

Short Communication

## HIGH-RISK BIODEGRADABLE WASTE PROCESSING BY ALKALINE HYDROLYSIS

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Biodegradable waste is by definition degraded by other living organisms. Every day, meat industry produces large amounts of a specific type of biodegradable waste called slaughterhouse waste. Traditionally in Europe, this waste is recycled in rendering plants which produce meat and bone meal and fat. However, feeding animals with meat and bone meal has been banned since the outbreaks of bovine spongiform encephalopathy (BSE). In consequence, new slaughterhouse waste processing technologies have been developed, and animal wastes have now been used for energy production. Certain parts of this waste, such as brains and spinal cord, are deemed high-risk substances, because they may be infected with prions. Their treatment is therefore possible only in strictly controlled conditions. One of the methods which seems to bear acceptable health risk is alkaline hydrolysis. This paper presents the results of an alkaline hydrolysis efficiency study. It also proposes reuse of the obtained material as organic fertiliser, as is suggested by the analytical comparison between meat and bone meal and hydrolysate.

**KEY WORDS:** *cow brain, BSE, organic fertilisers, slaughterhouse waste, rendering plants, spinal cord*

Wastes of all sorts are the essential issue of modern civilisation and lifestyle. By piling up waste, the humans severely disrupt the natural equilibrium.

One of the priorities in reducing environmental pollution is to resolve the problem of wastes. Taking a standpoint on waste issues, different states have developed different strategies and defined goals and measures aimed at preserving natural resources and protecting the environment (1). The fundamental principles are based on reducing the quantities of waste by continuously increasing their reuse and recycling through new technologies (2, 3). Animal by-products or slaughterhouse wastes have become a part of our everyday life (3, 4); their quantities have been

constantly increasing over the last twenty years, due to increasing consumption (5). In rendering plants, animal by-products are processed into meat and bone meal and grease for technical purposes (3, 6). Meat and bone meal is a source of animal protein and had been used in the production of feeding stuffs. However, since the outbreaks of bovine spongiform encephalopathy (BSE), this kind of use has been banned (7-9). Animal proteins have a very high biological value; their composition is the most similar to that of yeast, soy, and sunflower proteins. However, proteins of vegetal origin used in animal nutrition may achieve high biological value only when properly combined and mixed. Furthermore, meat and bone

meal has a market value in soil fertilisation, and the increasing quantities of proteins of animal origin open wide possibilities for their use, especially for generating energy, as one of the crucial issues today in Europe (5, 10). Current treatment technologies are still insufficient for efficient and safe processing. It is necessary to review various possibilities for using waste materials for energy generation without presenting any environmental hazard. This places the spotlight on integrated waste management systems, whose primary goal is to develop new technologies for efficient reuse high-risk materials, apart from heat treatment, currently used in rendering facilities (11). One such new, promising and environmentally acceptable technology is alkaline hydrolysis of high-risk biodegradable waste.

It could be a good solution for Croatia, which in 2010 still processed 100,000 tonnes of animal waste in rendering plants. Its advantage is that it combine sterilisation and digestion into one operation. This



**Figure 1** Laboratory-scale alkaline digester

process reduces the total volume of the mixture, completely destroys pathogens, and reduces the odour. There is no meat and bone meal fraction like in the rendering plants but only hydrolysed material, suitable for application in soil.

**Table 1** Comparison between meat and bone meal and high-risk biodegradable material after alkaline hydrolysis (mean  $\pm$  SD)

Chemical analyses	Meat-bone meal	High-risk biodegradable material (hydrolysate)
pH	8.5 $\pm$ 0.07	7.98 $\pm$ 0.03
EC / mS cm <sup>-1</sup>	24.22 $\pm$ 0.17	37.10 $\pm$ 0.07
DM (105 °C) / %	96.12 $\pm$ 0.12	17.31 $\pm$ 0.17
H <sub>2</sub> O / %	3.88 $\pm$ 0.13	82.70 $\pm$ 0.17
annealing residue (550 °C) / %	14.27 $\pm$ 0.06	21.43 $\pm$ 0.13
annealing loss / %	70.00 $\pm$ 0.03	78.60 $\pm$ 0.03
organic carbon / %	40.00 $\pm$ 0.03	40.02 $\pm$ 0.03
	in organic sample	4.30 $\pm$ 0.08
N / %	overall in DM	8.91 $\pm$ 0.02
	in other forms (105 °C)	9.29 $\pm$ 0.18
	NH <sub>3</sub> -N	9.10 $\pm$ 0.17
P <sub>2</sub> O <sub>5</sub> / %	5.64 $\pm$ 0.13	2.81 $\pm$ 0.18
K <sub>2</sub> O / %	0.83 $\pm$ 0.02	1.96 $\pm$ 0.12
Ca / %	2.35 $\pm$ 0.02	6.24 $\pm$ 0.02
Mg / %	0.11 $\pm$ 0.03	1.53 $\pm$ 0.02
Na / %	1.89 $\pm$ 0.17	1.90 $\pm$ 0.26
Mn / mg kg <sup>-1</sup>	92.00 $\pm$ 0.04	94.52 $\pm$ 0.04
Zn / mg kg <sup>-1</sup>	90.91 $\pm$ 0.05	68.13 $\pm$ 0.16
Cu / mg kg <sup>-1</sup>	12.55 $\pm$ 0.21	12.22 $\pm$ 0.24
Fe / mg kg <sup>-1</sup>	300.00 $\pm$ 0.05	441.3 $\pm$ 0.06
Pb / mg kg <sup>-1</sup>	0.79 $\pm$ 0.07	0.72 $\pm$ 0.06
Cd / mg kg <sup>-1</sup>	0.09 $\pm$ 0.07	0.11 $\pm$ 0.04

EC = electro-conductivity

DM = dry matter

NH<sub>3</sub>-N = ammonia nitrogen

The aim of this paper was to determine the composition of hydrolysed material used in agriculture as organic fertiliser and to present alkaline hydrolysis as a possible alternative in processing high-risk biodegradable wastes. The resulted product can be used in biogas production or directly as fertilizer. We also compared the composition of alkaline hydrolysates with the composition of meat and bone meal produced by a traditional rendering plant. This research is a contribution to the EU Directive 1774/2002 amendment (12), which defines methods for treating high-risk slaughterhouse wastes and refers to alkaline hydrolysis as economically and environmentally most acceptable model of certain high-risk biodegradable waste management.

## MATERIALS AND METHODS

Alkaline hydrolysis is a simple, natural process by which some complex molecules are broken into their constituent building blocks. The process occurs in nature when animal tissue and carcasses are buried in soil of neutral or alkaline pH. Hydrolysis can be catalyzed by enzymes, metal salts, acids or bases. Bases are typically water solutions of alkali metal hydroxides such as sodium hydroxide (NaOH) or potassium hydroxide (KOH). Alkaline hydrolysis is carried out in a tissue digester, an insulated, steam jacketed, stainless-steel pressure vessel with a lid that is manually or automatically clamped (Figure 1). The vessel contains a retainer basket for bone remnants and other materials like indigestible cellulose, latex, and metals. In our study, the vessel was operated in batch mode, at pressure of up to 4 bars, temperature 150 °C, reaction time 3 h, and use of KOH as catalyst. High-risk biodegradable material was taken from a Croatian rendering plant. Hand blender was used for mixing and chopping up the needed quantity of waste (400 g of cow brain) until the mixture became homogenous. The mixture was then placed in a reactor, and 30 mL of 45 % KOH solution and 600 mL of tap water were added. The same reaction mixture was prepared for three experimental cycles, under the same testing conditions. At the same time, we prepared meat and bone meal for chemical analyses.

Chemical analyses of the hydrolyzed material included the following: pH measurement with combined electrode (DIN EN 1256:2003) directly from hydrolysed material, electrical conductivity measurement with a conductivity meter MA5964 with

combined electrode (ISO 7888:1985), total nitrogen content using the Kjeldahl method (ISO 1871:1975), nitrate content ( $\text{NO}_3^-$ ) using spectrophotometry based on phenoldisulphonic acid, nitrogen ammonium content ( $\text{NH}_4^+$ ) using spectrophotometry with Nessler's reagent based on the Jackson's method, phosphorus content as phosphate ( $\text{PO}_4^{3-}$ ) using molybdate-blue spectrophotometry, potassium and sodium content using flame photometry, and microelement (Ca, Mg, Mn, Zn, Cu, Fe, Pb, Cd) content using atomic absorption spectrophotometry (AAS).

Statistical analysis was made in accordance with randomly selected experimental design, i.e., the experiments were made in triplicate and arithmetic mean values and standard deviation ( $\pm$ SD) were calculated.

## RESULTS AND DISCUSSION

The results of the multivariate chemical analysis for both meat and bone meal and hydrolysed high-risk biodegradable material (cow brain) are presented in Table 1. Alkaline hydrolysis ultimately degrades proteins by breaking chemical bonds (2). During hydrolysis, large protein molecules break into smaller (peptides), which is important for prion decontamination (13-15). All polypeptides contain carbon, hydrogen, nitrogen, and oxygen, with smaller proportions of other elements such sulphur and phosphorus. During hydrolysis, proteins degrade to small peptide chains and amino acids in the form of salts (2). Alkaline hydrolysis destroys all pathogens, including prions, converts fixatives, cytotoxic agents, and other toxins, and decomposes the harmful biodegradable derivatives (2, 15).

Alkaline hydrolysate is a sterile, coffee-coloured alkali solution with soup-like odour and high dry matter content (17.31 %), which can be used like liquid fertilizer (16), particularly because its organic matter content is high (40.02 %). The product may smell badly, which will have to be dealt with when it becomes applied in agriculture. Measurements of pH show that the hydrolysate is slightly alkaline (pH=7.98) and we are confident, that the hydrolysed material is good for use in acidic soils as pH-correction agent.

The tested samples were rich in nitrogen (9.42 %), phosphorus (2.81 %), and potassium (1.96 %). The N:P:K ratio was 3.35:1:0.70. This amount is several times higher than in common organic fertilisers, which renders hydrolysed material highly applicable as organic fertiliser (17).

The heavy metal content was below the Croatian maximum allowed levels stipulated by the Regulation on protection of agricultural land against pollution agents (18). According to this Regulation, a hydrolysate may be used on agricultural soil only if it has been analysed. The maximum applied quantity is 60 m<sup>3</sup> ha<sup>-1</sup> in the vegetation period and 30 m<sup>3</sup> ha<sup>-1</sup> over the rest of the year (October to April). This fertiliser must not be used in areas with high-risk pollution, on water-saturated soils such as those covered with snow and frozen soils. Hydrolysed material must not be used in the coastal and water flow areas at distances less than 10 m from a body of water, then in stationary water areas at distances of less than 70 m from a body of water, nor can they be used to fertilise vegetables, strawberries, and medical herbs within 30 days to harvest.

Based on our results and the Croatian regulations, the hydrolysed material may be classified as organic fertilizer with lower nutrient concentration, which can be used if analysis warrants its use and in soils where its application is allowed.

In waste management, alkaline hydrolysis is a very important step. The final product can be used directly for soil fertilisation and process is cheaper than meat and bone meal recovery. The alternative is to incinerate meat and bone meal, which significantly increases the cost in food industry.

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### **Sažetak**

#### **OPORABA VISOKORIZIČNOG BIORAZGRADIVOG OTPADA METODOM ALKALNE HIDROLIZE**

Biorazgradivi otpad definira se kao otpad koji razgrađuju živi organizmi. Klaonice i mesna industrija proizvode na dnevnoj bazi velike količine specifičnoga biorazgradivog otpada poznatog kao otpad životinjskog podrijetla. Tradicionalno se u Europi taj otpad reciklira u kafilerijama, pri čemu se proizvode mesno-koštano brašno i mast. No nakon pojave goveđe spongiformne encefalopatije (GSE) zabranjena je prehrana životinja mesno-koštanim brašnom. U potrazi za novim mogućnostima zbrinjavanja otpada životinjskog podrijetla razvijene su nove tehnologije uporabe i omogućena upotreba takvog otpada u energetske svrhe. Određeni dijelovi otpada životinjskog podrijetla, mozak i leđna moždina, pripadaju kategoriji visokorizičnog otpada zbog velike mogućnosti postojanja priona u njima. Njihova uporaba stoga je moguća samo u strogo kontroliranim uvjetima. Jedna od metoda koja se smatra prikladnom za uporabu visokorizičnih otpada jest metoda alkalne hidrolize. U radu su prikazani rezultati analiza mesno-koštanog brašna i hidroliziranog otpada te njegova uporaba kao organskoga gnojiva.

**KLJUČNE RIJEČI:** *kafilerije, klaonički otpad, kravlji mozak, leđna moždina, organsko gnojivo*

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