: 3294–3301.

Tao L, Yao H, Cheng Q. 2007. Genes from a *Dietzia* sp. for synthesis of C40 and C50 beta-cyclic carotenoids. *Gene* **386**: 90–97.

Teramoto M, Takaichi S, Inomata Y, Ikenaga H, Misawa N. 2003. Structural and functional analysis of a lycopene-monocyclase gene isolated from a unique marine bacterium that produces myxol. *FEBS Letters* **545**: 120–126.

Tsuchiya T, Takaichi S, Misawa N, Maoka T, Miyashita H, Mimuro M. 2005. The cyanobacterium *Gloeobacter violaceus* PCC 7421 uses bacterial-type phytoene desaturase in carotenoid biosynthesis. *FEBS Letters* **579**: 2125–2129.

Verdoes J, Krubasik P, Sandmann G, van Ooyen M. 1999. Isolation and functional characterisation of a novel type of carotenoid biosynthetic gene from *Xanthophyllomyces dendrorhous*. *Molecular and General Genetics* **262**: 453–461.

Vogl K, Bryant DA. 2011. Elucidation of the biosynthetic pathway for okenone in *Thiodictyon* sp. CAD16 leads to the discovery of two novel carotene ketolases. *Journal of Biological Chemistry* **286**: 38521–38532.

Wang S, Zhang L, Chi S, Wang G, Wang X, Liu T, Tang X. 2018. Phylogenetic analyses of the genes involved in carotenoid biosynthesis in algae. *Acta Oceanologica Sinica* **37**: 89–101.

Wei JH, Yin X, Welander PV. 2016. Sterol synthesis in diverse bacteria. *Frontiers in Microbiology* 7: 990.

Wieland B, Feil C, Gloria-Maercker E, Thumm G, Lechner M, Bravo J-M, Poralla K, Götz F. 1994. Genetic and biochemical analyses of the biosynthesis of the yellow carotenoid 4,4′-diaponeurosporene of *Staphylococcus aureus*. *Journal of Bacteriology* **176**: 7719–7726.

Wolfe GR, Cunningham FX, Durnfordt D, Green BR, Gantt E. 1994. Evidence for a common origin of chloroplasts with light-harvesting complexes of different pigmentation. *Nature* **367**: 556–568.

Yang L-E, Huang X-Q, Hang Y, Deng Y-Y, Lu Q-Q, Lu S. 2014. The P450-type carotene hydroxylase PuCHY1 from *Porphyra* suggests the evolution of carotenoid metabolism in red algae. *Journal of Integrated Plant Biology* **56**: 902–915.

Yang Y, Yatsunami R, Ando A, Miyoko N, Fukui T, Takaichi S, Nakamura S. 2015. Complete biosynthetic pathway of the C50 carotenoid bacterioruberin from lycopene in the extremely halophilic archaeon *Haloarcula japonica*. *Journal of Bacteriology* **197**: 1614–1623.

Yokoyama A, Sandmann G, Hoshino T, Adachi K, Sakai M, Shizuri Y. 1995. Thermozeaxanthins, new carotenoid-glycoside esters from thermophilic eubacterium *Thermus thermophilus*. *Tetrahedron Letters* **36**: 4901–4904.

Young AJ, Frank HA. 1996. Energy transfer reactions involving carotenoids: quenching of chlorophyll fluorescence. *Journal of Photochemistry and Photobiology B* **36**: 3–15.