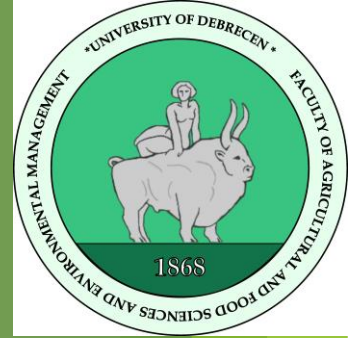




2nd INTERNATIONAL CONFERENCE
on Sustainable Solid Waste
Management



Feather waste recycle for Biogas production

Dr. Lili Mézes

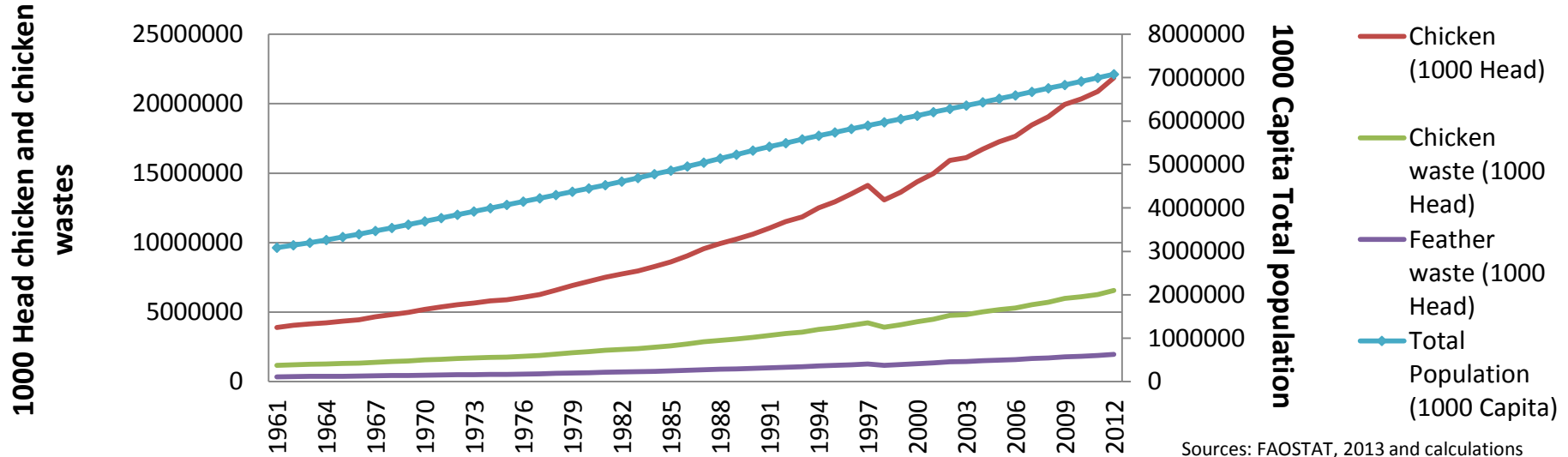
Institute of Water and Environmental Management, Faculty of
Agricultural and Food Sciences and Environmental
Management,

University of Debrecen, Hungary

Athens, 12.06.2014

Problems

World chicken and chicken waste production and population dynamics



- ▶ Modified constitutional law (1576/2007/EK decree, European Parliament)
 - => disable the feather utilization as feedstuff and depose to the landfill,
 - => innovative developments and methods are needed
- ▶ Feather waste recycling with anaerobic digestion
- ▶ Keratin-content of feather (90%) can be difficultly degraded



Objectives

- ▶ to determine the efficiency of enzymatic pre-treatment of chicken feather waste
- ▶ to apply pre-treated feather as raw material for biogas production
- ▶ to optimize the most effective treatment ratios
- ▶ and methane potential of feather waste



Materials and methods

I. Pre-treatment of feather waste

1. Preparation and analysing of slaughterhouse chicken feather:

A, Autoclaving (100° C, 30 min) => bacterial pre-treatment

B, Autoclaving and drying (80° C, 10 h) (DM, oDM content) and grinding (2-5 mm sieve) => C-, N-content analysing.



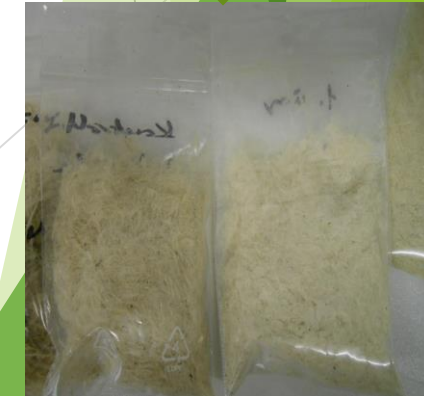
Raypa type autoclave



Grinder
(WARING®, Snijders)



Elementar VARIO EL universal
analyser



Materials and methods

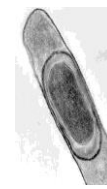
I. Pre-treatment of feather waste

2. Feather hydrolysis:

- Inoculation with *Bacillus licheniformis* KK1*:
4,55x10⁶ cell/ml (10 ml) except of the control experiment
 - produce extracellular protease, incubation (42 °C, 200 rpm) (Heidolph Unimax 1010)
 - pH optimum (7.2) setting with Tris-HCl
 - Medium: LB broth (MILLER et al. 1972)
- with 1:2 (w/v) feather: water ratio (0.67 kg whole feather: 1.33 l deionized water)
- in four 3 litre glass bottles for 5 days
- in a 45 l water bath (Memmert), triplicate.
- agitation (air pump) was controlled by flow meter (1.5 l/min air flow).

pH measuring: Hanna HI2550 multifunctional pH/ORP/temperature/EC/TDS/NaCl device

*isolated by Kovács et al., 2002; Perei et al., 2004; Bálint et al., 2005



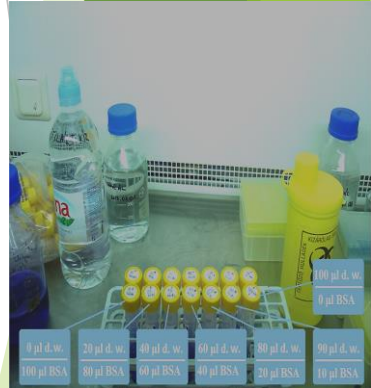
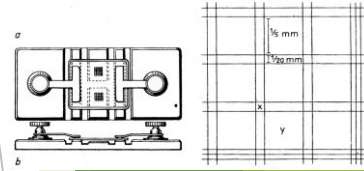
Materials and methods

I. Pre-treatment of feather waste

3. Analysis of feather degradation efficiency

I.

- **Cell counting:** in Burker chamber with Alpha BIO-3CCD light microscope
- **Determination of the protein concentration with Bradford (1976) method:**
 - 4 samples per day until 5 days.
 - Bacteria culture was filtrate with centrifuge (10000 rev/min for 20 min)
 - Absorbance of solutions was measured photometrically with Sacoman Athelie Junior spectrophotometer (2 min and 1 hour) at 595 nm
 - Calibration curve: Standard: bovine serum albumin (BSA)



3. Analysis of feather degradation efficiency II.

- Keratin degradation rate (%) was calculated from protein content of raw chicken feather and protein concentration of liquid culture filtrate.
- Determination of chemical oxygen demands:
 - ▶ After centrifuge with PF-10 compact photometer (Macherey-Nagel) with 0-15,000 mg/l sCOD test solution at 620 nm after 148 °C, 2h destruction.
- ▶ Solubilisation degree of feather (%) calculation was based on method described by Forgács et al. (2013).



Materials and methods

II. Biogas production

1. Biodegradation laboratory: Anaerobic digestion; Batch tests

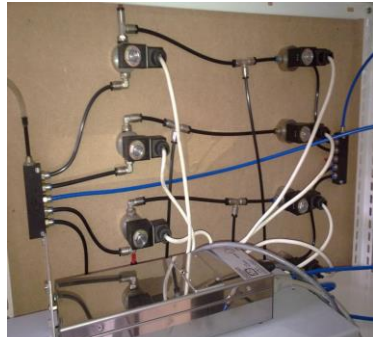
Biogas washing
(gas-washer bottle),
and dewatering
(cryogenic instalment)
system



Biogas quantity
analysis

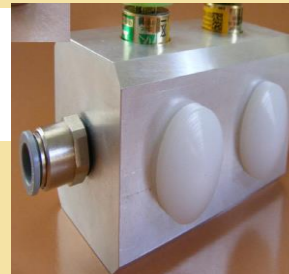


Cyclic, doubled
valve-system



Biogas quality analysis

CH_4
 CO_2



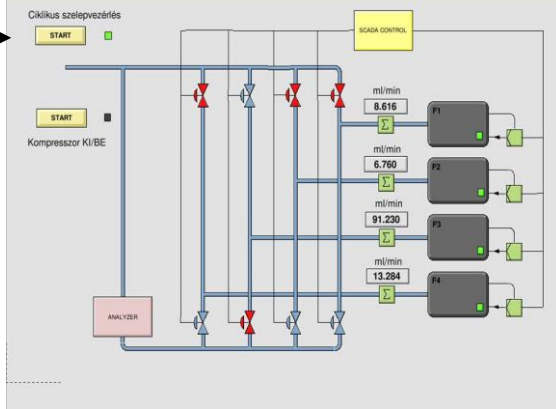
H_2S
 NH_3



Stainless steel digesters (6 l volume)
(mixing, heating, pH controlling) in incubators



Controlling system



Materials and methods

II. Biogas production

2. Batch digestion: raw materials and experiments:

Mesophilic conditions (38°C), 5 l volume, 30 days



Table 1. Experimental settings:

Digester	Corn silage (kg)	Mesophilic liquid digestate (kg)	Cattle slurry (kg)	Pre-treated feather (%)
1 (Control)	0.2	2.2	2.6	0
2	0.2	2.2	2.6	5
3	0.2	2.2	2.6	10
4	0.2	2.2	2.6	20

Results and discussion

I. Pre-treatment of chicken feather

Table 2. Quality parameters of feather samples:

Number of samples	Carbon-content (%)	Nitrogen-content (%)	DM%	oDM%
Mean±SD	51.61±1.78	14.09±0.995	39.13±0.49	93.35±4.11

The protein content of examined feather sample (nitrogen content*6.25) on average was 88.1±6.22% which was similar to results of Papadopoulos (1985) experiments (85-99%).

Table 3. Monitoring of growth of bacteria cells with Burkler chamber:

Bacterial cell number (x10 ⁶ cell/ml)	1. day	2. day	3. day	4. day	5. day
Infected experiments	4.55±0.00	11.2±0.58	23.7±1.11	52.32±1.33	70.8±4.62



Results and discussion

I. Pre-treatment of chicken feather

Determination of feather protein concentration

- ▶ Protein concentration of samples could be determined with comparison of absorbance values and calibration curve
- ▶ The dissolved protein concentration in our case was 0.19 mg/ml.

Table 4. Efficiency of feather pre-treatment:

Experiments and differences	Protein concentration changes (mg/ml)				
	1. day	2. day	3. day	4. day	5. day
Infected experiments	0.012±0.001	0.09±0.014	0.16±0.018	0.24±0.031	0.36±0.014
Control	0.00±0.0	0.00±0.0	0.008±0.002	0.013±0.003	0.026±0.012
Differences%	1.2	9.0	16.0	22.7	33.4

Table 5. Keratin degradation rate (%):

Experiments and differences	Keratin degradation rate (%)				
	1. day	2. day	3. day	4. day	5. day
Infected experiments	0.91±0.052	6.79±2.14	12.08±0.96	18.12±1.66	27.17±0.75
Control	0.00±0.0	0.00±0.0	0.60±0.10	0.98±0.16	1.96±0.64
Differences%	0.91	6.79	11.48	17.14	25.21

Pre-treated and control experiments

Determination of sCOD (g/l):

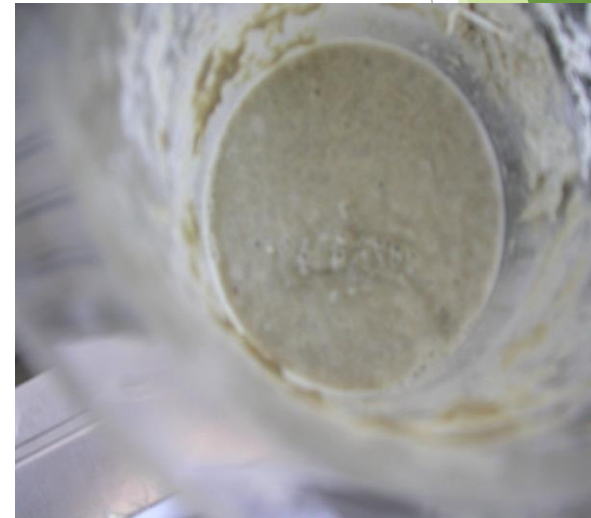
Control

- ▶ sCOD: 6.8 ± 0.56 g/l
- ▶ Solubilisation degree(%): $4.53\% \pm 0.37$



Pre-treated feather

- ▶ sCOD: 106.15 ± 3.18 g/l
- ▶ Solubilisation degree(%): $70.77\% \pm 2.12$



- ▶ Solubilisation degree (%): 66.23% difference was determined compared the control experiments

Results and discussion

II. Biogas production

Batch digestion experiments:

Table 6. Quality parameters of biogas raw materials:

Raw materials	Corn silage	Cattle slurry	Liquid digestate	Pre-treated feather
DM%	26.0±2.73	3.6±0.52	2.8±0.91	19.94±1.36
oDM %	93.0±3.35	82.7±3.89	72.4±4.02	96.4±2.31
C:N ratio	27.6	13,0	18.0	1.47
C-content	45.8±0.87	40.4±2.40	47.3±2.07	15.09
N-content	1.7±0.29	3.1±1.21	2.6±0.25	10.26

Table 7. Calculated average of DM% and oDM%:

Digester number	1	2	3	4
Pre-hydrolysed feather%	0	5	10	20
DM%	3.9±0.5	3.7±0.6	3.6±0.4	3.4±0.5
oDM% in DM%	3.1±0.4	2.8±0.3	2.5±0.2	2.3±0.2



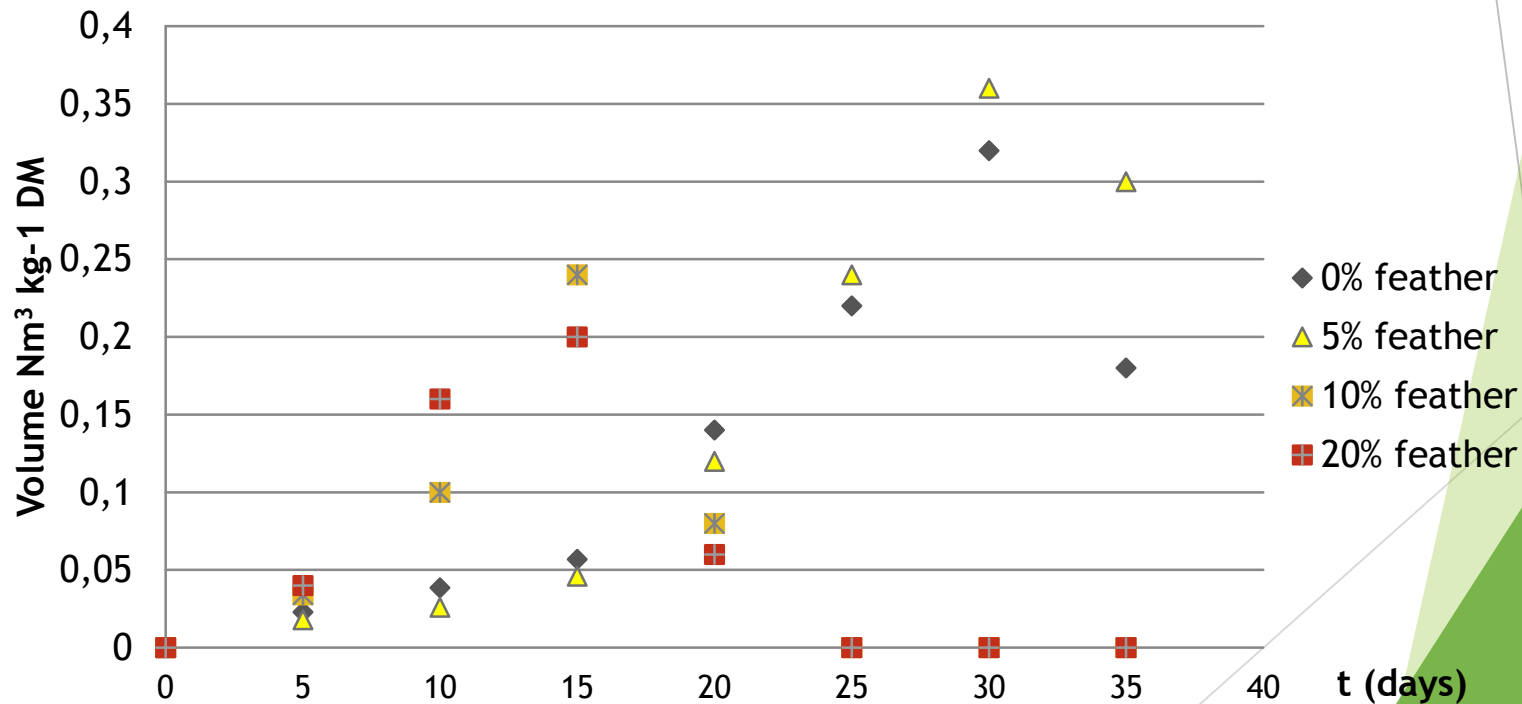
Results and discussion

II. Biogas production

Biogas production of biodegraded chicken feather:

Methane production during co-digestion of different pre-treated feather ratios:

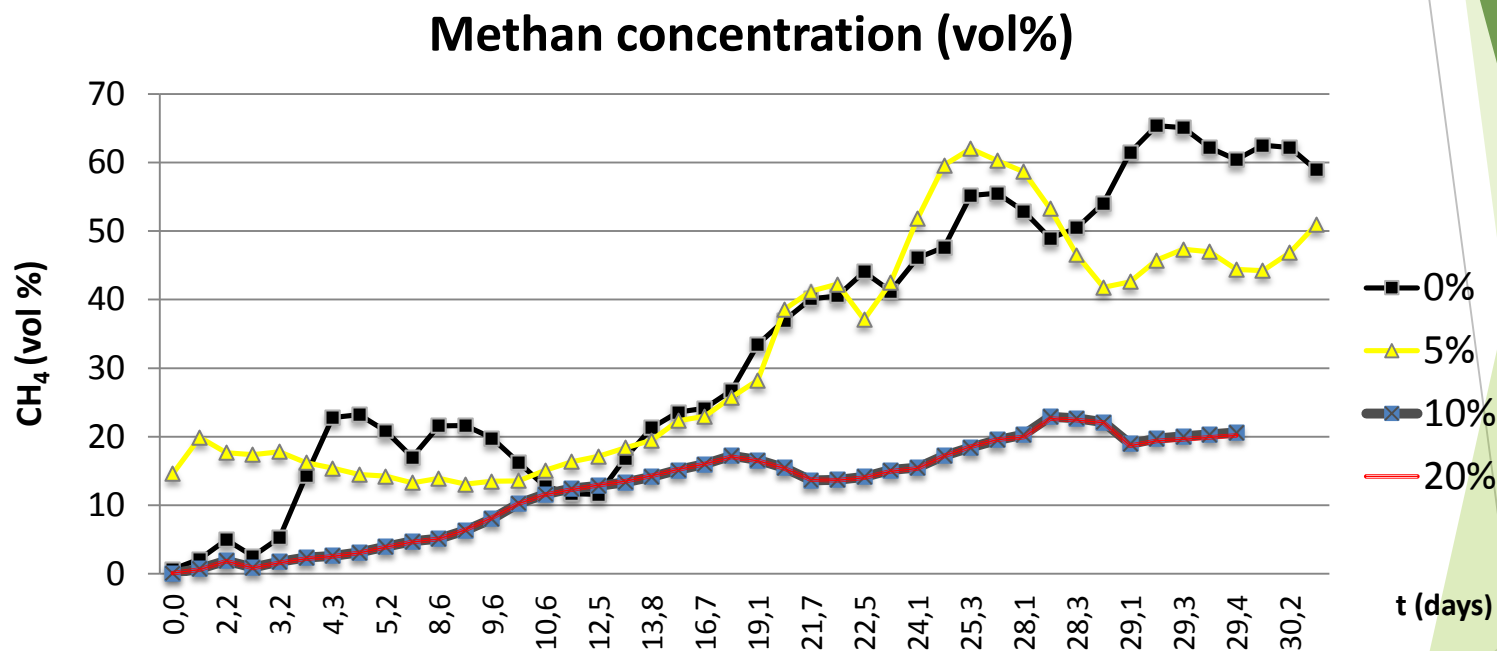
Methane production



Results and discussion

II. Biogas production

Methane concentration (vol%) during co-digestion of different pre-treated feather ratios:



- ▶ The biogas quality in case of the poultry feather mixture rate of 5 showed better results and differed significantly from the rates of 10 and 20%.
- ▶ In case of treatments with a feather mixture rate of 5 methane concentrations around 60% stayed stable.

Conclusion I.

Pre-treatment of feather waste:

- ▶ The protein concentration values and keratin degradation ratios (%) of infected and control experiments showed significant difference applying t-test and variance analysis (Tukey's test) (SD 0.1%).
- ▶ Solubilisation degree of feather (%) - calculated from sCOD - shows 66.23% difference compared the control experiments.

Positive effects on biogas production:

- ▶ In our case 5% pre-treated feather ratio result in a favourable biogas production, the highest methane yield was $0.36 \pm 0.13 \text{ Nm}^3/\text{kg DM}$,
- ▶ second was the control experiment ($0.32 \pm 0.13 \text{ Nm}^3/\text{kg DM}$) after 30 days.
- ▶ Control, 5% experiments and 10, 20% experiments showed significant differences.

Conclusion II.

Negative effects on biogas production:

- ▶ The ratio, which is more than 10%, reduce the biogas yields significantly.
- ▶ Due to the amount of produced H_2S (ppm) the critical mixing ratio of feather proved to be 10% in laboratory environment.
- ▶ The production of the H_2S reached its maximal value (200 ppm) already on the 9th day in case of the 10% treatment.
- ▶ Three treatments group could be selected in case of the highest ammonia concentrations (control and 5% and 10, 20%), which were detected after 20 days.
- ▶ Above 10% inhibitory effect was observed in case of produced ammonia.

Acknowledgements

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Thank you for your kind attention!