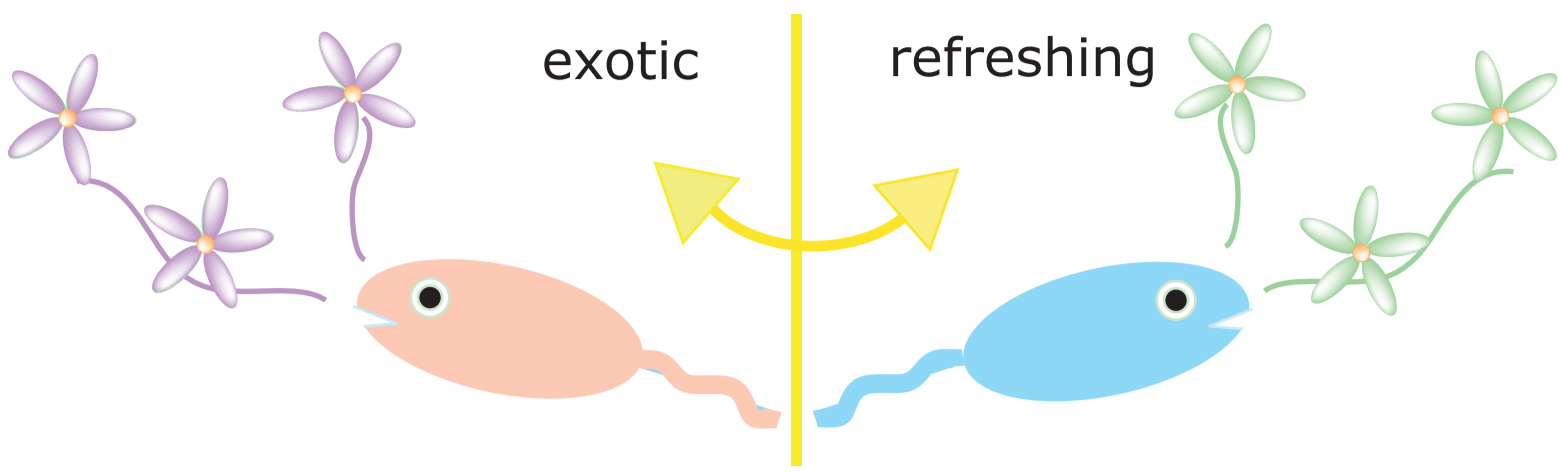


Aromatic E.coli

K.Nakajima, S.Fukushima
K.Suzuki, M.Ikezumi, D.Ave

Goal ~ aroma switching



We aimed to create *E.coli* that produce aroma.

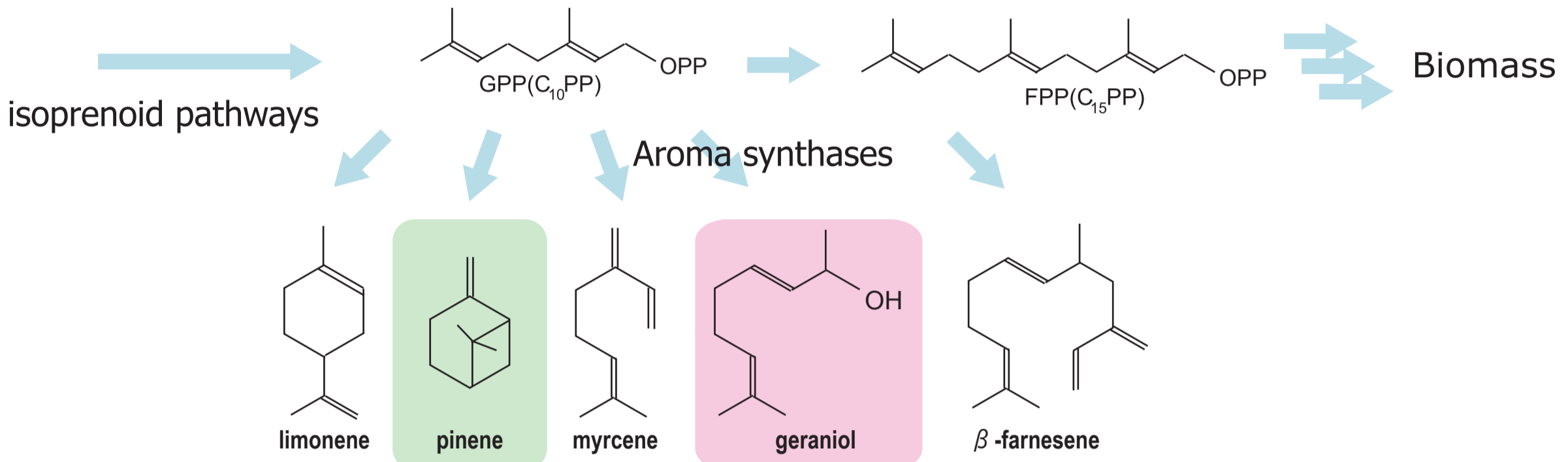
We would like *E.coli* to emit various aroma in response to light implement.

Our Aroma ~ terpenoids

Aroma terpenes are biosynthesized by wide variety of plants, generating a variety of gorgeous scent.

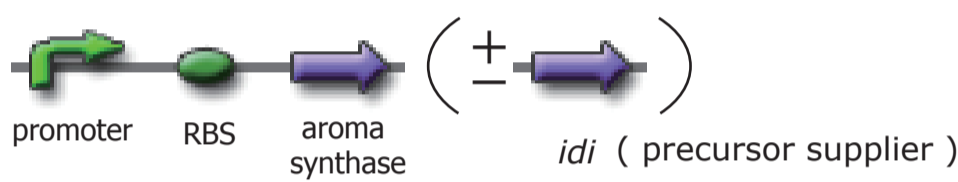
1. **Ubiquitous** ; precursors available in all organisms
2. **Similarity and Diversity** ; various enzymes, very different smells, but share the chemistry and precursors
3. **More than the Scent** ; - anti insecticides
- pain relief / relaxation
- anti-inflammatory.....etc

Synthetic Pathway



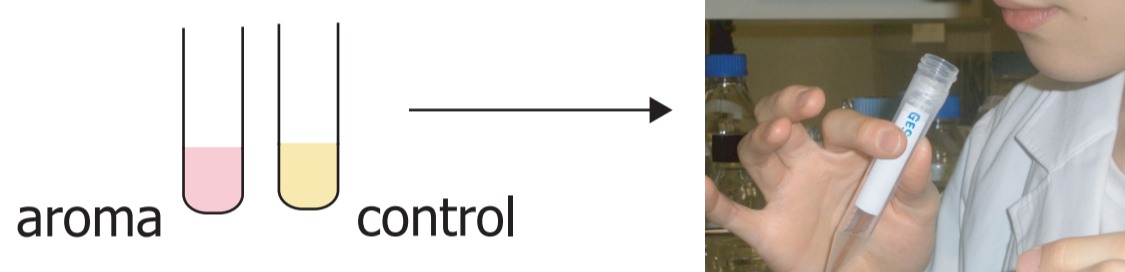
Experiments / Result

Smell Test of Various Aroma Synthase



idi (isopentenyl diphosphate isomerase, *Phaffia rhodozyma*)
... enhances precursor production

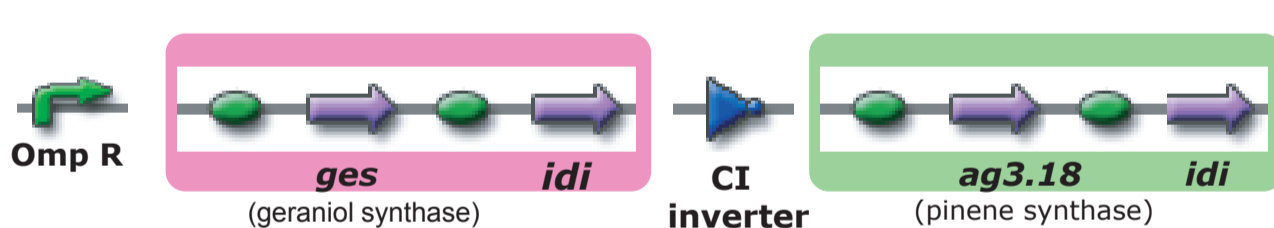
Method:
Instead of GC/MS (that we couldn't assess within iGEM time), we use "human nose" analysis.



Results

Gene	Terpene	Origin	Scent		Courtesy of...	
			idi -	idi +		
<i>ges</i>	Geraniol	<i>Ocimum basilicum</i>	○	◎	soothing & lemony	Picherskey Lab (U.Michigan)
<i>ag3.18.118</i>	Pinene	<i>Abies grandis</i>	○	◎	refreshing & woody	Croteau Lab (W.S.U.)
<i>ag10</i>	Limonene	<i>Abies grandis</i>	○	◎	sweet & sour	Croteau Lab (W.S.U.)
<i>p43</i>	(E)-β-farnesence	<i>Mentha x piperita, L.</i>	○	◎	soft mellow smell	Croteau Lab (W.S.U.)
<i>pts-5526</i>	Myrcene	<i>Perilla frutescens</i>	✕	—	—	Honda Lab (Kyoto Univ.)

Step2: Harnessing by Osmotic Switch



...Just constructed (on 11/2 /2006)
Genotype; OK
Phenotype; To be tested (soon!)

Geraniol & Pinene compete for GPP (precursor)

Sharp Switching!!

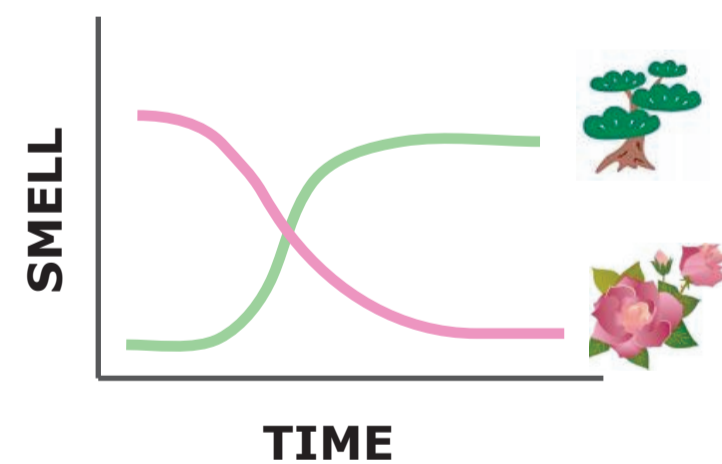
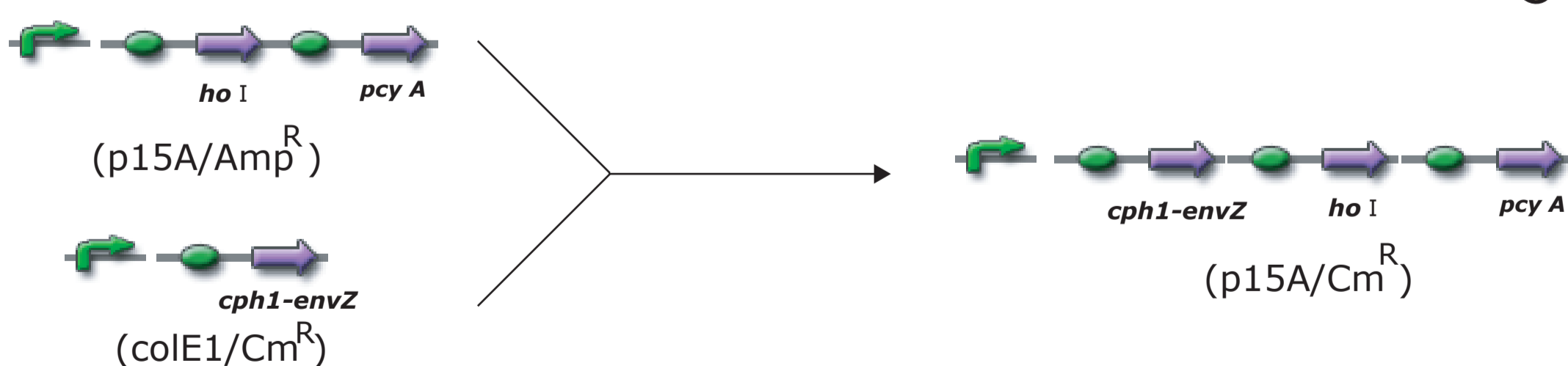


Image. The ratio of aroma output

In Very Near Future

Step3: Control of Scent Generator by Light

UCSF/UT Austin's light sensor integrated into single plasmid(p15A,Cm^R)



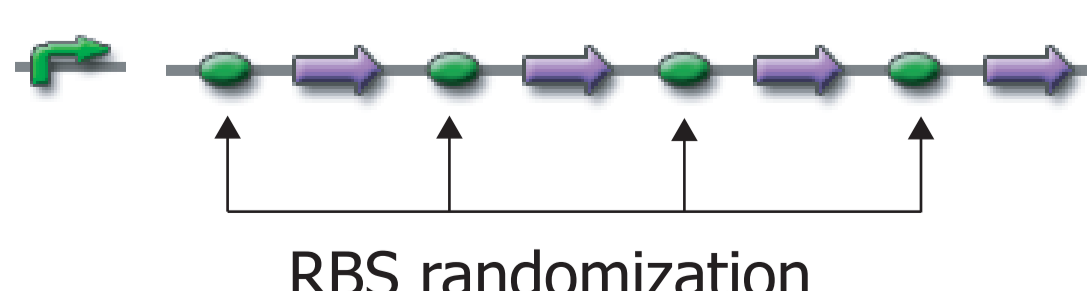
given by AS+Chris (Levska et al., Nature **438**: 441-442,2005)



A litte Far Future

Aroma Revolution

Create your favorite smell with several aroma genes!



RBS randomization

High throughput "sniffing" analysis



Compound name	%
Limonene	14.04
Verbenone	21.37
α-Terpinol	8.83
Perillyl alcohol	7.86
Carophyllene	6.98
Geraniol	2.50
Neral	2.04
B-Ocimene	1.98
Germaerene-D	1.78
Terpinene-4-Ol	1.69
Geranyl acetate	1.45
Methyl geranate	1.40
Carvone	1.32
P-Menth-1-en-9-ol	1.29
Sabinene	1.27
Different compounds	Trace (<1%)