Tom Ellis - CSynBI

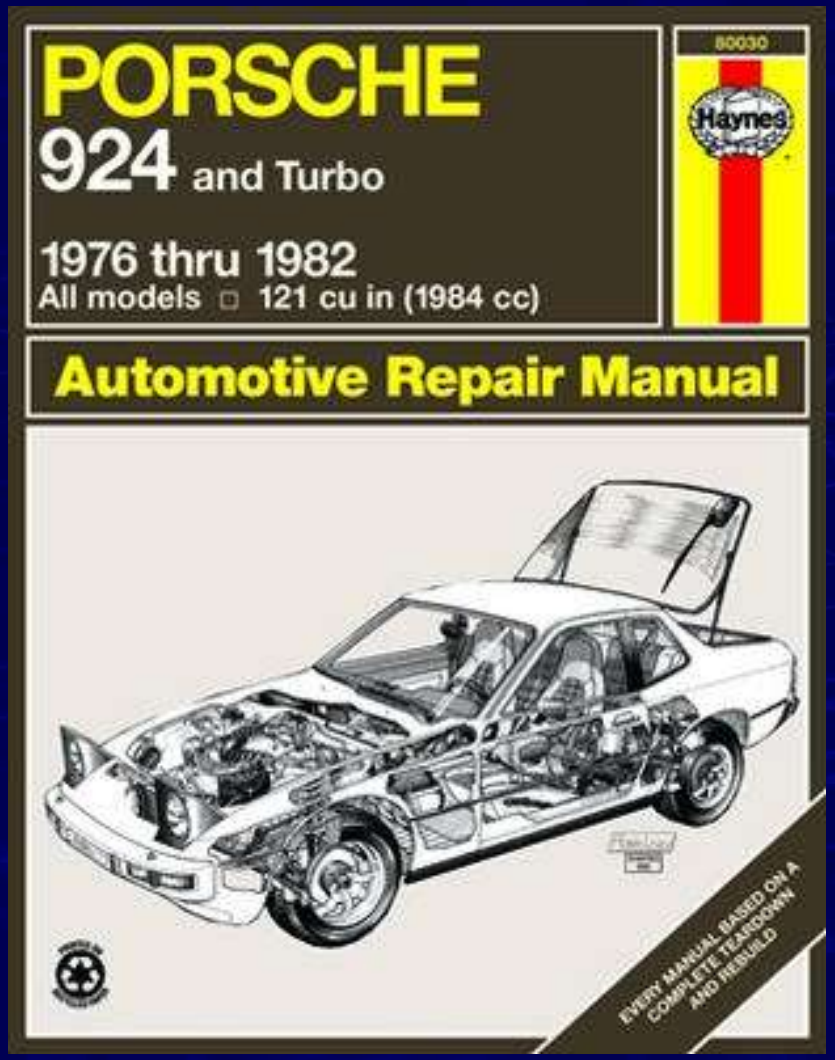
Developing chassis that are fit for purpose

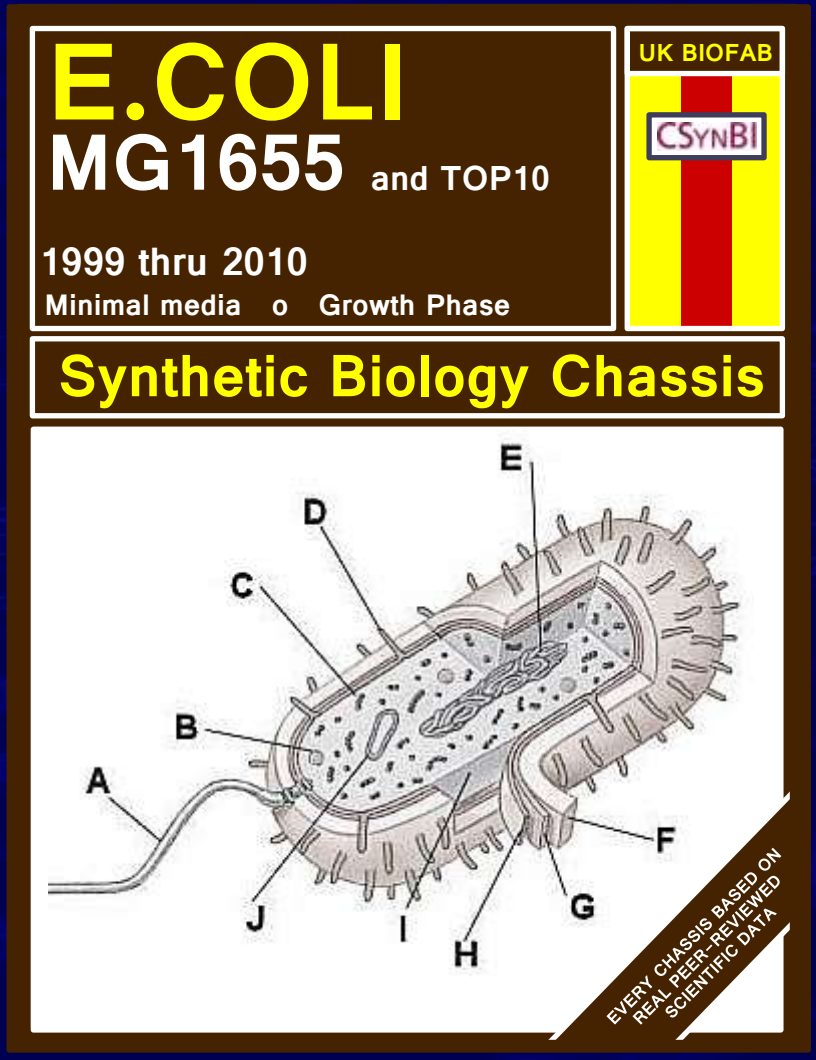
Imperial College

Synthetic Biology Club

March 2010

Chassis for synthetic biology





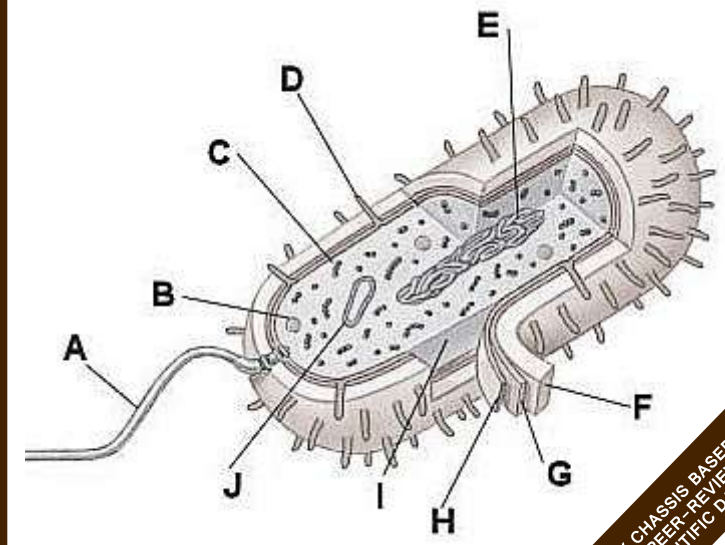
1st Generation Synthetic Biology

E.COLI
MG1655 and TOP10

1999 thru 2010
Minimal media o Growth Phase

UK BIOFAB
CSynBI

Synthetic Biology Chassis



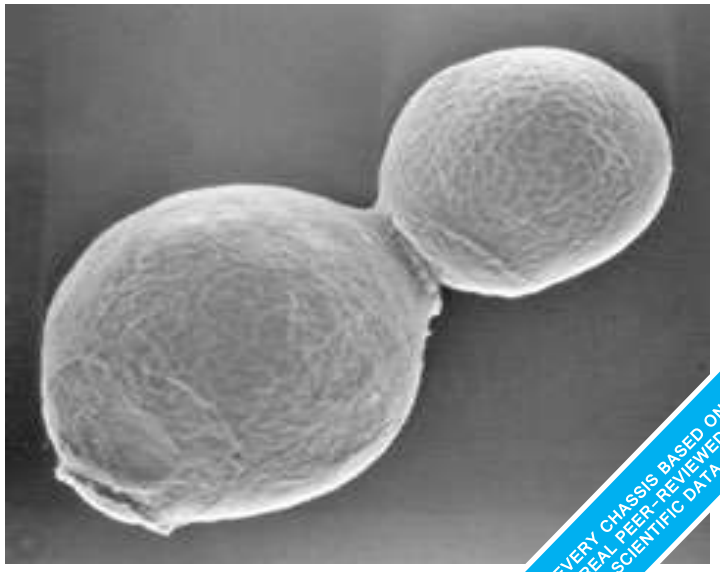
EVERY CHASSIS BASED ON REAL PEER-REVIEWED SCIENTIFIC DATA

YEAST
S.CEREVISIAE

2002 thru 2010
Synthetic Defined Media o Growth Phase

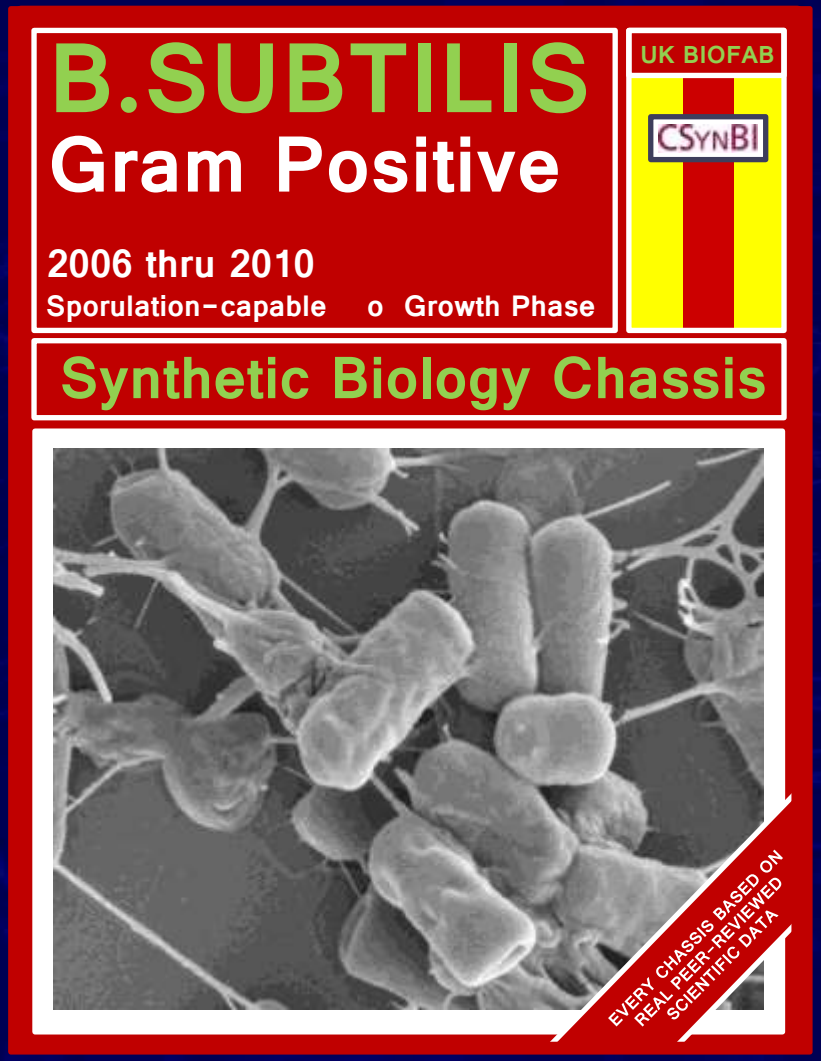
UK BIOFAB
CSynBI

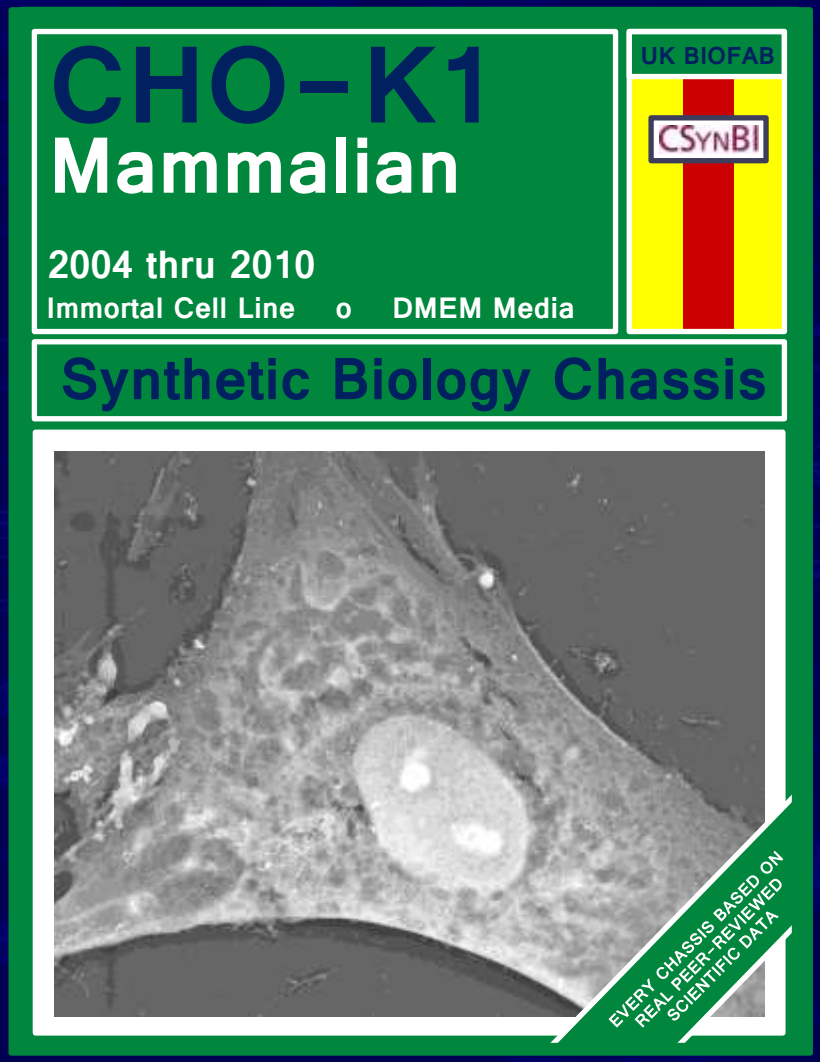
Synthetic Biology Chassis



EVERY CHASSIS BASED ON REAL PEER-REVIEWED SCIENTIFIC DATA

2nd Generation Synthetic Biology





Relevance of current chassis cells

E.coli	Advanced molecular cloning Industrial-scale application
B.subtilis	Commonly used in industry Well-understood genetic regulation
S.cerevisiae	Major industrial organism Extensively characterised
CHO-K1 cells (+ others)	Easy to use immortal mammalian cell line Good transfection efficiency

Industrial-scale biosynthesis

Ease of re-engineering

Importing function

Synthetic Biology

Create new function
Add a foreign function to a cell

What is function?

Arsenic biosensor
Biofilm detection
Synthesis of isobutanol biopetrol
Secretion of spider silk

Growth in waste water
Safe growth on human skin
Sustainable growth on desert soils
Cheap industrial-scale growth

<u>Import the easiest</u>

List of model organisms

Prokaryotes

Escherichia coli (*E. coli*) - This common, Gram-negative gut bacterium is the most widely-used organism in molecular genetics.
Bacillus subtilis - an endospore forming Gram-positive bacterium
Caulobacter crescentus - a bacterium that divides into two distinct cells used to study cellular differentiation.
Mycoplasma genitalium - a minimal organism
Vibrio fischeri - quorum sensing, bioluminescence and animal-bacterial symbiosis with Hawaiian Bobtail Squid
Synechocystis, a photosynthetic cyanobacteria widely used in photosynthesis research.
Pseudomonas fluorescens, a soil bacterium that readily diversifies into different strains in the lab.

Table of model genetic organisms

This table indicates the status of the genome sequencing project for each organism as well as whether the organism organism's biochemical pathways.

Organism	Genome Sequenced	Homologous Recombination	Biochemistry
Prokaryote			
Escherichia coli	Yes	Yes	Excellent
Eukaryote, unicellular			
Dictyostelium discoideum	Yes	Yes	Excellent
Saccharomyces cerevisiae	Yes	Yes	Good
Schizosaccharomyces pombe	Yes	Yes	Good
Chlamydomonas reinhardtii	Yes	No	Good
Tetrahymena thermophila	Yes	Yes	Good
Eukaryote, multicellular			
Caenorhabditis elegans	Yes	Difficult	Not so good
Drosophila melanogaster	Yes	Difficult	Good
Arabidopsis thaliana	Yes	No	Poor
Vertebrate			
Danio rerio	Yes	Difficult?	Good
Mus musculus	Yes	Yes	Good
Homo sapiens (Note:not a model organism)	Yes	Yes	Good

http://en.wikipedia.org/wiki/Model_organism

Also - lists of sequenced genomes

The Chassis Challenge of Synthetic Biology

Will it be possible to perform synthetic biology in diverse organisms?

Q: Can we develop universal gene-engineering protocols and tools like those used in *E.coli* and Yeast?

e.g. Electroporation of DNA

Stable insertion of DNA into host genome

Q: Do standards for synthetic biology carry between organisms?

e.g. Measurement of fluorescent proteins (GFP)

Relative measurement

Q: What role will be played by any minimal synthetic cells?

Example: Halophile energy from desalination

Halobacterium halobium

Thrives in waste brine from desalination

Engineered to produce isobutanol biopetrol from sunlight and CO_2

Provides an local source of energy for desalination

Example: Heavy-metal biosensors for water

Arsenic, Antimony, Lead

Small molecules that are expensive to detect

Natural proteins can bind these

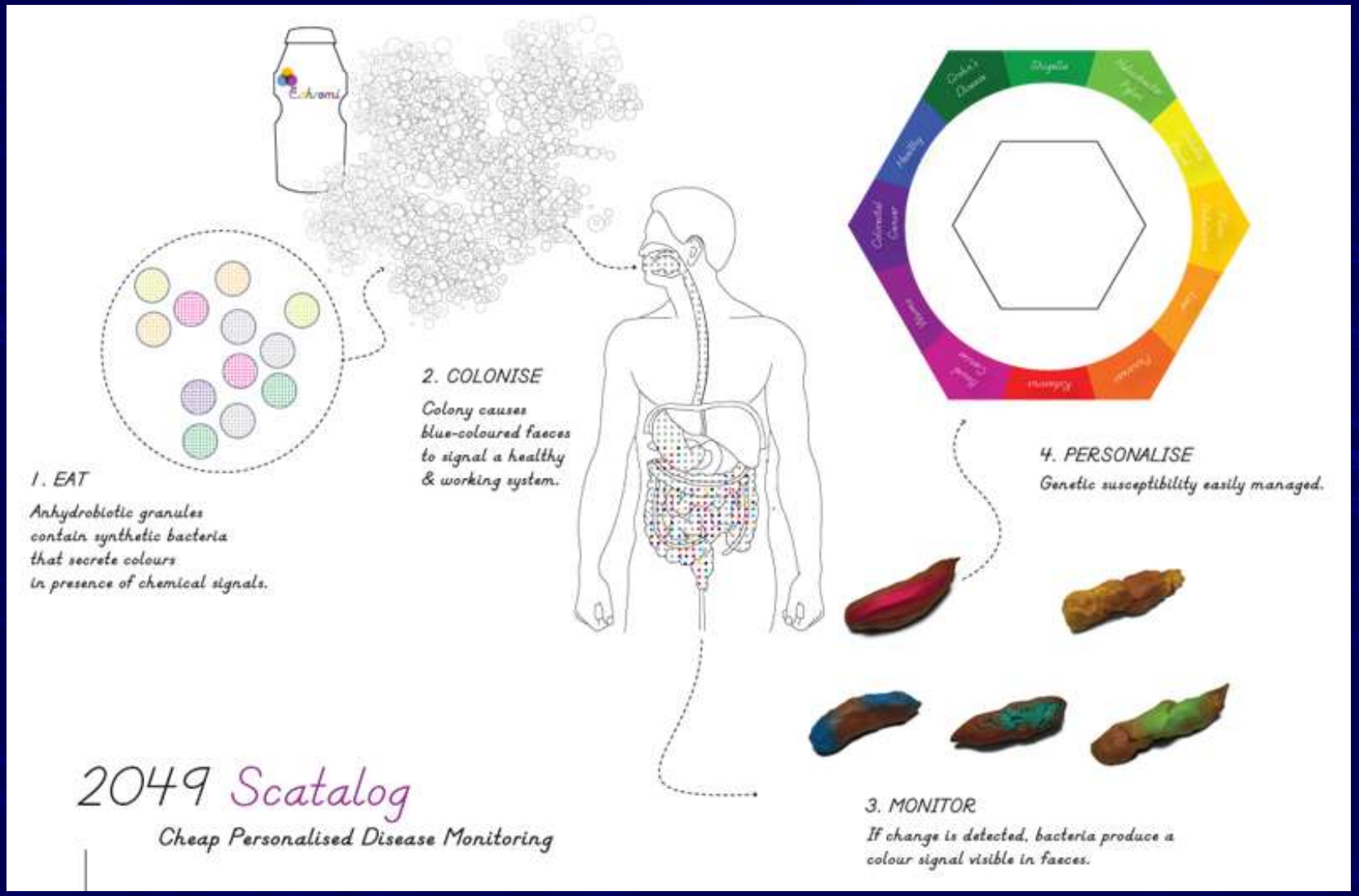
Microbial two-component signalling systems are modular

Bind – Detect – Signal

What microbes /organisms can be safely added to points in the water supply?

- Coliform bacteria – *E.coli*, *Citrobacter*
- Algae, pond weed plants

Example: Lactobacillus health biosensor



Future Chassis

Cyanobacteria	Human cells	*C.elegans*
Plant cells (Coleochaete)	*D.melanogaster*	Algae
JCVI *M.Genitalium*	Synthetic cell	Stem cells

Energy, exobiology and bioremediation: extremophiles

Halophiles	Thermophiles	Piezophiles
Lithoautotrophs	Metalotolerant	*D. radiodurans*

Medically-relevant microbes

Pathogens:	*Pseudomonas*	Tuberculosis	Plasmodia
Human flora:	*Lactobacillus*	*Candida*	*Staphylococcus*