Invited Trends Article

The Non-Mevalonate Isoprenoid Biosynthesis of Plants as a Test System for New Herbicides and Drugs against Pathogenic Bacteria and the Malaria Parasite

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Higher plants and several photosynthetic algae contain the plastidic 1-deoxy-D-xylulose 5-phosphate / 2-*C*-methyl-D-erythritol 4-phosphate pathway (DOXP/MEP pathway) for isoprenoid biosynthesis. The first four enzymes and their genes are known of this novel pathway. All of the ca. 10 enzymes of this isoprenoid pathway are potential targets for new classes of herbicides. Since the DOXP/MEP pathway also occurs in several pathogenic bacteria, such as *Mycobacterium tuberculosis*, and in the malaria parasite *Plasmodium falciparum*, all inhibitors and potential herbicides of the DOXP/MEP pathway in plants are also potential drugs against pathogenic bacteria and the malaria parasite. Plants with their easily to handle DOXP/MEP-pathway are thus very suitable test-systems also for new drugs against pathogenic bacteria and the malaria parasite as no particular security measures are required. In fact, the antibiotic herbicide fosmidomycin specifically inhibited not only the DOXP reductoisomerase in plants, but also that in bacteria and in the parasite *P. falciparum*, and cures malaria-infected mice. This is the first successful application of a herbicide of the novel isoprenoid pathway as a possible drug against malaria.

Introduction

In the last five years the occurrence of a novel non-mevalonate pathway for isoprenoid biosynthesis in photosynthetic organisms, such as higher plants and algae, as well as in various heterotrophic eubacteria has been established (Lichtenthaler, 1999; Rohmer, 1999). This isoprenoid pathway had first been detected in some eubacteria where it is used for the biosynthesis of the sterol surrogates hopanoids and for the side-chain of ubiquinone (Rohmer *et al.*, 1993). In plants and most algae this DOXP/MEP pathway is responsible for the biosynthesis of the plastidic isoprenoids such as carotenoids, phytol (side-chain of chlorophylls), plastoquinone-9, isoprene, as well as mono- and diterpenes (see the review by Lichtenthaler, 1999 and references cited therein). This non-mevalonate isoprenoid pathway also occurs in the malaria parasite (Jomaa *et al.*, 1999) and in various pathogenic bacteria, but does not exist in humans or other mammals. This novel pathway opens possibilities for developing new herbicides as well as drugs against pathogenic organisms using plants and their isoprenoid biosynthesis as easy to handle test-systems as is shown here.

Abbreviations: CDP-ME, 4-(cytidine 5′-diphospho) -2-*C*-methylerythritol; DMAPP, dimethylallyl diphosphate; DOXP, 1-deoxy-D-xylulose 5-phosphate; DXR, 1-deoxy-D-xylulose 5-phosphate reductoisomerase; DXS, 1-deoxy-D-xylulose 5-phosphate synthase; IPP, isopentenyl diphosphate; MEP, 2-*C*-methyl-D-erythritol 4-phosphate.

The non-mevalonate DOXP/MEP pathway for isoprenoid biosynthesis

In plants the biosynthesis of isopentenyl diphosphate (IPP), the common C_5-precursor of all isoprenoids and terpenoids, proceeds via two independent biochemical pathways: 1) The classical acetate/mevalonate pathway is present in the cytosol and is responsible for the biosynthesis of sterols, sesquiterpenes and triterpenoids. 2) The alternative, non-mevalonate 1-deoxy-D-xylulose-5-phosphate/2-*C*-methylerythritol 4-phosphate

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(DOXP/MEP) pathway proceeds in the plastids and catalyzes the biosynthesis of the plastidic isoprenoids. The acetate/mevalonate pathway and its enzymes, as originally established in animal cells and yeasts by Conrad Bloch and Feodor Lynen, have been known for about forty years (for original literature see the review by Lichtenthaler *et al.*, 1997a). In contrast, the existence of the plastidic DOXP/MEP pathway of IPP formation in plants was detected only in the past five years applying the ^{13}C- or ^{2}H-labeling technique and high resolution NMR spectroscopy (Lichtenthaler *et al.*, 1995 and 1997b; Schwender *et al.*, 1996 and 1997; Arigoni *et al.*, 1997; Disch *et al.*, 1998). For four years this DOXP/MEP pathway has been partially elucidated with its intermediates and enzymatic steps as reviewed up to 1998 (Lichtenthaler, 1999; Rohmer, 1999).

This non-mevalonate plastidic isoprenoid pathway starts with the formation of DOXP from D-glyceraldehyde 3-phosphate and pyruvate through the action of the DOXP synthase (Fig. 1). The next step is the transformation of DOXP into 2-*C*-methyl-D-erythritol 4-phosphate (MEP) consisting of a C-C skeletal rearrangement and a reduction step which is catalyzed by the enzyme DOXP reductoisomerase (DXR). The occurrence of this isoprenoid pathway in plants and various photosynthetic algae groups was proven by the incorporation of ^{2}H- and ^{13}C-labeled 1-deoxy-D-xylulose into isoprene and phytol (Schwender *et al.*, 1997; Zeidler *et al.*, 1997), into β-carotene and methylerythritol (Arigoni *et al.*, 1997) as well as of radiolabeled DOXP and MEP into β-carotene by plastid preparations of different plants (Fellermeier *et al.*, 1999). The first enzyme of this non-mevalonate isoprenoid pathway, the DOXP synthase (DXS), has been cloned from several higher plants (Lange *et al.*, 1998; Bouvier *et al.*, 1998), the green alga *Chlamydomonas* (Lichtenthaler, 1999; Schwender *et al.*, 1999), *E. coli* (Sprenger *et al.*, 1997; Lois *et al.*, 1998) and a *Streptomyces* strain (Kuzuyama *et al.*, 2000a). The second enzyme of this plastidic IPP pathway, the DOXP reductoisomerase DXR) was recently cloned in the plants *Arabidopsis thaliana* (Schwender *et al.*, 1999) and *Mentha piperita* (Lange and Croteau, 1999a), and also in the eubacterium *E. coli* (Takahashi *et al.*, 1998). DOXP, made from D-glyceraldehyde 3-phosphate and pyruvate by the action of DOXP synthase, was readily transformed to MEP by recombinant DOXP reductoisomerase from *Arabidopsis* (Schwender *et al.*, 1999). Fosmidomycin, an antibiotic and herbicidal substance, efficiently inhibits the plants' carotenoid, phytol and isoprene biosynthesis (Zeidler *et al.*, 1998) by specifically blocking the DOXP reductoisomerase (Fig. 1) as shown for *E. coli* (Kuzuyama *et al.*, 1998) and *Arabidopsis* (Schwender *et al.*, 1999).

Recently an enzyme from *E. coli* was described that catalyzes the conversion of MEP to 4-(cytidine 5′-diphospho)-2-*C*-methylerythritol (CDP-ME) by reaction with CTP (Rohdich *et al.*, 1999). The corresponding gene, the up to now unannotated ORF *ygbP* of *E. coli*, is also present in the genomes of several micro-organisms and seems to correlate with the occurrence of the DOXP/MEP pathway (Table I). A gene similar to *ygbP* carrying a putative plastid transit peptide sequence is also found in *A. thaliana*. CDP-ME was incorporated into carotenoids by isolated *Capsicum* chromoplasts (Rohdich *et al.*, 1999), although one might argue, that it could have been hydrolyzed to MEP before incorporation. The intermediacy of CDP-ME in the DOXP/MEP pathway is, however, supported even more convincingly by a study of Kuzuyama *et al.* (2000b) which showed the CDP-ME synthase encoded by *ygbP* as essential for IPP biosynthesis in *E. coli*. The fourth enzyme of the DOXP/MEP pathway, the CDP-ME kinase, was also described recently (Lüttgen *et al.*, 2000). It phosphorylates CDP-ME at the 2-hydroxy group in an ATP-dependent reaction (Fig. 1) and is encoded by the hitherto unannotated *ychB* gene of *E. coli*, which shows the same distribution among bacteria and plants as the genes *dxs*, *dxr* and *ygbP* (Table I). This CDP-ME kinase is identical with an enzyme recently described as a presumable isopentenyl monophosphate kinase (Lange and Croteau, 1999b). Since the latter had only extremely low enzyme activities with isopentenyl monophosphate as a substrate, it is more likely, that *ychB* is indeed coding for the CDP-ME kinase. The next steps in the biosynthesis of IPP could possibly be the formation of 2-*C*-methyl-D-erythritol-2,4-cyclodiphosphate (Fig. 2) which is accumulated in several bacteria under oxidative stress conditions (Ostrovsky *et al.*, 1998; Duvold *et al.*, 1997). Further steps are possibly an intramolecular elimination of diphosphate to yield compound **3** followed

Table I. Presence of gene sequences of the non-mevalonate isoprenoid pathway in higher plants, apicomplexa (*P. falciparum*) and in eubacteria. The genes code for the enzymes 1-deoxy-D-xylulose 5-phosphate synthase (*dxs*), 1-deoxy-D-xylulose 5-phosphate reductoisomerase (*dxr*), 4-(cytidine 5′-diphospho)-2-*C*-methyl-D-erythritol synthase (*ygbP*) and 4-(cytidine 5′-diphospho)-2-*C*-methyl-D-erythritol 2-phosphate kinase (*ychB*). The *dxs, dxr, ygbP* and *ychB* homologues have not been found in mammals and archaebacteria and do not exist in the complete sequenced genomes of yeast (*Saccharomyces cerevisiae*) and the nematode *Caenorhabditis elegans.*

Organism	***dxs***	***dxr***	***ygbP***	***ychB***
Plants				
Arabidopsis thaliana	Q38854*	AJ242588#	O64726#	AAC32234§
Mentha piperita	O64904#	AF116825#		AF179283§
Capsicum annuum	O78327#			
Oryza sativa	O22567*			
Chlamydomonas reinhardtii	O81954#			
Protozoan parasite				
Plasmodium falciparum	O96694#	O96693#		
Eubacteria				
Photosynthetic bacteria:				
Synechocystis sp.[a]	P73067*	Q55663*	P74323*	P72663*
Synechoccocus leopoliensis	Y18874#	AJ250721#		
Rhodobacter capsulatus	P26242*		Q08113*	
Chlorobium tepidum	+	+	+	+
Pathogenic bacteria:				
Escherichia coli[a,b]	P77488*	P45568*	Q46893*	P24209*
Haemophilus influenzae[a]	P45205*	P44055*	O05029*	P45271*
Helicobacter pylori[a]	Q9ZM94#	AAD05777§	AAD5981§	
Treponema pallidum[a]	O83796#	AE001235#	O83525*	AE001216§
Chlamydia trachomatis[a]	O84335#	AE001281#	O84468#	AAC68399§
Chlamydia pneumoniae[a]	Q9Z6J9#	Q9Z8J8#	AAD18718§	AE001363§
Mycobacterium tuberculosis	O07184*	Q10798*	P96864*	+
Mycobacterium leprae	Q50000*	+	+	+
Pseudomonas aeruginosa	+	+	+	+
Salmonella typhimurium	+	+	+	+
Yersinia pestis	+	+	+	+
Vibrio cholerae	+	+	+	+
Neisseria meningitidis	+	+	+	+
Neisseria gonorrhoeae	+	+	+	+

,#,§ The accession numbers given in the table are those of the Swissprot database, the European Molecular Biology Laboratory (EMBL)# database and GenBank database§.

[a] The genome of these organisms is completely sequenced.

[b] Of *E.coli* there exist also pathogenic strains. + Highly similar sequences to the *E. coli dxs, dxr, ygbP* and *ychB* genes have been found in these organisms as given as preliminary data in the database of the Institute of Genomic Research (http://www.tigr.org).

by two reductases and two dehydratases (Fig. 2). The resulting IPP is then transformed to its isomer dimethylallyl diphosphate (DMAPP) the starter molecule of isoprenoid biosynthesis to which more IPP molecules are added by head-to-tail condensation to eventually yield the final isoprenoid. In *E. coli* the requirement of an IPP/DMAPP isomerase for the formation of DMAPP is, however, questionable (Hahn *et al.*, 1999), and additional reactions leading to DMAPP therefore cannot be excluded in *E. coli.*

The occurrence of the DOXP/MEP pathway in photosynthetic algae and in photosynthetic and heterotrophic bacteria

The DOXP/MEP pathway of IPP and isoprenoid formation is generally present in photosynthetic organisms i.e. higher plants (Lichtenthaler *et al.*, 1997b), green algae (Lichtenthaler, 1999; Lichtenthaler *et al.*, 1995; Schwender *et al.*, 1996 and 1997; Disch *et al.*, 1998), the red alga *Cyanidium*, the chrysophyte *Ochromonas* as well as in the oxygenic photosynthetic blue-green bacteria *Sy-*

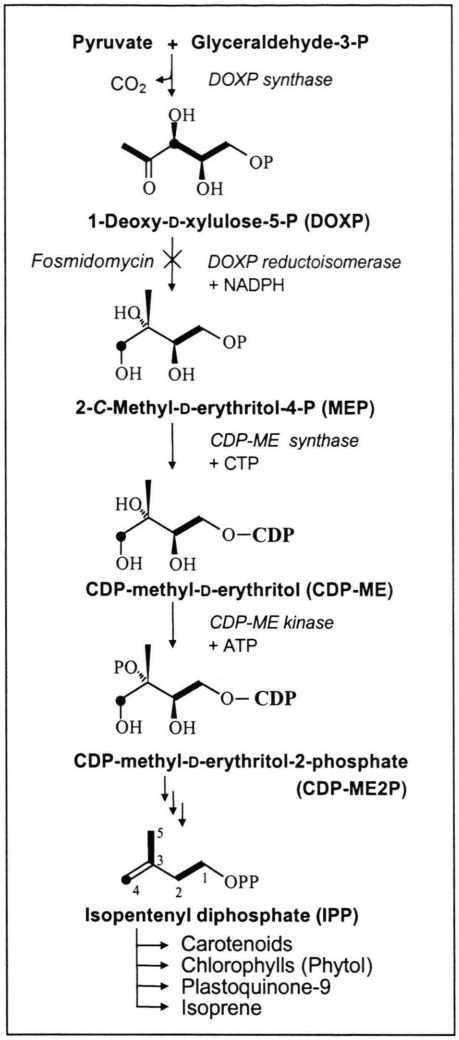

Fig. 1. Biosynthesis of the isoprenoid precursor IPP via the non-mevalonate DOXP/MEP pathway as present in plants, algae and various heterotrophic eubacteria (including pathogenic bacteria) and also in the malaria parasite *Plasmodium falciparum*. The first four enzymes DXS (DOXP synthase), DXR (DOXP reductoisomerase), 4-(cytidine 5′-diphospho)-2-*C*-methyl-D-erythritol synthase (CMS) and 4-(cytidine 5′-diphospho)-2-*C*-methyl-D-erythritol kinase (CMK) have been characterized. Several others, possibly two dehydratases and two reductases can be anticipated (see Fig. 2) but are not yet known. The second enzyme of the pathway, the DOXP reductoisomerase can specifically be blocked in plants, bacteria, and the malaria parasite by the antibiotic herbicide fosmidomycin (Takahashi *et al.*, 1998; Zeidler *et al.*, 1998; Schwender *et al.*, 1999; Jomaa *et al.*, 1999). The C-atoms 1, 2 and 4 of isopentenyl diphosphate, the first isoprenic C_5-unit, come from glyceraldehyde-3-P and the C-atoms 3 and 5 from pyruvate.

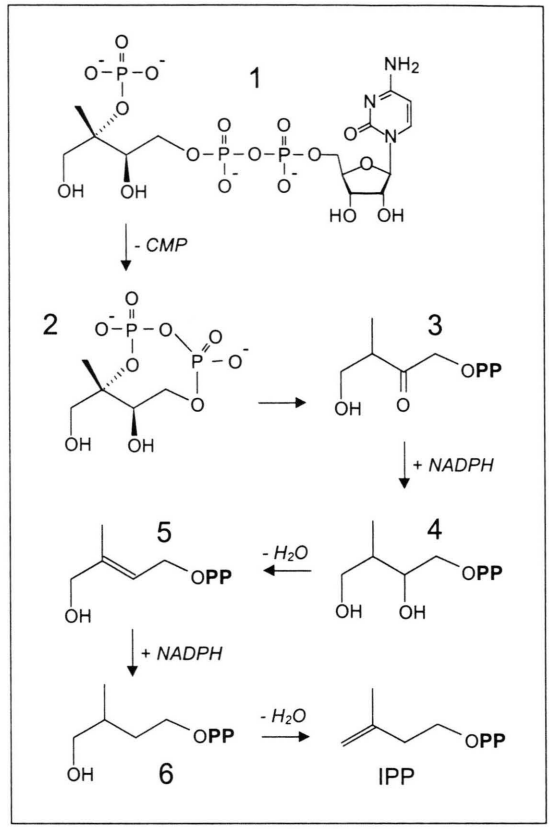

Fig. 2. Suggested sequence of the biosynthesis of isopentenyl diphosphate from CDP-2-*C*-methyl-D-erythritol-2-phosphate (1). The enzymic reaction possibly proceeds via 2-*C*-methyl-D-erythritol-2,4-cyclodiphosphate (2), followed by an intramolecular elimination of diphosphate (from compound 2 to 3) and requires two reductases (+NADPH) and two dehydratases (-H_2O).

nechocystis (Schwender *et al.*, 1997; Disch *et al.*, 1998; Proteau, 1998) and *Synechococcus leopoliensis* (Miller *et al.*, 1999). The available data indicate that during the evolution of algae and higher plants (Lichtenthaler, 1999) the photosynthetic eukaryotes have conserved the DOXP/MEP pathway which originally had been imported into the eucyte by endosymbiontic cyanobacteria-like progenitors of the present chloroplasts of algae and higher plants. Genes of the DOXP/MEP pathway are also found in the two photosynthetic bacteria with anoxygenic photosynthesis *Rhodobacter* and *Chlorobium* (Table I).

The DOXP/MEP pathway of isoprenoid formation is found in many different eubacterial taxonomic groups, and most eubacteria seem to use it

(Rohmer, 1999). In fact, it was first detected by Rohmer and his group in 1993 during studies of ^{13}C-incorporation into the bacterial isoprenoid hopanoids (Rohmer *et al.,* 1993). The eubacteria with the DOXP/MEP pathway of IPP formation, as deduced from ^{13}C-incorporation of applied ^{13}C-glucose or ^{13}C pyruvate into hopanoids or the prenyl side-chain of ubiquinones, were *Zymomonas mobilis*, *Methylobacterium fujisawaense*, *Alicylobacillus acidoterrestris* and *E. coli* (Rohmer *et al.*, 1993).

In several data bases orthologs of the four known genes of IPP formation, *dxs*, *dxr, ygbP* (*CDP-ME-synthase*) and *ychB* (*CDP-ME-kinase*) can be found in higher plants as well as in many bacteria including pathogenic bacteria (Table I). The following organisms belong to the latter category: *Haemophilus influenzae* (fever), *Mycobacterium tuberculosis*, *Mycobacterium leprae* (leprosy), *Chlamydia trachomatis* (urethritis, trachoma, eye disease), *Treponema pallidum* (syphilis), *Helicobacter pylori* (gastric ulcer), *Vibrio cholerae* and *Pseudomonas aeruginosa* (wound infection). Our database research showed in bacteria, for which the entire genome has been sequenced (e.g. *E. coli*, *Helicobacter pylori, Chlamydia trachomatis as well* as the non-pathogenic *Bacillus subtilis*, *Thermotoga maritima*, *Deinococcus radiodurans*), orthologs to *dxs*, *dxr*, *ygbP* and *ychB*, but no orthologs for the key enzyme of the acetate/mevalonate pathway, the HMG-CoA reductase. In contrast, in completely sequenced Archaebacteria (*Pyrococcus horikoshii*, *Methanococcus jannaschii*, *Methanobacterium thermoautotrophicum*) we found in the database search no orthologs of the four known enzymes of the DOXP/MEP pathway, but only orthologs of HMG-CoA reductase. In most cases only one of the two IPP pathways seemed to appear in a bacterial organism. *Helicobacter pylori* may be an exception to this rule since its genome also contains a HMG-CoA reductase ortholog.

Due to the strong conservation of the amino acid sequences of the *dxs*, *dxr*, *ygbP* and *ychB* genes it can be expected that the DOXP/MEP pathway enzymes from plants and pathogenic bacteria share the same enzymic mechanisms and the same binding sites for inhibitors. As a consequence, it should be possible to control all these pathogenic bacteria by inhibitors of the DOXP/MEP pathway of IPP formation. In fact, it had been shown already in 1989 that the herbicide fosmidomycin inhibits the isoprenoid biosynthesis in several bacteria (Shigi, 1989).

Occurrence of the DOXP/MEP pathway in the malaria parasite *P. falciparum*

Plasmodium falciparum, the parasite causing malaria, is a protozoan organism that possesses a unique plastid-like cell organelle, the apicoplast (McFadden *et al.*, 1997). The latter with its circular 35-kb genome resembles in many aspects the chloroplasts in algae and plant cells (Köhler *et al.*, 1997) but, being pigment free, does not perform photosynthesis. The apicoplast was apparently acquired by members of the phylum *Apicomplexa* by secondary endosymbiosis of an alga, presumably a green alga. The metabolic function of the apicoplast is essential for the survival of the parasites (Fichera and Roos, 1997). Unicellular green algae only possess the plastid-bound DOXP/MEP pathway of IPP and isoprenoid formation (Lichtenthaler, 1999; Lichtenthaler *et al.*, 1995; Schwender *et al.*, 1996; Disch *et al.*, 1998). Apparently *P. falciparum* needs the DOXP/MEP pathway of IPP formation of the apicoplast for its metabolism and replication and seems not to have the mevalonate pathway. In fact, when searching for genes encoding the enzymes of the DOXP/MEP pathway in *P. falciparum* a DOXP synthase (DXS) and a DOXP reductoisomerase (DXR) were found which have a great similarity to the corresponding enzymes of bacteria and plants (Jomaa *et al.*, 1999). This DXR exhibited at the NH_2-terminal side a transit peptide sequence that is typical for targeting the enzyme in plants into the plastid, and in the case of *P. falciparum* into the apicoplast. The existence of the DOXP/MEP pathway in *P. falciparum* was further confirmed as the recombinant *P. falciparum* DXR converted DOXP into MEP (Jomaa *et al.*, 1999). This then opens up new possibilities to fight against the malaria parasite *P. falciparum* with inhibitors that specifically block the isoprenoid biosynthesis of the apicoplast (see below).

Inhibition of the DOXP/MEP pathway in plants, bacteria and the malaria parasite

In our search for inhibitors of the plants' DOXP/MEP pathway of IPP and isoprenoid formation we used two approaches: 1) we were look-

ing for structural analogues to inhibitors of the plastidic ketol acid reductoisomerase (KARI) which, like the DXR, also catalyzes a C-C skeleton rearrangement followed by an NADPH-dependent reduction step (Lichtenthaler, 1999; Zeidler *et al.*, 1998) and 2) we checked agrochemicals with herbicidal activity (Kamuro *et al.*, 1988; Patterson, 1987) that had been shown to also affect isoprenoid biosynthesis in several bacteria (Shigi, 1989). Fosmidomycin (Fig. 3) proved to be an efficient inhibitor of the plastidic and bacterial isoprenoid biosynthesis (Zeidler *et al.*, 1998; Shigi, 1989), and it specifically blocked the recombinant DXR of *E. coli* and *Arabidopsis* (Kuzuyama *et al.*, 1998; Schwender *et al.*, 1999). Fosmidomycin is a structural analogue to 2-*C*-methylerythrose 4-phosphate (Fig. 3), the intermediate in the conversion of DOXP to MEP by DXR. In addition it was demonstrated that fosmidomycin and its derivative FR-900098 also block the recombinant DXR of *P. falciparum*, suppress the growth of multidrug resistant *P. falciparum* strains and cure malaria infected mice (Jomaa *et al.*, 1999). This is a first example to demonstrate that a herbicidal inhibitor of the DOXP/MEP pathway can also be applied as an antibiotic against pathogenic bacteria as well as an anti-malaria drug.

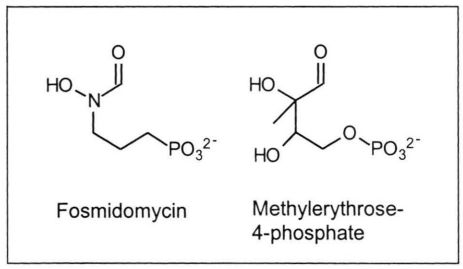

Fig. 3. Chemical structure of fosmidomycin, a specific inhibitor of the DOXP reductoisomerase DXR of plants, bacteria, and the malaria parasite. Fosmidomycin is a structural analogue of 2-*C*-methylerythrose 4-phosphate, the intermediate in the enzymatic transformation of 1-deoxy-D-xylulose 5-phosphate (DOXP) into 2-*C*-methyl-D-erythritol 4-phosphate (MEP) (see Fig. 1).

Plant test systems for new drugs and inhibitors of the DOXP/MEP pathway

The efficiency of herbicides and antibacterial substances is essentially determined by the uptake rate, the intracellular transport of the active compound to the enzyme target site and the breakdown and inactivation processes of the active ingredient. These processes differ in various organisms. Thus, one cannot expect that newly developed inhibitor compounds will be equally effective in plants, pathogenic bacteria, and the parasite *P. falciparum*. For the first screening of chemicals as potential inhibitors of the DOXP/MEP pathway of isoprenoid biosynthesis in different organisms plant test-systems are, however, a preferential and first choice. All the precautions during research with pathogenic bacteria and the parasite *P. falciparum*, such as particular security laboratories or special safety handling of the micro-organisms are not required when working with whole plant test-systems or isolated plant enzymes of the DOXP/MEP pathway. Once a new structural class of potential inhibitors/drugs has been detected in plant test-systems, the lead structure and its chemical derivatives can be further tested for maximum efficiency of inhibition in the individual pathogenic bacteria and in the parasite *P. falciparum*. Below some examples of test-systems that have been used by the authors for screening potential new drugs and inhibitors of the DOXP/MEP pathway:

Measuring isoprene emission in plant leaves

Many herbaceous plants and leaves of trees possess the ability to emit the volatile isoprene. Isoprene is made via the DOXP/MEP pathway (Schwender *et al.*, 1997; Zeidler *et al.*, 1997) from IPP via DMAPP. It is thus a very direct IPP product which requires only two additional enzymes and not as many as in the case of phytol or carotenoids. Thus, further enzymatic steps of plastidic isoprenoid synthesis can be excluded as potential targets by studying the effect of chemicals on isoprene emission. It is emitted by leaves at high irradiances and temperatures above 30 °C. The formation of isoprene can easily be studied with a cuvette-test based on the UV absorption of gaseous isoprene in air (Zeidler and Lichtenthaler, 1998).

Test with green ripening tomatoes

Injecting the potential chemicals into green ripening tomatoes, the accumulation of the red carotenoid lycopene will efficiently be suppressed when the active ingredient is an inhibitor of the DOXP/

MEP pathway of IPP formation (Zeidler *et al.*, 1998). This is an easily available test-sytem for visual screening.

Light-induced greening of etiolated barley leaves

A dose-dependent inhibition of carotenoid and chlorophyll accumulation can be tested during the light-induced pigment formation in etiolated leaves as shown for fosmidomycin (Zeidler *et al.*, 1998).

Assays with recombinant enzymes of the DOXP/MEP pathway

In assays with individual enzymes of the DOXP/MEP pathway all potential inhibitors can be recognized by a dose-dependent inhibition of the correponding enzyme product. Assay systems have been developed for the DXS from the green alga *Chlamydomonas* (Lichtenthaler, 1999; Schwender *et al.*, 1999), and also for DXR cloned from *Arabidopsis* (Schwender *et al.*, 1999) and from *E. coli* (Kuzuyama *et al.*, 1998). Others including CDP-ME synthase and CDP-ME kinase of plants are in development.

Besides plant systems, it is also possible to apply non-pathogenic bacteria as test-systems, to which the enzymes of carotenoid biosynthesis have been transferred (Harker and Bramley, 1999).

Concluding remarks

So far there exists only one inhibitor lead structure of the DOXP/MEP pathway of isoprenoid formation, fosmidomycin and its derivatives, which block the DOXP reductoisomerase (DXR). Further targets for potential herbicides, antibiotics, and drugs are the DOXP synthase (DXS) and the enzymes CDP-ME synthase and the CDP-ME kinase (cf. Fig. 1) as well as the other yet unknown enzymes (possibly six) which are required for the transformation of MEP into the final C_5-diphosphate IPP (cf. Figs. 1 and 2). The fosmidomycin example is a successful transfer of a plant DOXP/MEP pathway inhibitor to a malaria cure in an animal system, and fosmidomycin can also be used to control bacterial pathogens that possess the DOXP/MEP pathway. This makes the development of new drugs blocking this novel isoprenoid pathway a rewarding task and challenge. Since the DOXP/MEP pathway of IPP formation does not occur in mammals, inhibitors and drugs of this IPP pathway cannot directly affect the isoprenoid metabolism of humans. The plant test-systems of the DOXP/MEP pathway require no particular safety precautions and are readily available. Plants produce various types of plastidic isoprenoids in larger amounts and in a shorter time than bacteria and can thus easily be applied in the search for new herbicides as well as drugs against pathogenic microorganisms.

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