Dark and photo fermentative hydrogen production as a route toward biomass energy generation

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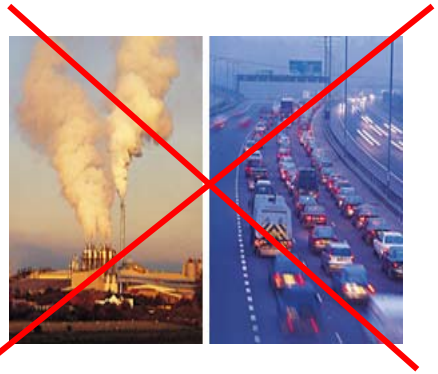
Developing renewable energy for energy/environmental crisis



Fossil fuel:
coal, oil, natural gas

Solar

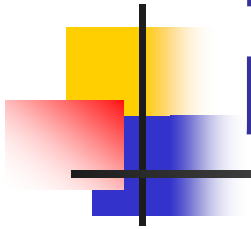
Wind

Biomass

Nuclear

Hydro

Geothermal



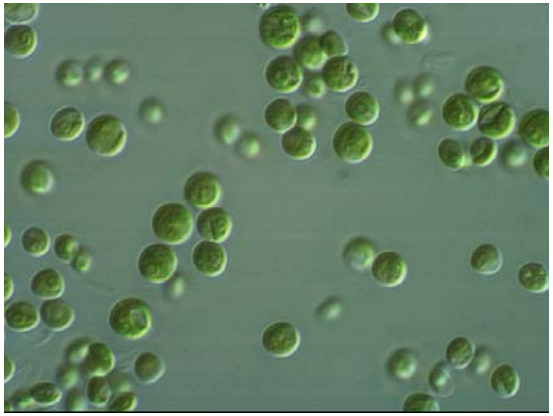
Energy that produced from biological systems

- Liquid fuels
  - Ethanol
  - Butanol
  - Biodiesel
- Gaseous fuels
  - H_2
  - Methane

Microorganisms that produce energy

CH_4

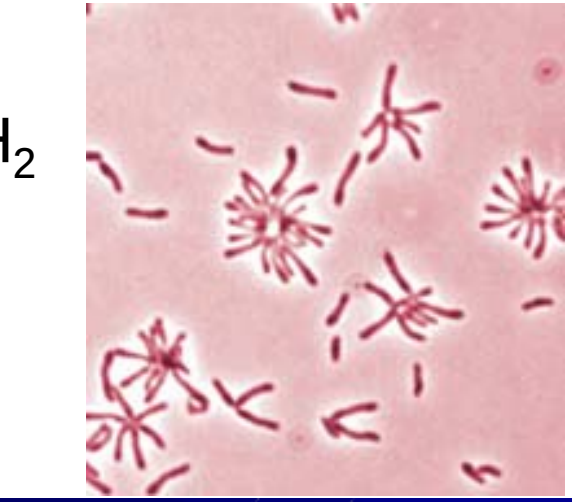
Methanococcus jannaschii



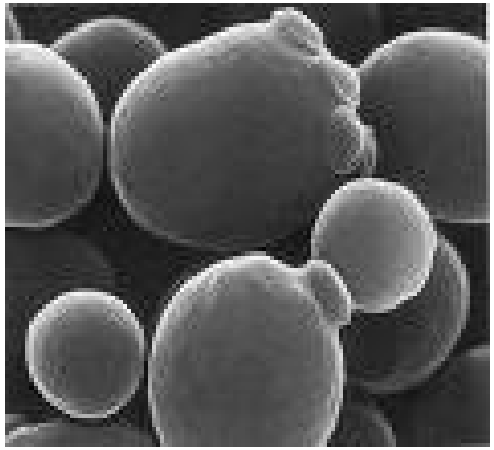
Microalgae

H_2, biodiesel

H_2



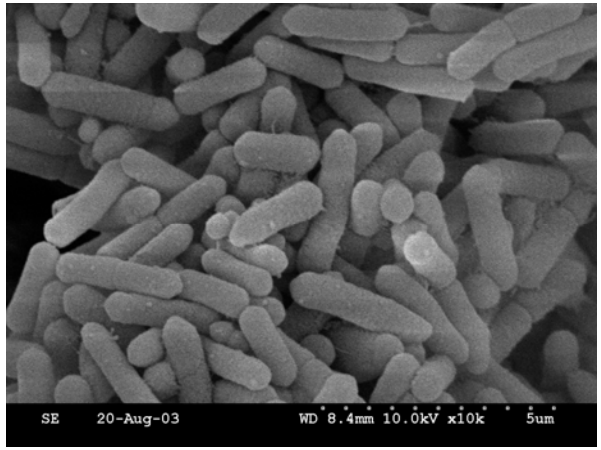
Rhodopseudomonas palustris



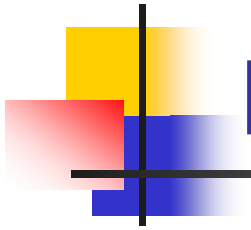
ethanol

Saccharomyces cerevisiae

H_2

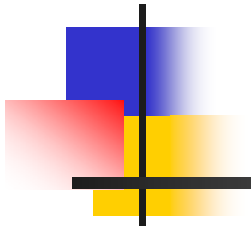


Clostridium sp.



Energy Biotechnology Platform

- Feedstock technology
  - Energy crop
  - Feedstock pretrement
    - Enzymatic hydrolysis (Cellulase, xylanase, pectinase, amylase, glucoamylase, Lipase, Protease, etc)
    - Chemical/physical pretreatment
- Energy production technology
  - Fermentation technology
  - Metabolic engineering
  - Energy product separation

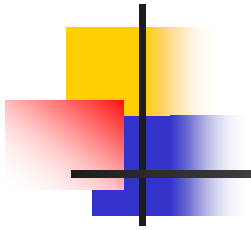


Bio-Hydrogen

Why hydrogen?

- **Clean:** CO_2 free; only water after combustion
- **High energy yield:** 122 KJ/g
- **Wide application** (i.e. fuel cell)
- **Versatile production methods**
- **H_2 is a recyclable and sustainable energy carrier**

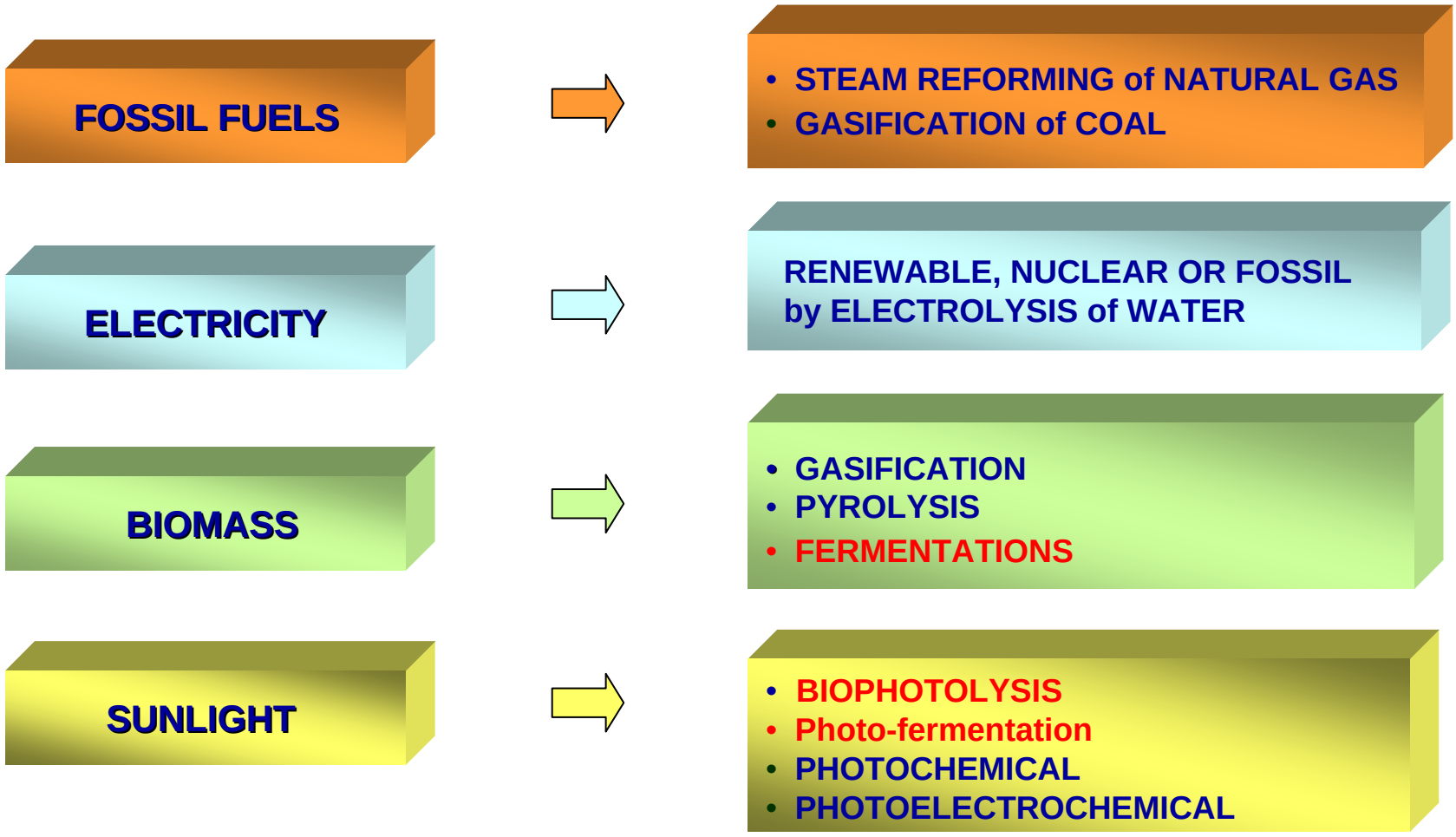
Source of Energy	KJ/g
Hydrogen gas	122
Methane gas	56
Petrol (octane, C_8H_{18})	48
Coal (carbon, C)	33
Ethanol (C_2H_5OH)	30
Methanol (CH_3OH)	23



Platform technologies of H_2 energy

- H_2 production
  - Conventional (Chemical/physical) methods
  - Biological methods (under development)
- H_2 storage and transportation
  - H_2 storage devices (compressed or liquidized or metal hydride)
  - H_2 fueling devices
- H_2 energy application
  - Fuel cell
  - H_2 ICE (internal combustion engine)

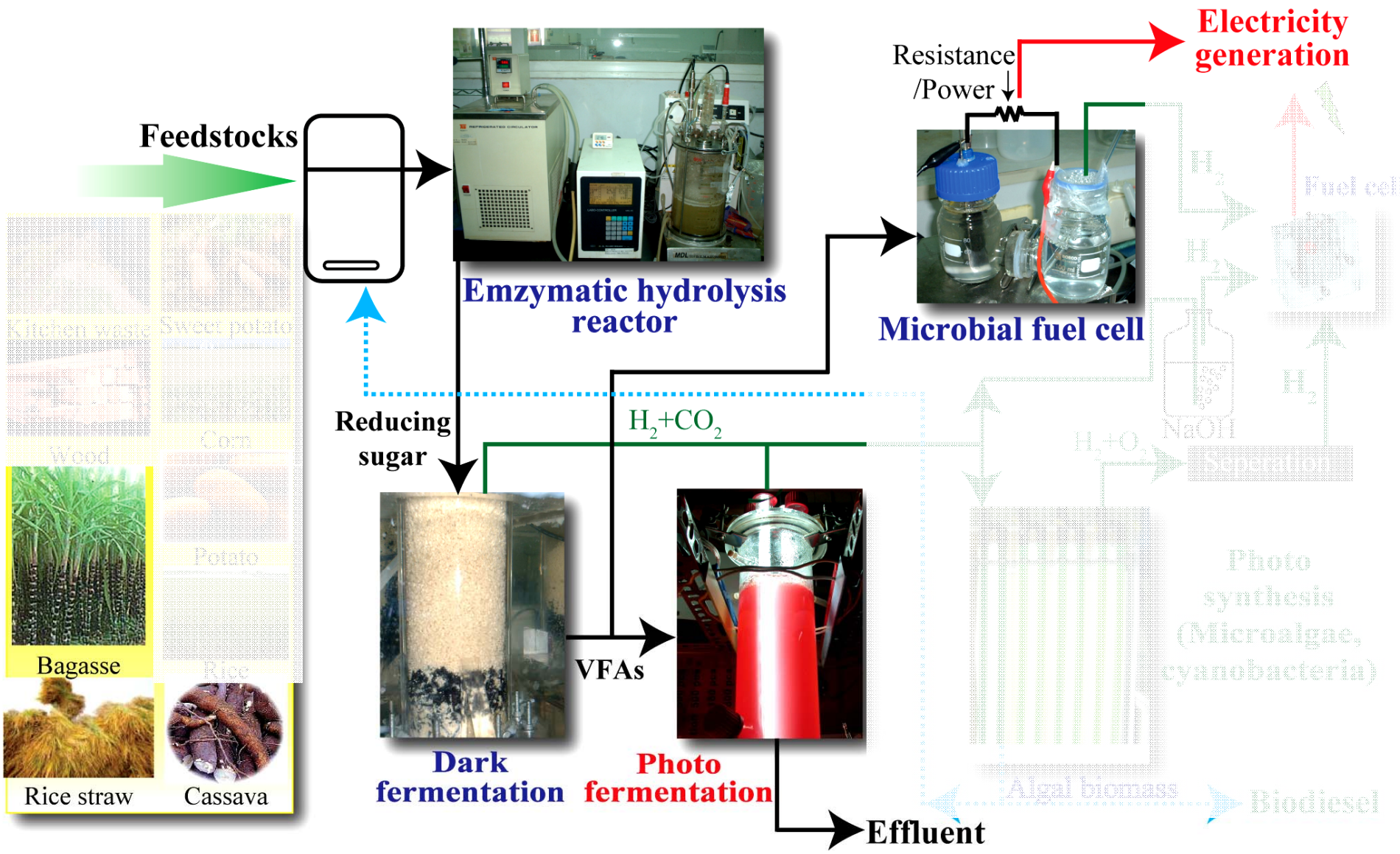
SOURCES and PROCESSES for H_2 PRODUCTION



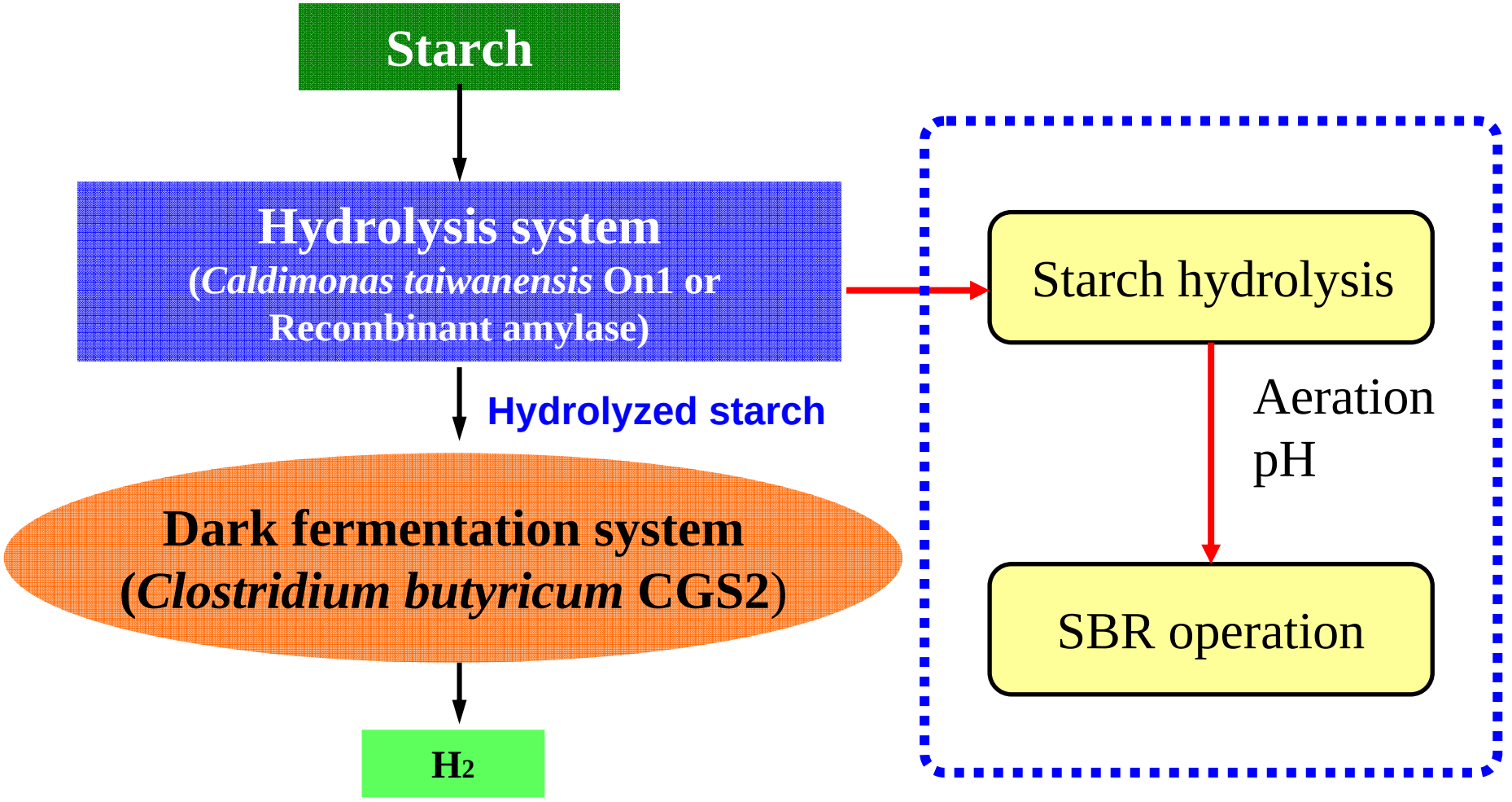
Types of bio-H_2 production

Type	Function	H_2-producing microorganism
Photosynthesis	Photolysis of water	Microalgae, cyanobacteria
Photo fermentation	Carbon source: organic substrate Energy source: light energy	photosynthetic bacteria
Dark fermentation	Carbon/energy source: organic substrate	Anaerobic acidogenic bacteria (e.g., *Clostridium* spp.)

Our vision of an integrated bioH_2 energy system

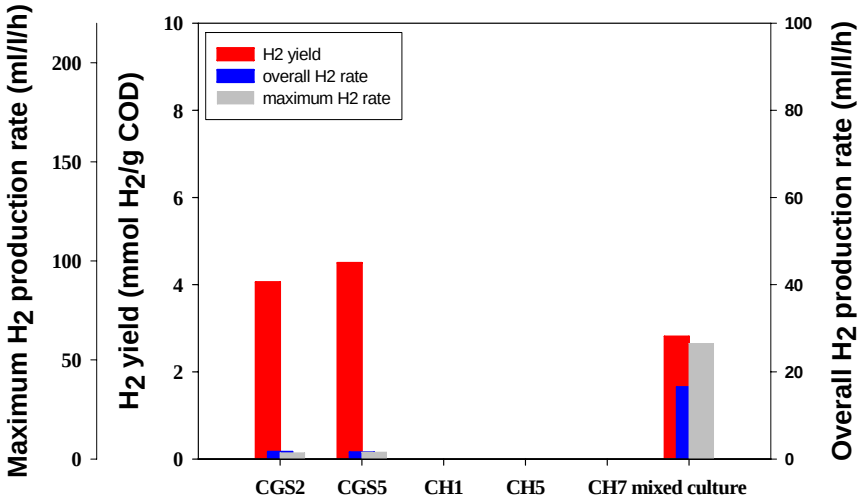


Converting starch to bioH2

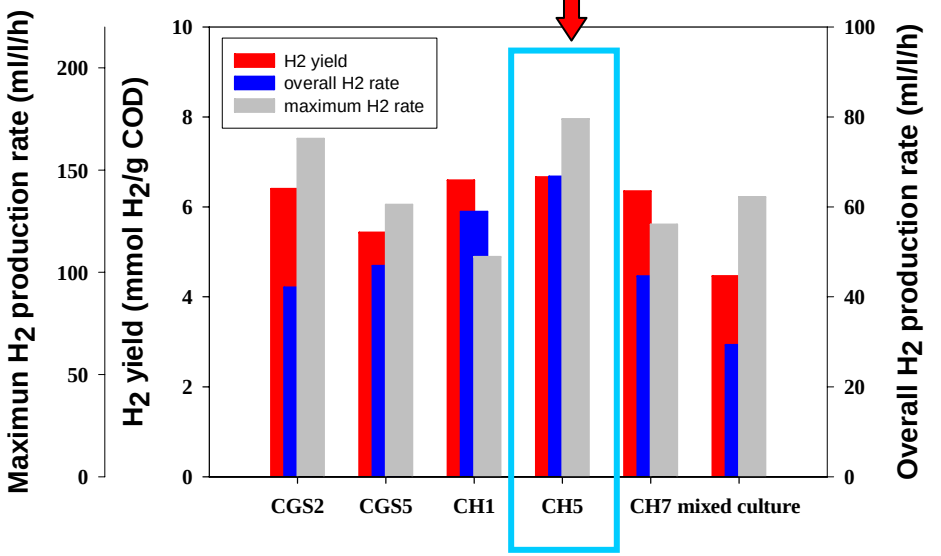


Comparison of H_2 production from original and hydrolyzed starch

H_2 production rate of 175 ml/h/l
H_2 yield of 6.66 mmol H_2/g COD.

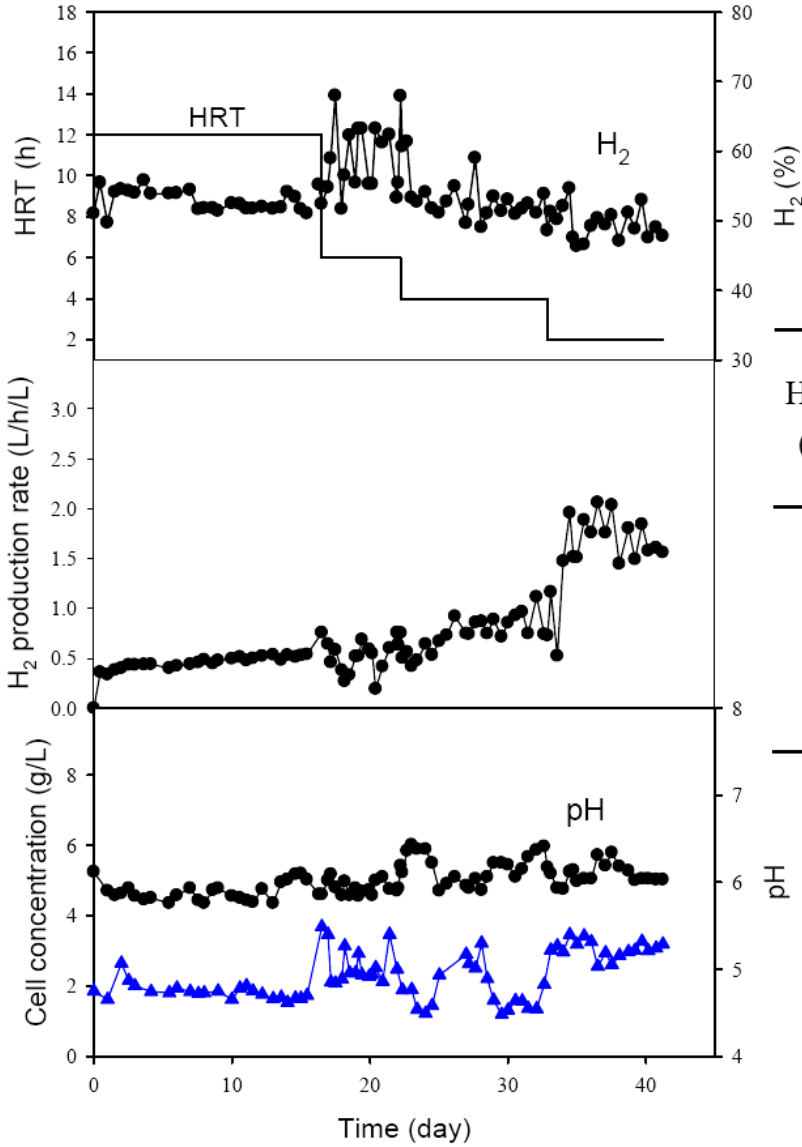


Original starch



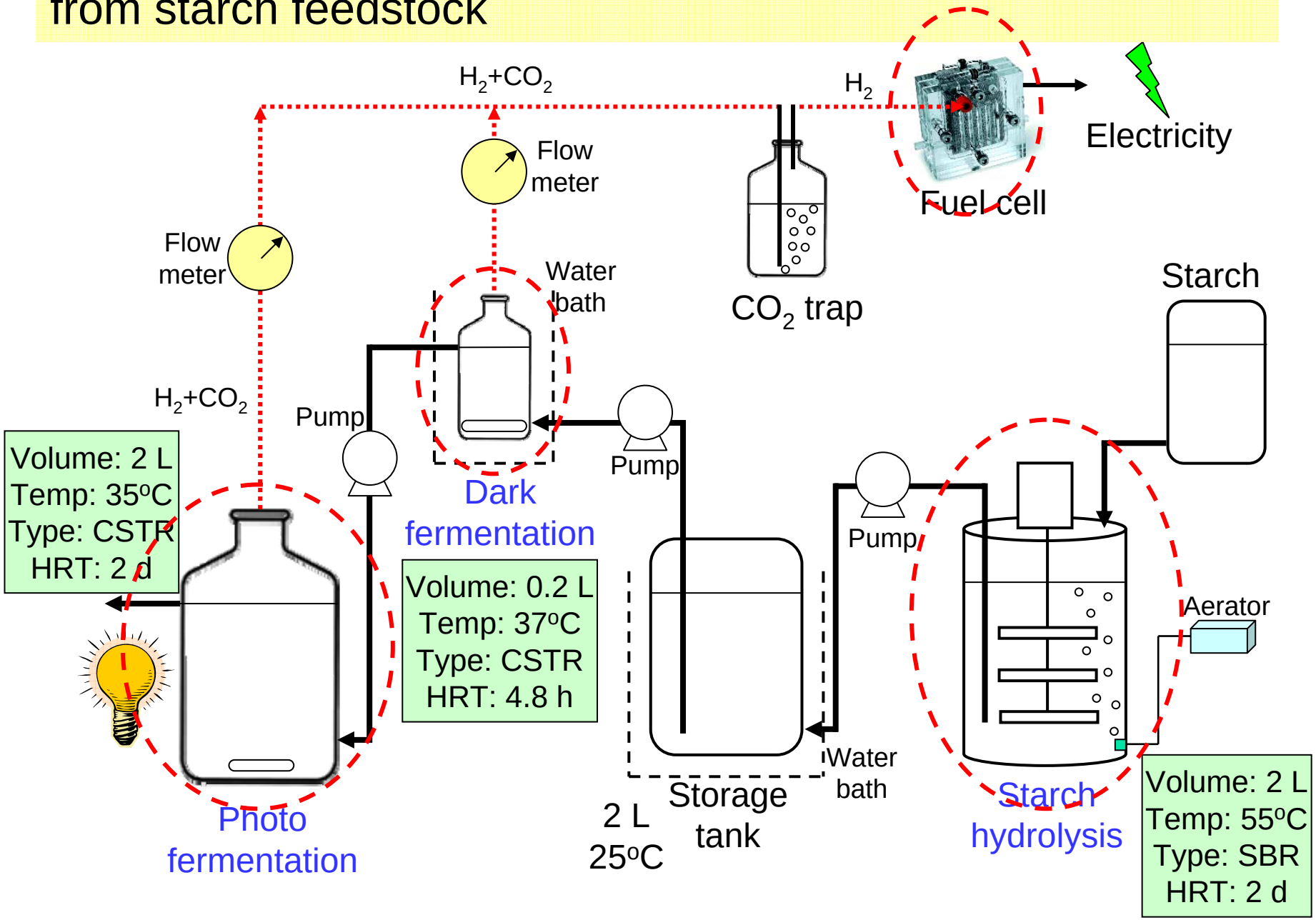
Hydrolyzed starch

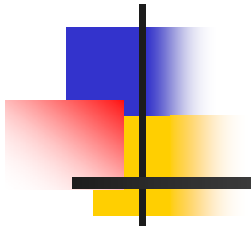
Continuous H_2 production with hydrolyzed starch



HRT (h)	Specific H_2 production rate (mmol H_2/g VSS/h)	H_2 yield (mol H_2/mol glucose)	H_2 yield (mmol H_2/g COD)	Overall H_2 yield (mmol H_2/g Starch)
12	10.2	2.03	10.56	9.63
6	10.4	1.64	8.55	9.12
4	13.1	1.41	7.34	7.83
2	21.8	1.49	7.75	8.27

Three stage bioH_2 system generating electricity via fuel cell from starch feedstock

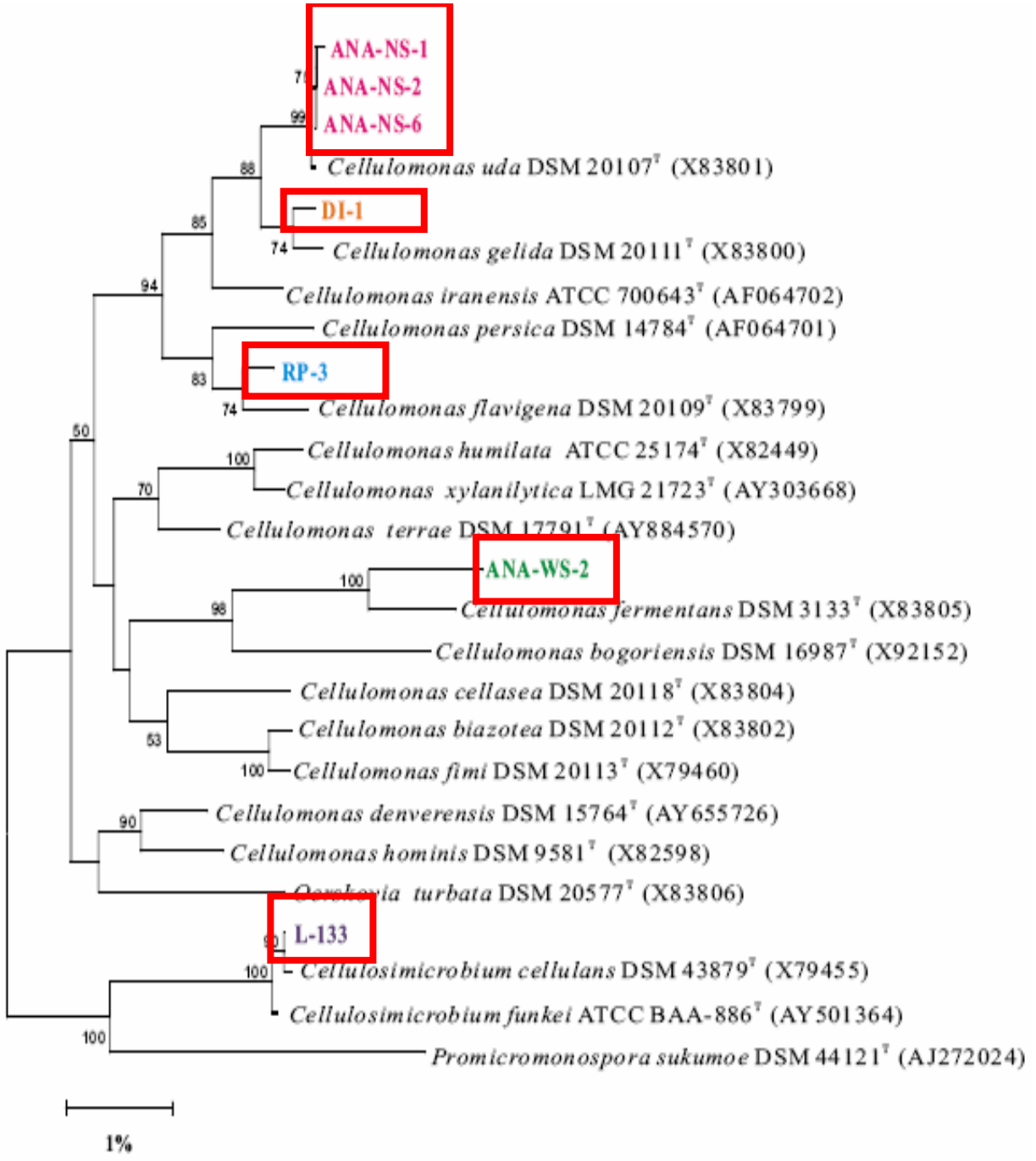




Cellulosic bioH_2

Cellulosic bioH2

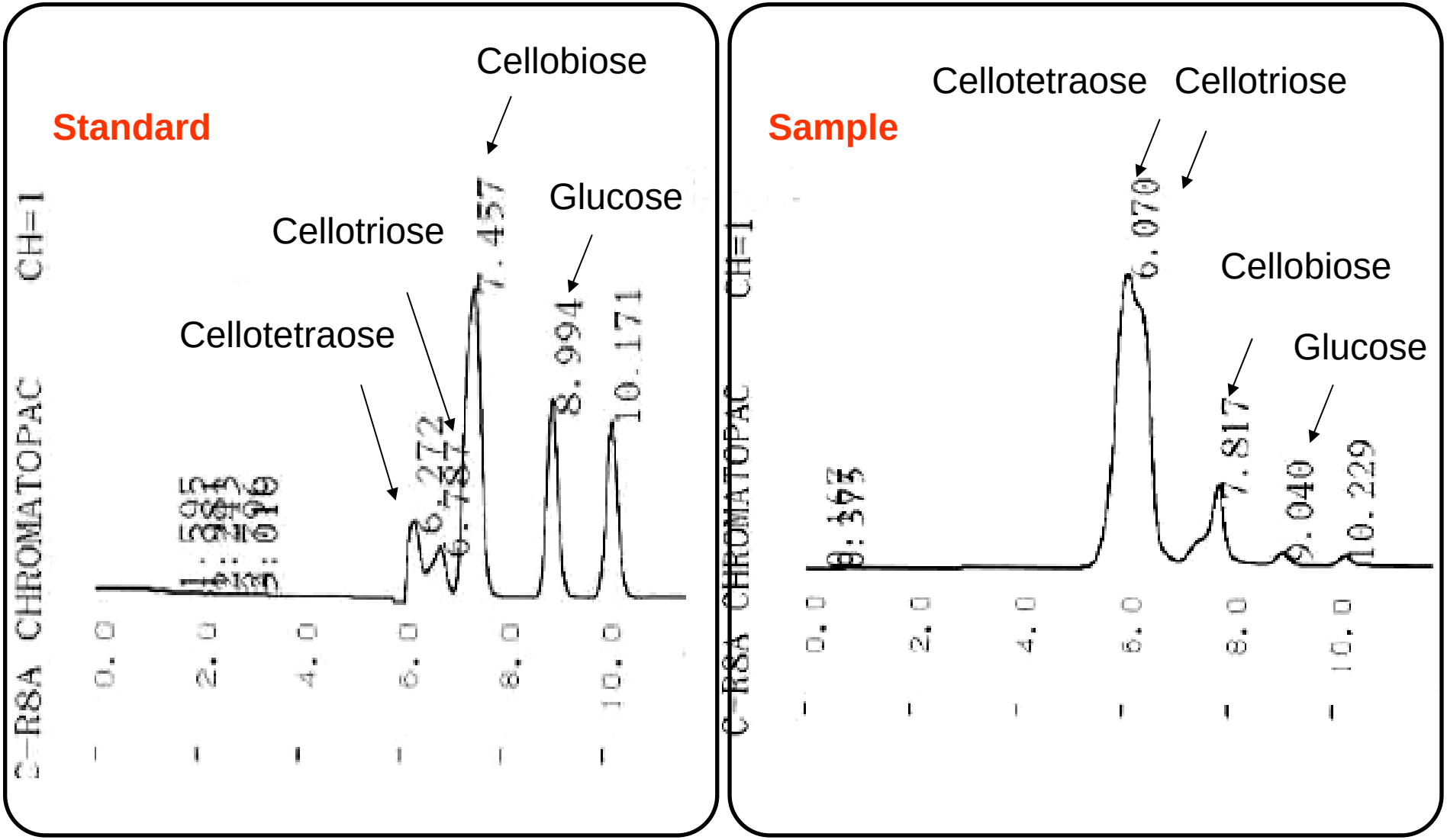
Isolating cellulase producing bacteria



Cellulose-hydrolytic bacterial strains isolated from soil

Strain	Sequence similarity
ANA-NS1	*Cellulomonas uda* DSM 20107^T 99.7%
ANA-NS2	*Cellulomonas uda* DSM 20107^T 99.7%
ANA-NS6	*Cellulomonas uda* DSM 20107^T 99.7%
ANA-FP2	*Cellulomonas uda* DSM 20107^T 99.7%
FP4	*Cellulomonas uda* DSM 20107^T 99.7%
RP-3	*Cellulomonas flavigena* DSM 20109^T 98.5%
I-1	*Cellulomonas gelida* DSM 20111^T 99.5%
ANA-WS2	*Cellulomonas fermentans* DSM 3133^T 97.8%
L-133	*Cellulosimicrobium cellulans* DSM 43879^T 99.8%

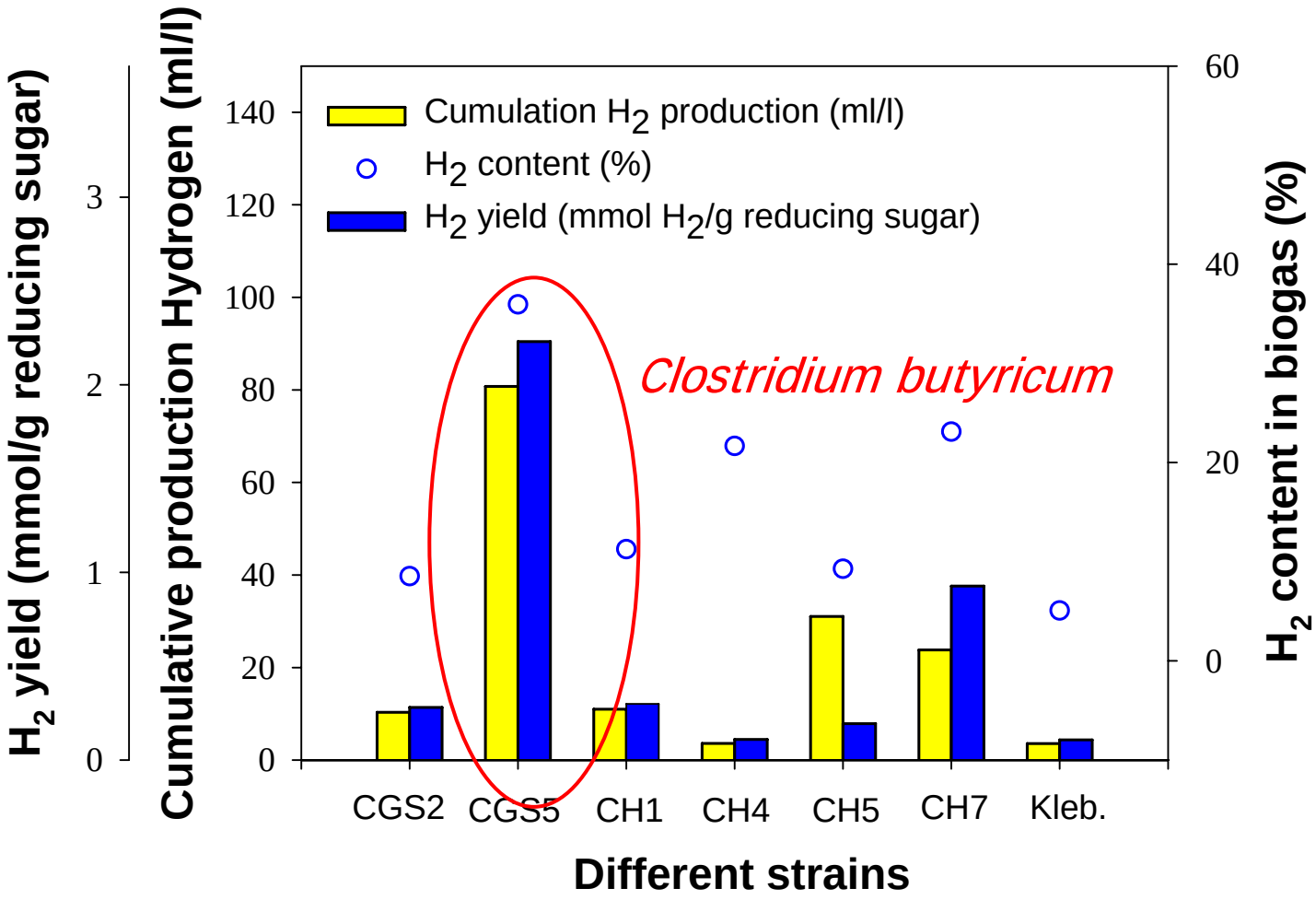
HPLC Analysis of cellulose hydrolysate



Enzyme location and activity (on xylan)

Location	Pure strain / Enzyme	Ana-NS-1	Ana-NS-2	Ana-NS-6	Ana-FP 2	FP-4	RP3	I-1	Ana-WS-2	L-133
Extra-cellular	Exoglucanase (μg reducing sugar /min)	0.899	6.591	5.292	3.894	0.799	1.698	0	0.300	0
	Endoglucanase (μg reducing sugar /min)	11.72	26.36	10.39	17.58	2.93	2.40	9.85	13.05	28.0
	Cellobiase (μg glucose/min)	0.029	0	0.019	0.014	0	0.298	0	0	0.001
	Xylanase (μg reducing sugar /min)	14.0	3.17	10.8	25.4	1.27	0	0	6.341	40.0
Intra-cellular	Exoglucanase (μg reducing sugar /min)	0	0	0	0	0	0	0	0.032	0
	Endoglucanase (μg reducing sugar /min)	0	0.056	0	0	0	0.082	0	0	0
	Cellobiase (ug glucose/min)	0	0	0	0	0	0	0	0	0
	Xylanase (μg reducing sugar /min)	0	0.200	0	0	0.133	0	0.400	0	0
Cell-bound	Exoglucanase (μg reducing sugar /min)	0	0.032	0.116	0	0	0	0	0.053	0
	Endoglucanase (μg reducing sugar /min)	0.056	0.056	0	0.028	0	0.165	0	0.140	0
	Cellbioase (μg glucose/min)	0	0	0	0	0	0	0	0	0
	Xylanase (μg reducing sugar /min)	0	0	0	0	0.267	0	0	0	0

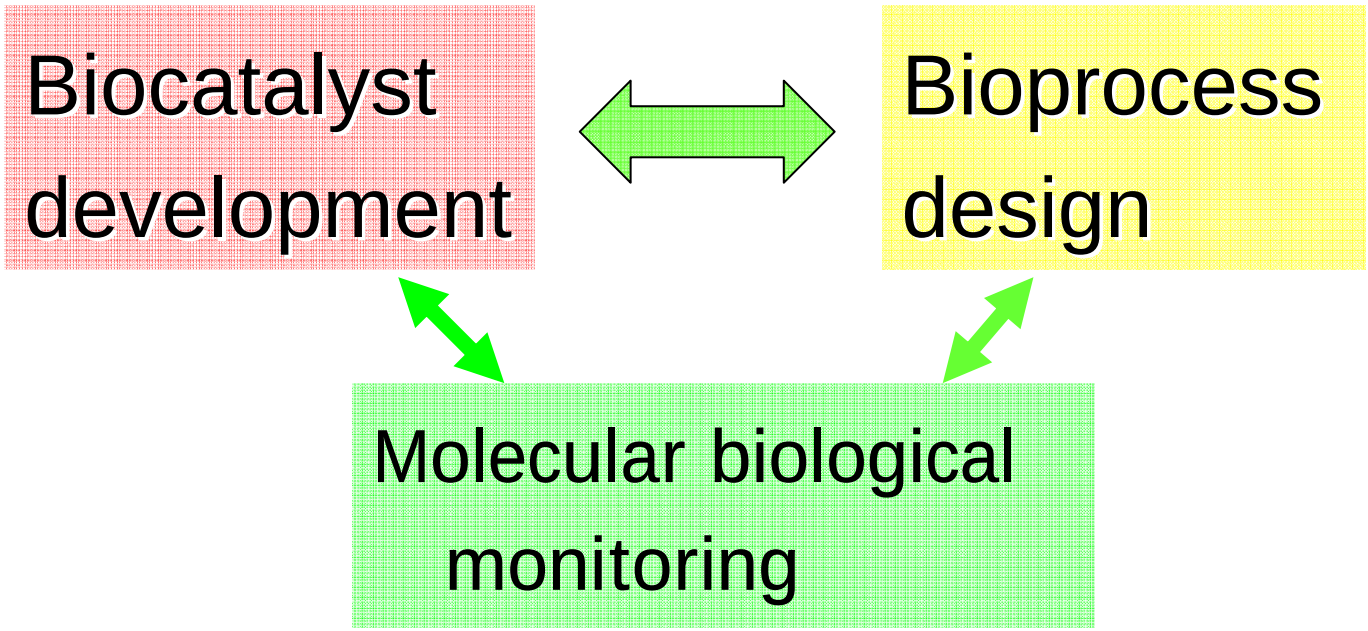
H_2 production from hydrolyzed cellulose with pure strains



How to produce H_2 efficiently?

Our approach:

Using biochemical engineering approaches to optimize and control biohydrogen production



Immobilized-cell H_2 producing system

Cells with high H_2 producing activity

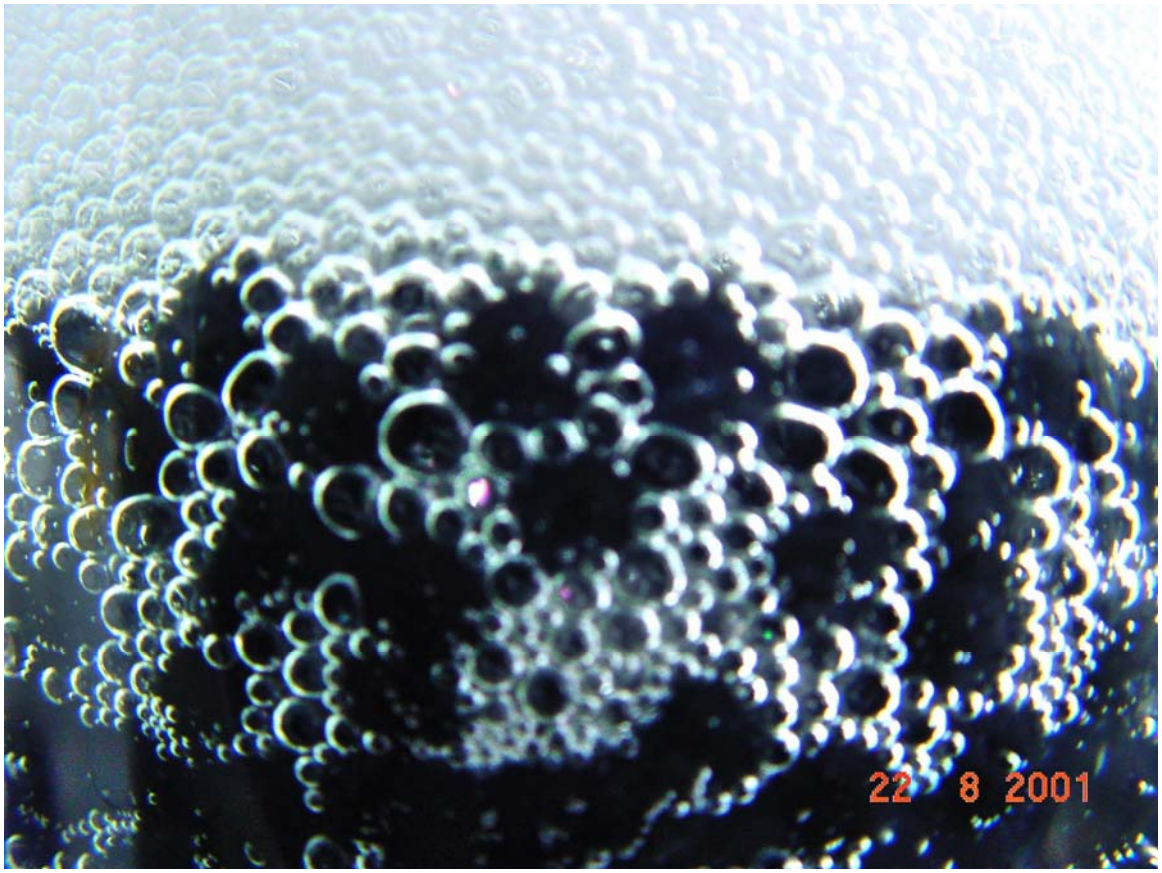
Mixed with AC and polymeric matrix

Making immobilized cells

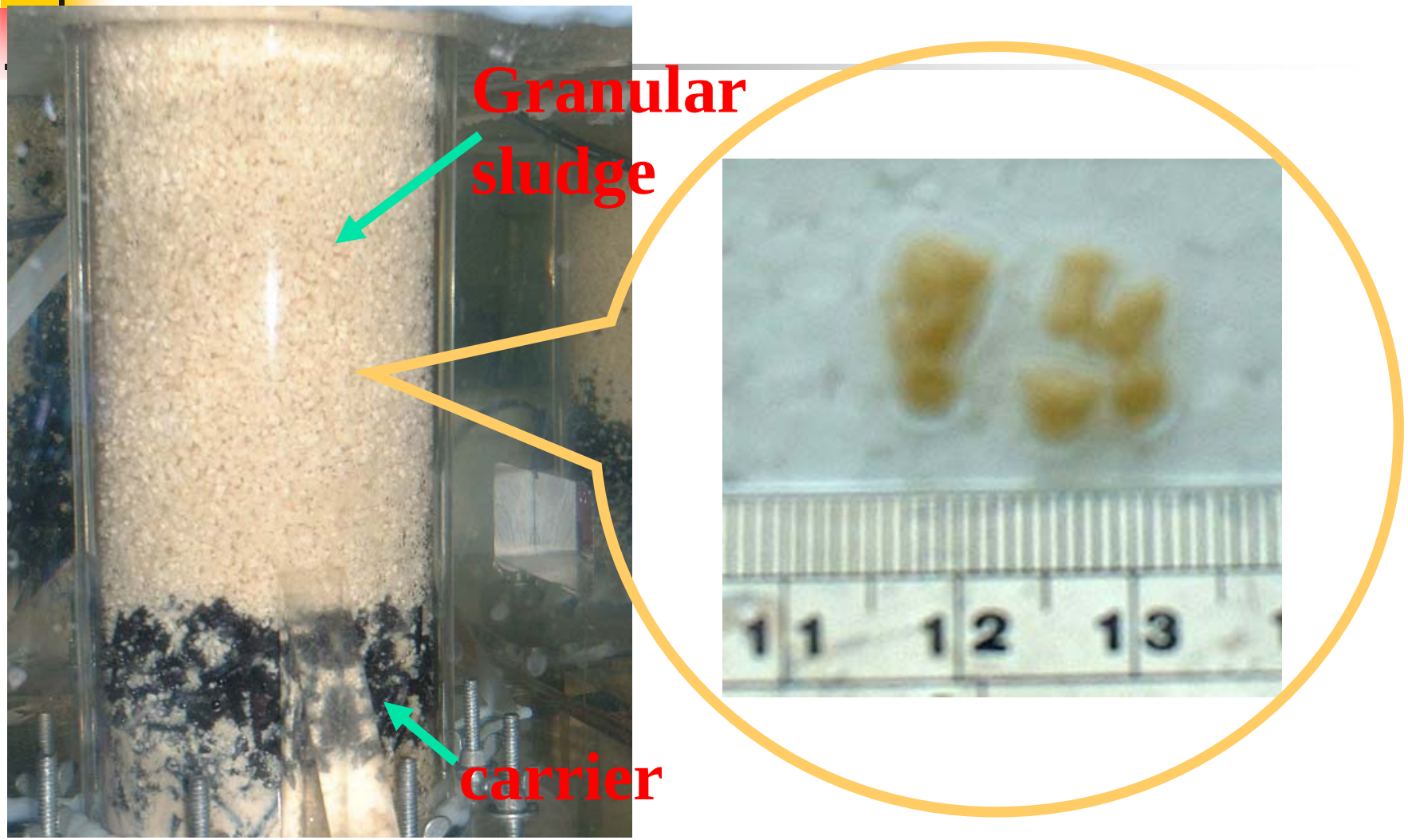
Introduced into a bioreactor

Bioreactor operation

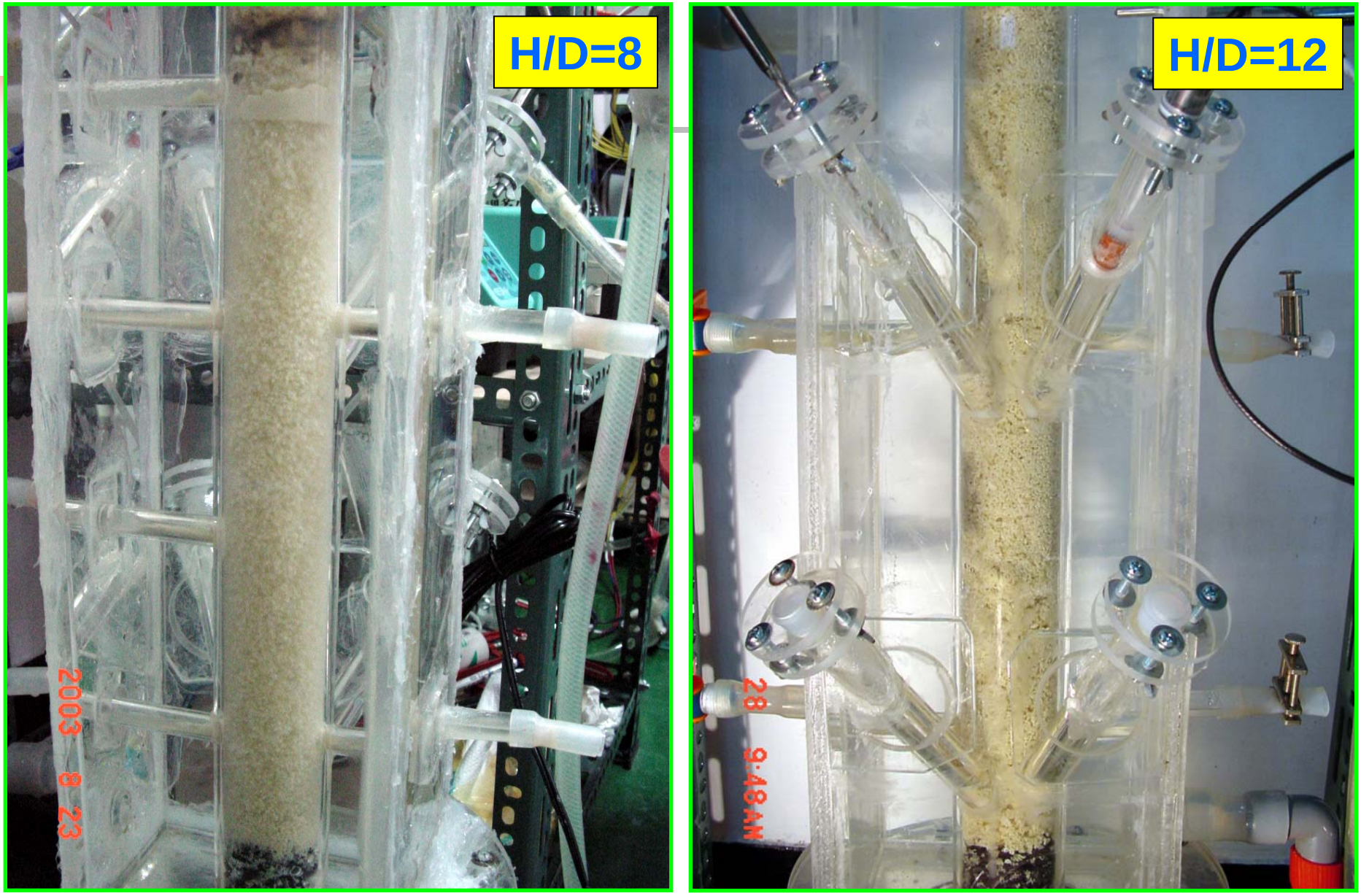
H_2 production in immobilized cells



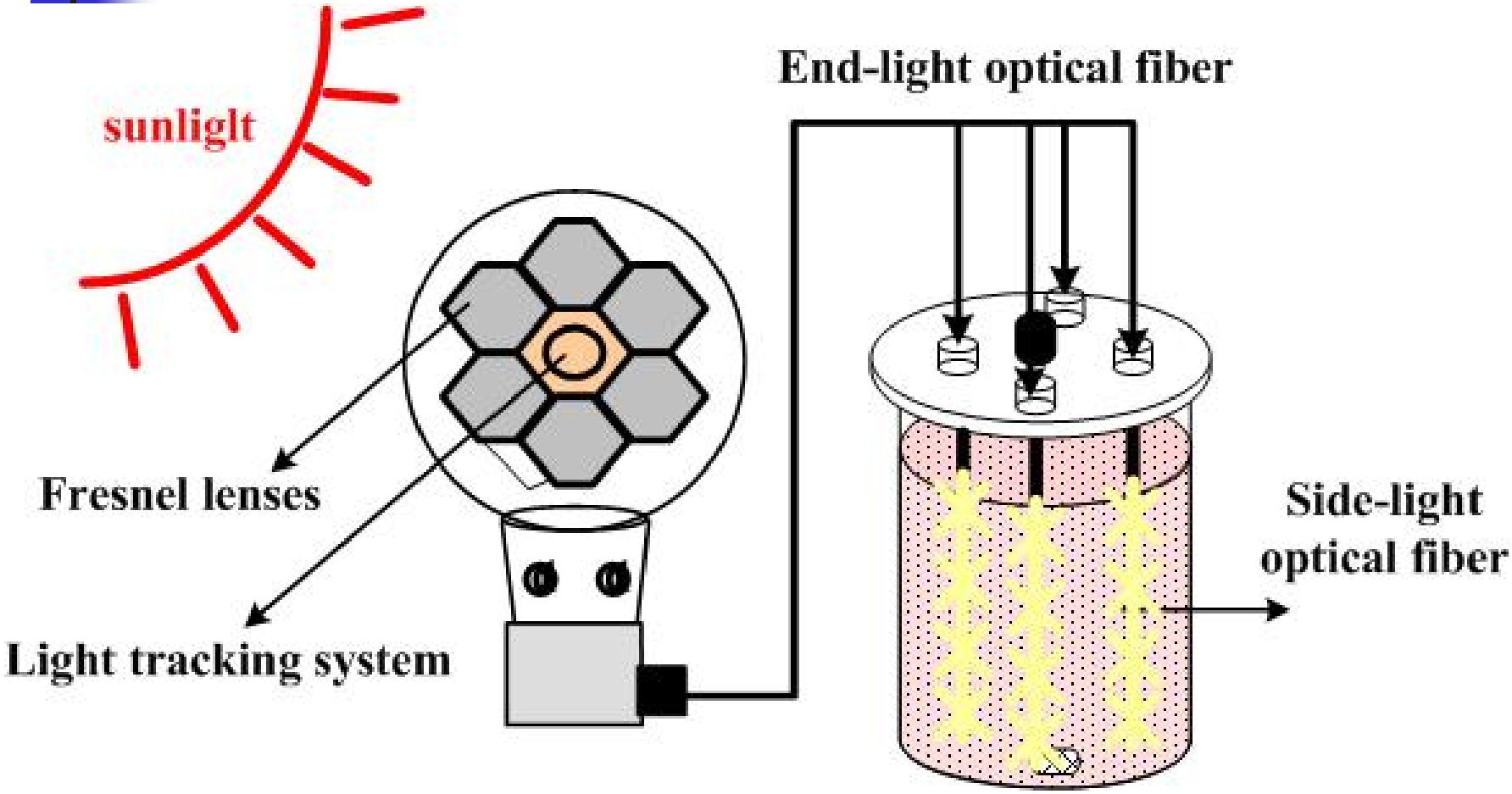
Granular sludge bed system



Picture of a CIGSB system



A solar-energy-excited optical fiber (SEEOF) system



Solar energy ➔ Hydrogen ➔ Electricity