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An Evolutionary Diversity of the Primary Metabolism for Plant Synthetic Biology

Faahan Simon*

Department of Life and Environmental Sciences, University of Sydney, NSW 2006, Australia

Introduction

The numerous and frequently plentiful chemical substances that plants make are essential for these sessile and multicellular organisms to occupy a range of environmental niches. Several of these phytochemicals are referred to as specialised or secondary metabolites since they are produced in a lineage-specific manner. Many of these plant-based natural products also give human society precious resources and crucial nutrients for the creation of medications and biomaterials. Rapid identification of plant-specific metabolic enzymes is made possible by next-generation sequencing and sophisticated mass spectrometry technology.

Microbial hosts have been engineered to create chemical production platforms that are optimised for a specific primary metabolic branch on which various downstream pathways, including plant-specific metabolic pathways, have been introduced. These hosts have well-developed genetic tools and industrial-scale culture methods (for example, yeast). Microbial production of some classes of plant natural products [1], such as alkaloids and phenolics, appears to be more difficult, likely because of their toxicity, pathway complexity, and inefficiency of plant-derived enzymes, despite the fact that significant progress has been made in the industrial-scale production of terpenoid in microbes.

Description

Many plant lineages have developed with a tremendous diversity of chemicals (left). Synthetic biology (green, right) allows for the efficient manufacture of target molecules by identifying and reconstructing the underlying specialised metabolic pathways in a heterologous host, or chassis (e.g. nutraceuticals, pharmaceuticals, and bio-based materials). Moreover, the host's upstream major metabolic pathways can be modified to improve the availability of a particular precursor(s) (blue, right). If their benefits and drawbacks (table) are carefully considered and addressed, plants can provide alternative platforms for the sustainable and perhaps efficient production of natural plant products [2,3].

Although it is still in its infancy, the use of heterologous plant hosts offers an alternative and sustainable way to generate plant-based natural products that makes use of large-scale culture systems that are driven by endogenous photosynthetic energy generation and carbon fixation. Opportunities to cultivate high-yielding plants in marginal lands, which can avoid direct rivalry with food crop production and limit environmental impacts, have advanced thanks to recent investments and efforts in creating bioenergy crops. In comparison to microbial hosts, plant hosts may also have superior storage capacity and toxin resistance for the synthesis of phytochemicals. Hence, plant chassis could potentially offer interesting alternate platforms to make some of these difficult-to-produce metabolites in microorganisms. This is especially true if appropriate plant hosts are carefully chosen and created.

Engineering plant primary metabolism presents a number of significant hurdles not present in microorganisms. (a) Due to low transformation efficiency and lengthy generation cycles of most plants, the ability to conduct genetic engineering and mutagenesis screening in plants is significantly more constrained than in microorganisms (months to years versus hours to days). (b) Compared to microorganisms, which can use a variety of carbon sources, plant metabolism is probably more limited since it relies nearly exclusively on the carbon intake from photosynthetic CO2 fixation. (c) Plant major metabolic pathways frequently compromise overall growth and yield since they are intimately entwined with one another and directly related to the growth and development of these complex multicellular organisms.

Studies on microbial metabolic engineering and synthetic biology have shown that efficient provision of a particular primary precursor or precursors as well as carbon flux redirection are necessary for the effective creation of downstream target products. Hence, for effective and significant production of natural products in plants, a comprehensive understanding and engineering of both primary and specialised metabolisms are essential [4].

There are some examples of evolutionary diversification of primary metabolic pathways, particularly at the interface between primary and specialised metabolism, even though primary metabolism is typically thought to be conserved across the plant kingdom in contrast to highly-diversified specialised metabolism. Achieving efficient production of plant natural products in carefully chosen plant hosts will require optimising plant primary metabolism in coordination with downstream specialised metabolic pathways, which can be accomplished by exploring and utilising such relatively uncommon but important evolutionary innovations of plant metabolism. One approach to overcoming these difficulties is to carefully select host plants that are naturally geared towards the synthesis of particular classes of compounds, and to then design the primary metabolism in a rational and exact manner to maximise the supply of a particular precursor. Here, I go over a promising strategy for achieving this objective by studying nature's millions of years of experiments [5].

The precursor and building blocks of several isoprenoid chemicals, such as sterols (such as cholesterols), dolichol, and quinones, are isopentenyl diphosphate (IPP) and its allylic isomer dimethylallyl diphosphate (DMAPP) (e.g. ubiquinone). IPP and DMAPP are also used in plants to make rubbers, isoprene, mono- and sesquiterpene volatiles, and a variety of di- and tri-terpenoids, as well as photosynthetic pigments like chlorophylls and carotenoids, quinones like plastoquinone and phylloquinone, plant hormones like gibberellins, brassinosteroids, and abscisic acid. The 2-C-methyl-d-erythritol 4-phosphate (MEP) and mevalonate (MVA) pathways can both be used to generate IPP and DMAPP.

The MEP pathway is present in many bacteria, including Escherichia coli and cyanobacteria, whereas the MVA pathway is present in many mammals,

***Address for Correspondence:** Faahan Simon, Department of Life and Environmental Sciences, University of Sydney, NSW 2006, Australia; E-mail: simon.faahan@yahoo.au*

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fungi, and archaea. Most species have one of the two processes. Nonetheless, it is noteworthy that plants and a large number of algae possess both MEP and MVA routes for IPP and DMAPP synthesis, supporting the creation of these various isoprenoid compounds in various subcellular compartments. There seems to be some, but not much, metabolic cross-talk between these two pathways. Although certain isoprenoids, like the sesquiterpene artemisinin from plants, have been effectively generated by microbial synthetic biology, it is also possible to manufacture diverse isoprenoid molecules using plant hosts by taking use of their inherent ability to produce abundant IPP.

The MVA route begins with acetyl coenzyme A (CoA), of which three are condensed to 3-hydroxy-3-methylglutaryl-CoA (HMG-CoA), reduced to MVA, and then phosphorylated and decarboxylated by ATP to produce IPP. IPP:DMAPP isomerase then converts IPP and DMAPP into one other (IDI). According to fluorescence protein-tagged subcellular localization studies, the MVA pathway in plants primarily occurs in the cytosol, but the later steps, which are catalysed by phosphomevalonate kinase (PMK), mevalonate diphosphate decarboxylase (MPDC), and IDI, also seem to be localised in the peroxisomes [6]. Similar to bacteria, fungi, and animals, HMG-CoA reductase (HMGR), this converts HMG-CoA into MVA irreversibly and is attached to endoplasmic reticulum (ER), appears to be the primary regulating enzyme of the MVA pathway in plants.

Conclusion

One could hypothesise that the cyanobacterial endosymbiosis, which likewise produces IPP and DMAPP by the MEP pathway, is the source of the plastidic MEP pathway of plants and algae. While some of these genes were horizontally transferred to a common ancestor of plastid-bearing eukaryotes, evolutionary analyses of individual MEP pathway enzymes of plants and algae revealed that these enzymes have mosaic evolutionary origins and share last common ancestors with either cyanobacteria, -proteobacteria, or Chlamydia (57, 58). The plant MEP pathway is probably regulated differently from that of bacteria due to its complicated evolutionary history and the great and diverse demand for synthesising numerous and plentiful isoprenoid chemicals. DXP synthase (DXS) catalyses the first reaction, which is irreversible and commits carbon to the MEP route.

Acknowledgement

Not applicable.

Conflict of Interest

None.

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