Acetone-butanol-ethanol (ABE) production from sweet potato residue of ethanol fermentation by *Clostridia acetobutylicum* CICC8012

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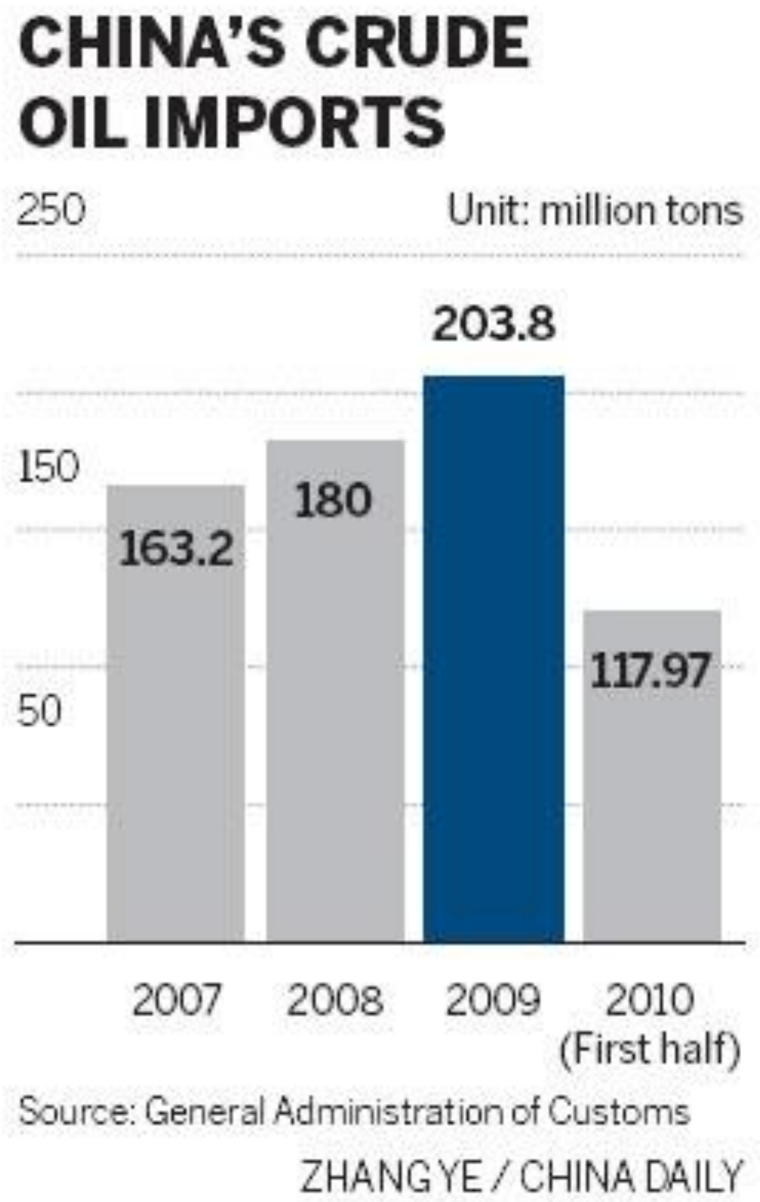
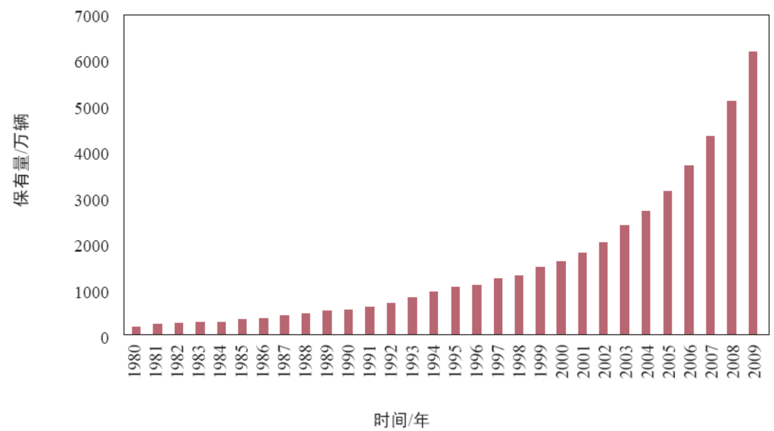
Energy and environment challenges of China



➢China's crude oil import dependence in 2009 reached **51.29** percent, exceeding the **warning line** of 50 percent for the first time.

➢China surpassed the US as the world's **largest energy consumer** in 2010.

- There are more than 100 million vehicles in China.(1 billion vehicles in the world)
- The quantity of vehicle grows rapidly, 18 million vehicles were sold out in 2010.



- Gasoline price rose from 2.3 Yuan/liter to 7.5 Yuan/liter from 2004-2010, and more expansive than that in USA.

Data from China Vehicle Emission Control Annual Report 2010

World CO_2 emission

CO_2 emissions in 2010 (million tonnes CO_2) and CO_2/capita emissions 1990-2010 (unit: tonne CO_2/person)

		Per capita emissions						
	Emissions 2010	1990	2000	2010	Change 1990-2010	Change in %	Change in CO_2, %	Change in population, %
Annex I *								
United States *	5.250	19,7	20,8	16,9	-2,8	-14%	5%	23%
EU-27	4.050	9,2	8,5	8,1	-1,1	-12%	-7%	6%
EU-15 **	3.150	9,1	8,8	7,9	-1,2	-13%	-5%	9%
- Germany	830	12,9	10,5	10,0	-2,9	-22%	-19%	4%
- United Kingdom	500	10,2	9,2	8,1	-2,2	-21%	-15%	8%
- Italy	410	7,5	8,1	6,8	-0,7	-9%	-3%	7%
- France	370	6,9	6,9	5,9	-1,0	-15%	-5%	11%
- Poland	320	8,2	7,5	8,3	0,1	1%	2%	1%
- Spain	290	5,9	7,6	6,3	0,4	7%	26%	18%
- Netherlands	180	10,8	10,9	10,6	-0,2	-2%	9%	12%
Russian Federation	1.750	16,5	11,3	12,2	-4,2	-26%	-28%	-4%
Japan	1.160	9,5	10,1	9,2	-0,4	-4%	0%	4%
Australia	400	16,0	18,6	18,0	1,9	12%	46%	30%
Canada	540	16,2	17,9	15,8	-0,4	-2%	20%	23%
Ukraine	310	14,9	7,2	6,9	-8,0	-54%	-59%	-12%
Non Annex I								
China	8.950	2,2	2,9	6,8	4,6	205%	257%	17%
India	1.840	0,8	1,0	1,5	0,8	100%	180%	40%
South Korea	590	5,9	9,7	12,3	6,4	109%	134%	12%
Indonesia	470	0,9	1,4	1,9	1,1	126%	194%	30%
Brazil	430	1,5	2,0	2,2	0,7	51%	96%	30%
Mexico	430	3,7	3,8	3,8	0,1	4%	39%	35%
Saudi Arabia	430	10,2	12,9	15,6	5,3	52%	159%	70%
Iran	400	3,7	5,2	5,4	1,6	44%	94%	35%
South Africa	380	7,3	6,9	7,6	0,3	4%	42%	36%
Taiwan	270	6,3	10,1	11,1	4,8	77%	118%	23%
Thailand	240	1,6	2,7	3,4	1,8	115%	160%	21%

China is the biggest CO_2 emitter

- China totally energy consumption almost depends on domestic supply
- Petroleum resource is extremely limited. Shortage of oil supply is becoming serious challenge for the economy and society smoothly develop.
- Reducing GHG emission is another challenge issue for China.

- Challenges drive China to develop the production of bio-liquid fuel (such as ethanol and butanol) to substitute gasoline

- Benefit : reducing GHG emission
  economy growth healthily
  society development harmony and sustainable

Advantages of butanol

As a kind of biofuel, butanol is superior to ethanol

- More hydrophobic property
- Higher energy density
- Allows the use of existing pipeline infrastructures for transportation
- Can be mixed with gasoline at any ratio as vehicle fuel

Feedstocks for butanol production

Feedstocks are critical for economically feasible butanol fermentation

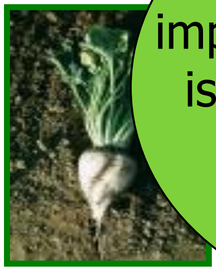
Corn wheat

It is not sustainable especially for China which has the largest population to feed.

Starch crops

Canna e

It is urgent to find low-cost energy feedstocks such as crop residues and municipal or industrial solid waste



Sugar beets sugar cane sweet sorghum

China is still a sugar import country , there is not enough sugar crops for sugar production

Cellulosic materials

Woody fiber grasses

- According to the Chinese government planning, more than 5M tons ethanol will be produced from sweet potato in 2020
- This will emerge number of different fermentation residues
- These residues are presented in the form of solids and liquids and have to be processed for avoiding the pollution of environment.

Major components of the dry sweet potato residue of ethanol fermentation

Which can be hydrolyzed to monomeric sugars, and then be used as substrates for the butanol fermentation.

Non-starch polysaccharides	protein	cel[illegible]
30-40%	5-15%	20-30%

Issue of butanol production from sweet potato residue of ethanol fermentation

- Residues must be hydrolyzed to simple sugars for butanol fermentation using both physical-chemical pretreatment and enzymatic hydrolysis .

- Unfortunately, during acid hydrolysis, the microbial **inhibitors** are generated such as furfural, hydroxymethyl furfural, acetic, ferulic and glucuronic.

Research Progress

1.Residues characterization

glucose	21.41%
xylose	1.96%
galactose	4.09%
arabinose	0.92%
initial pH	3.7
average moisture content	71.50%
protein content	4.70%
ashes	5.61%
fibers	6.10%
lipids	0.60%
C	45.61%
H	6.24%
N	3.16%
Na	1274(ug/g)
K	4321(ug/g)
Ca	876(ug/g)
Mg	586(ug/g)
P	1489(ug/g)
Cu	10.1(ug/g)
Mn	15.2(ug/g)
Zn	6.9(ug/g)
Fe	49.1(ug/g)

2. Effects of sulfuric acid pretreatment conditions (dilute sulphuric acid concentration and time) on ABE fermentation

Dilute sulphuric acid concentration %（v/v)	0.5			1			1.5			2			0
Pretreatment time (min)	15	30	45	15	30	45	15	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[Total sugar] initial (g/kg)	21.43	21.94	23.15	31.54	42.37	44.18	39.25	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[Reducing sugar] final (g/kg)	4.52	4.98	4.82	15.24	16.87	18.75	19.54	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[Butanol] final (g/kg)	1.902	2.021	1.922	2.926	3.749	2.988	1.543	1.134	[illegible]	0.735	0.602	0.601	1.38
[ABE] final (g/kg)	3.17	3.26	3.15	4.14	5.71	4.98	2.53	1.89	1.88	1.05	0.94	0.91	2.61
Rate of non-fermentable sugars %	21.09	22.7	20.82	16.61	16.21	26.6	49.78	63.52	86.17	90.8	91.96	92.61	17.97

The reason for the higher sulphuric acid concentration pretreatment the poorer ABE production may have been the presence of **inhibitory** chemicals in the sweet potato residues hydrolysate and need further research.

3.Effects of different detoxification methods on the ABE fermentation

[Glucose]	[illegible]	[illegible]	[illegible]	[illegible]
[Xylose]	[illegible]	[illegible]	[illegible]	[illegible]
[Galactose]	[illegible]	[illegible]	[illegible]	[illegible]
[Total reducing s	[illegible]	[illegible]	[illegible]	[illegible]
[Total reducing su	[illegible]	[illegible]	[illegible]	[illegible]
[Batanol] final (g/kg)	1.77	3.75	[illegible]	3.716
[ABE] final (g/kg)	2.89	6.14	8.92	5.56

It is reported that 9.3 g/L of ABE were produced by *C. berjerinckii* BA101 from the XAD-4 treated sulfuric acid treated corn fiber hydrolysate, and some fermentation inhibitors may still be present after treatment (Qureshi et. al., 2008). Ranjan and Moholkar reported that 1.6 g/L butanol and 0.12 g/L ethanol were fermented from acid-assisted enzyme hydrolysate of rice straw by *C. acetobutylicum* MTCC 481 after 120h of fermentation.

Three methods used in the present study resulted in production of varied amount of ABE solvents from the sweet potato residues which were treated with 1% v/v sulfuric acid at 121 ℃ for 30 min.

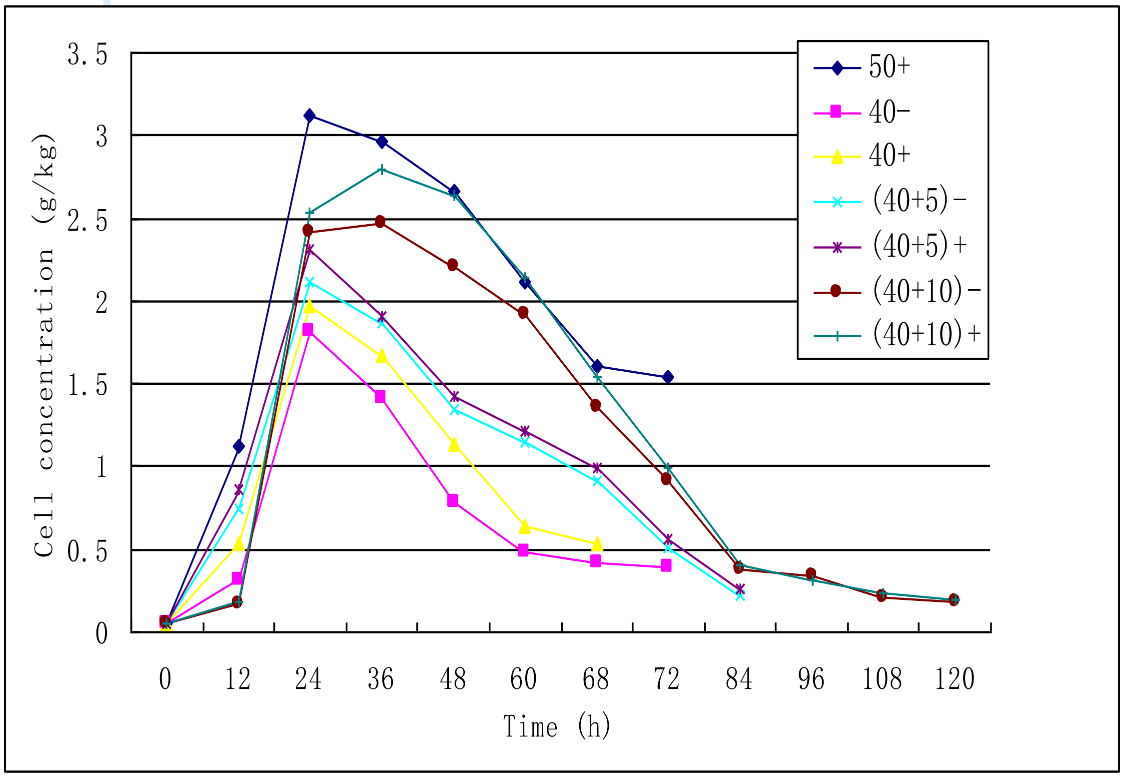
4. Study of ABE fermentation using detoxified hydrolysate with/without adding nutrients and glucose

[Total reducing sugar] initial (g/kg)	50	40		45		50	
[Glucose] initial (g/kg)	50	0	0	5	5	10	10
[sugar of hydrolysate] initial (g/kg							
Nutrition							
[Total reducing sugar] final (g/kg							
[Glucose] final (g/kg)							
verage ABE productivity rate (g /k							
Fermentation time (h)							
[Butanol] final (g/kg)	9.62	6.02	6.21	6.49		6.51	6.55
[ABE] final (g/kg)	13.19	8.57	8.82	9.14	9.28	9.31	9.44

The total ABE production that attained in the broth with hydrolysate was much less than that achieved in the control broth. This may due to the acid hydrolysis, which generated a complex mixture of microbial inhibitors

No mater what kind of medium used in fermentation, if the total initial sugar concentration of hydrolysate was above 45 g/kg, the yield of ABE and productivity was below 0.22 g ABE/g sugar and 0.12 g/kg/h.

Cell growth of ABE fermentation using detoxified hydrolysate with/without adding nutrients and glucose



- In the presence of nutrition cell growth was slightly improved
- It is reported that only 1.67 g/L of *C. beijerinckii* BA101 was detected in corn fiber hydrolysate treated with Ca(OH)2 at 48h (Ezeji et. al., 2007).

+:adding nutrients; -: no nutrients; 5/10: glucose concentration, g/L

Conclusions

- ➢Our research found a possible way of using sweet potato residues as a potential feedstock for ABE production.
- ➢According to the results, it was observed that 8.92 g/kg ABE solvents could be produced from $Ca(OH)_2$ over-limed sweet potato hydrolysate in 72 h, and the yield, productivity, total reducing sugar and rate of non-fermentable sugar were 0.12 g/kg/h, 0.22 g ABE/g sugar, 4.46 g/kg and 10.82 %, respectively.
- ➢Compared with dialysis and XAD-4 absorption, $Ca(OH)_2$ over-liming could remove fermentation inhibitors in hydrolysate medium.
- ➢ We also demonstrated ABE production and cell growth was slight improved with supplement of glucose and nutrients in medium.

Thank you very much for attention!