



Steviol Synthesis in Yeast a student project

An iGEM project on alternative sweeteners

Several diseases are thought to be connected with an increased sugar consumption. The use of non-sugar sweeteners could be a possible approach for both reducing the risk of sickening and a proper ingestion without high sugar intakes.

Several of the known problems with common sweeteners could be overcome by diterpene glycosides which are produced by the paraguayan herb *Stevia rebaudiana*.

One possibility to improved sweetening

Need an ER? Go Yeast!

Yeast is an eukaryotic unicellular organism. In addition to this it is a popular model organism and in our case it is the yeast *Saccharomyces cerevisiae*. But *E.coli* is also a popular model organism and even grows faster than yeast. Moreover there are more biobricks available for *E.coli*. So why do we use yeast? For the simple reason that we need an endoplasmatic reticulum to produce steviol, because the kauren oxidase is localized in the ER. In addition yeast has got further advantages. It is an established food additive producer and it is closer related to *Stevia*.

Furthermore we utilize the yeast's ability for homologue recombination. Yeast only needs about 40 bp of a homologue area for recombination. That means that DNA sequences can be integrated into another DNA molecule if the DNA sequence has got homologue areas to the target DNA. We want to clone the required genes and the promoters and terminators into plasmids via homologue recombination in *S.cerevisiae*.

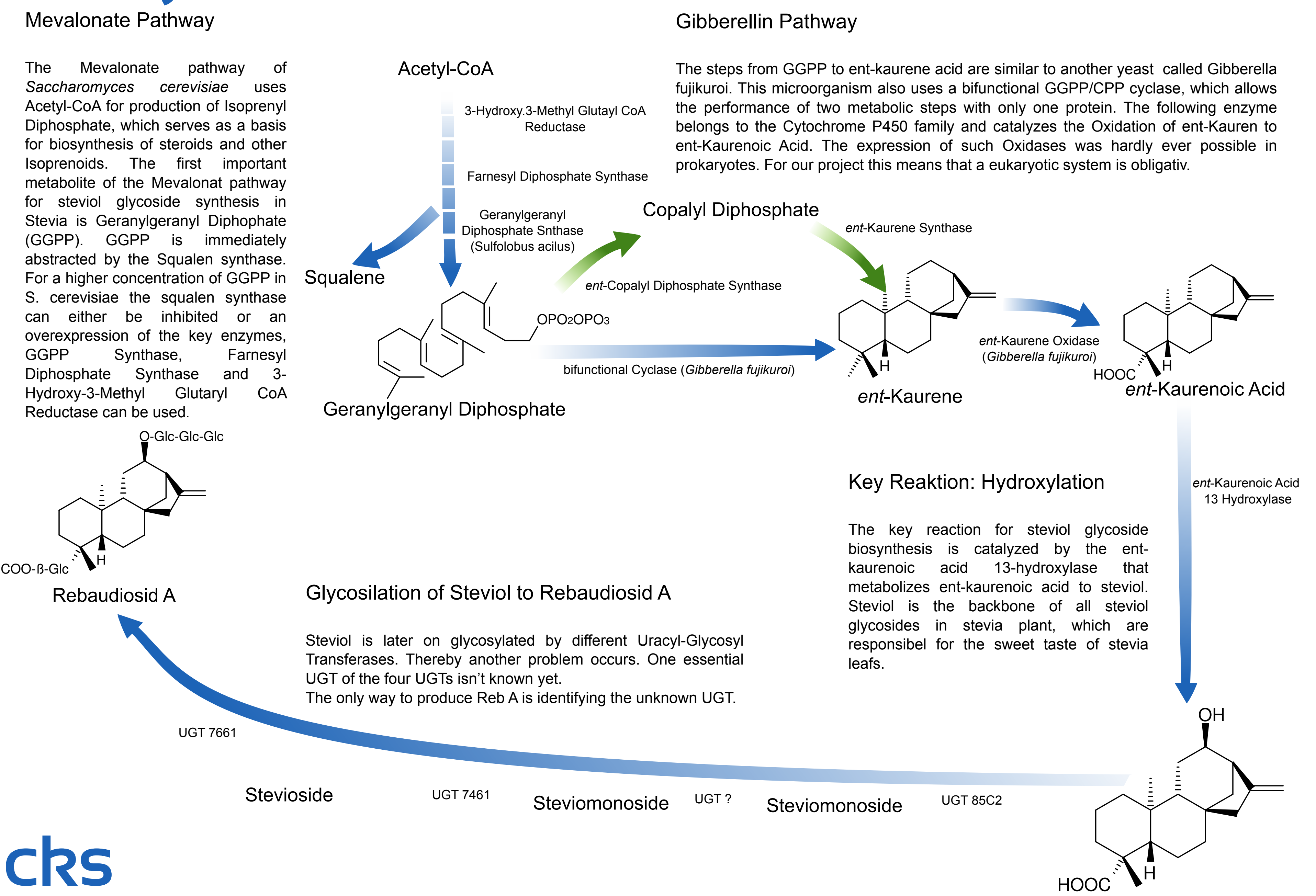
Biological Parts: Biobricks

Biobricks are standardized open source DNA parts specified in RFCs. They are functionally defined molecules that can be assembled and integrated into model organisms to construct a new biological pathway. Especially the synthetic biology benefits of such standardized DNA parts, because biobricks are freely accessible resources to the public and therefore a support for progress in biological research and development.

The aim of synthetic biology is to assembly synthetic and biological units to create new substances and systems. For such a research is an interdisciplinary team needed. A possibility for an interdisciplinary work, even in the stage of education, is the iGEM compe-

products based on steviol glycosides is the microbial production of the compound Rebaudioside A which could also lower the enviromental costs of sweetener production. In our interdisciplinaty project in the context of the annual international genetically engineered machine competion (iGEM) we want to investigate the possibilty of such a microbial sweetener production in the baker yeast *Saccharomyces cerevisiae*.

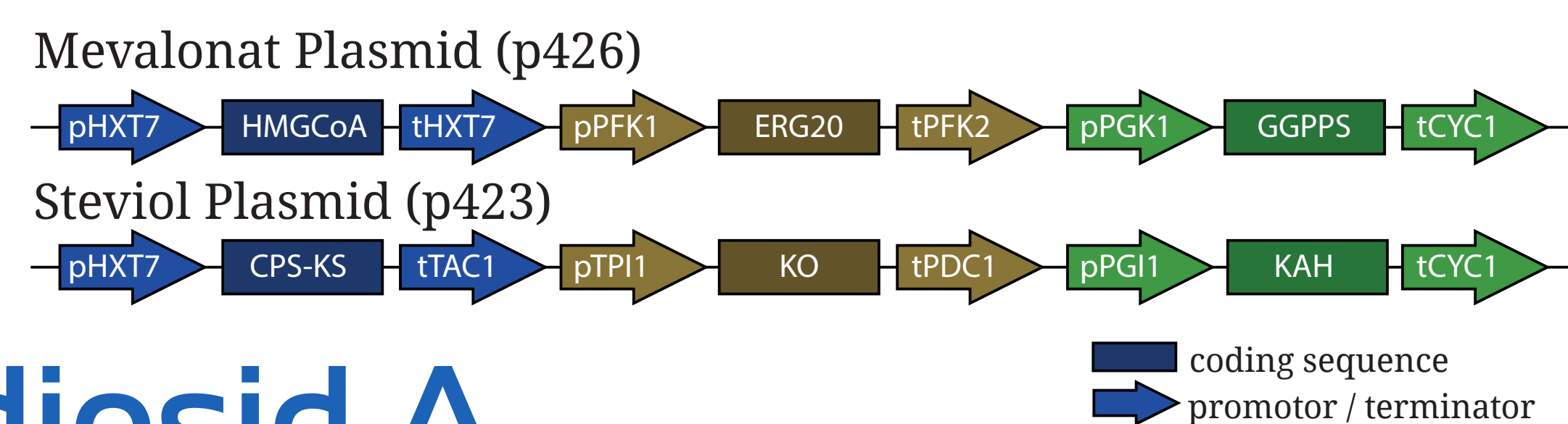
Acetyl-CoA to Rebaudiosid A



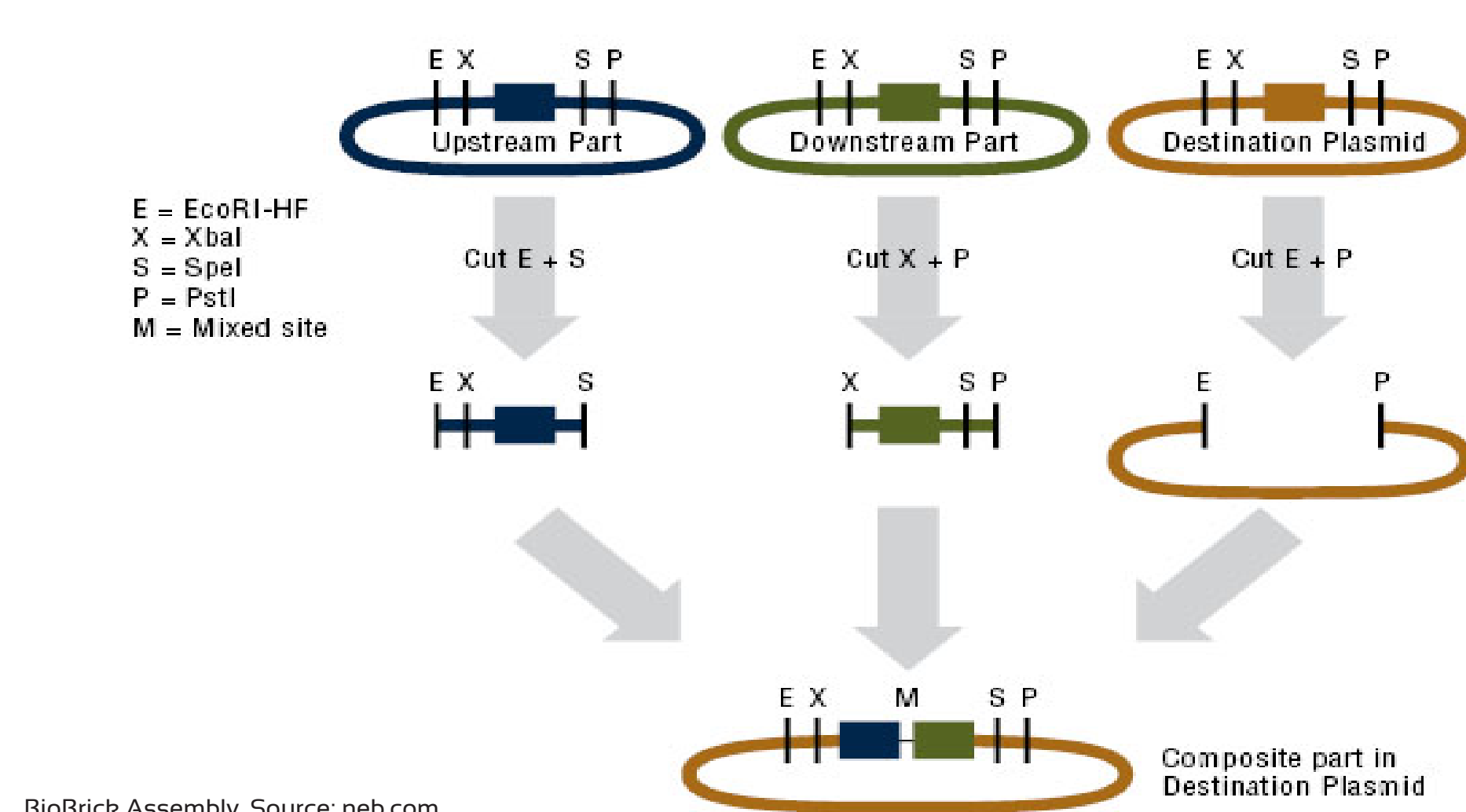
Schedule

- November:** First Team Meeting
- March:** iGEM Registration
- June:** First Sponsoring
Achema
Night of Science
PCR
- July:** Transformation Mevalonat Plasmid
- August:** GC-MS for GGPPS
- September:** Transformation Steviol Plasmid
GC-MS for Steviol, ent-Kauren
- October:** European Jamboree Amsterdam
- November:** World Championship Boston

Plasmids



tion (international Genetically Engineered Machine Competition). The idea of iGEM is to motivate research groups of students to deal with ideas related to synthetic biology, whereas the concept of biobricks should be realized.



The Team

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