### Pilot-scale investigation of bioethanol production from source-separated food waste

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# Introduction

Food waste management constitutes a serious environmental issue, since one-third of food produced for human consumption is lost or wasted globally, amounting to about 1.3 billion tonnes per year. In Greece, the amount of food wasted is estimated to more than twice the global average and more than any other European country (UNEP, 2021). Therefore, the development of environment-friendly solutions for the valorization of these wastes, especially in countries like Greece, constitute the highest priority. There are several reports in the literature that examine the potential of food waste valorization through the bioethanol pathway. However, in most of them, the research is limited to lab scale. In the present study, the optimization of the process was made on pilot scale trials. The aim of this paper was to optimize the production of bioethanol from source-separated food waste at pilot scale.

### **Materials and Methods**

The source-separated food waste utilized in this study was collected from the Vari-Voula-Vouliagmeni Municipality, Attica, Greece. The biowaste was collected and transferred to the Unit of Environmental Science and Technology (UEST), School of Chemical Engineering, NTUA, where it was dried and milled by a GAIA biowaste dryer (model GC-100). The raw material had approximately 75% moisture. The average composition of the dried feedstock was in % w/w dry basis: cellulose  $17.47 \pm 3.19$ , hemicellulose  $5.81 \pm 2.04$ , starch  $4.96 \pm 2.21$ , fats and oils  $11.97 \pm 1.51$ , acid soluble lignin  $1.39 \pm 0.62$ , acid insoluble residue  $10.42 \pm 2.75$ , water soluble solids  $33.85 \pm 1.84$ , volatile solids  $88.38 \pm 1.84$ , moisture  $5.40 \pm 2.38$ .

All chemicals used were of analytical grade. Non-commercial enzymatic cocktails such as NS22109 or Spirizyme that belong to amylases family and, NS22177 or NS87014 known as cellulolytic enzymes were kindly donated by Novozymes (Denmark) and used in this study. Amylase activity of NS22109 and Spirizyme was measured according to the method described by Xiao et al. (Xiao et al., 2006) and found 2425 and 2337 U/mL respectively. Total cellulase activity (FPU) of NS22177 and NS87014 was measured against filter paper by the standard IUPAC method, as described by Ghose (Ghose, 1987), and found to be 227 and 333 FPU/ mL respectively. The fermentation yeast used was Saccharomyces cerevisiae, added in the form of dry baker's yeast.

The characterization of structural carbohydrates and lignin in biomass of raw and treated material was determined following the NREL laboratory analytical procedures (Sluiter et al., 2012). The liquid fractions were analyzed by HPLC for the determination of sugars, ethanol and volatile fatty acids. All analyses were performed in duplicate.

# **Pilot plant**

A bioconversion pilot scale system installed at UEST was utilised for the treatment train of dried food waste. The pilot plant includes two stainless steel agitated horizontal rotating reactors having a capacity of 200 L each, which can operate independently. In the first reactor, the enzymatic saccharification took place while in the second the fermentation. The temperature in the reactors is set through the circulation of water in the double walls of the vessels. At the end of fermentation, the bioethanol produced was recovered a distillation unit at 70 °C with the contribution of a vacuum pump. The distillation unit is a coil type heat exchanger which cools the ethanol vapours by heating a water reflux system. The operation of the pilot plant is fully controlled through a PLC.

#### Enzymatic hydrolysis and fermentation

Within this study, two fermentation modes were examined; separate hydrolysis fermentation (SHF) and simultaneous saccharification fermentation (SSF). The effect of solids loading (10, 15, 20 % w/w) was evaluated, while the enzyme loading was stable at 40  $\mu$ L/g starch for amylases (NS22109 or Spirizyme) and 175  $\mu$ L/g cellulase for cellulases (NS22177 or NS87014). In all trial, the dosage of yeast was 2% of the initial solid load. In the SHF case, the hydrolysis of starch was conducted at 65 °C for 2h, the hydrolysis of cellulose at 50 °C for 6 h and the fermentation step at 35 °C for 7 h. In the SSF case, the process was conducted at 35 °C for 7 h based on preliminary laboratory trials. Samples were collected in hourly time intervals and analyzed for the determination of glucose and ethanol concentration in order to monitor the progress of bioprocess. The performance of enzymatic hydrolysis was evaluated based on the glucose yield  $Y_{\rm G}$  (g/g of theoretical glucose) and, the performance of fermentation was estimated based on ethanol yield  $Y_{\rm Etoh}$  (g/g of theoretical ethanol).

### **Results and discussion**

Table 1 presents the experimental trials with the respective saccharification and ethanol yields

Trials	Loading (%w/w)	SHF	SSF	Mix 1	Mix 2	Y <sub>G</sub> (g/g)	Ethanol concentration (g/L)	Y <sub>EtOH</sub> (g/g)
1	10	$\checkmark$		$\checkmark$		0.74	10.0	0.48
2	15	$\checkmark$		$\checkmark$		0.69	10.5	0.43
3	20	$\checkmark$		$\checkmark$		0.58	11.1	0.38
4	10		$\checkmark$	$\checkmark$			6.2	0.47
5	15		$\checkmark$	$\checkmark$			5.9	0.27
6	15		$\checkmark$		$\checkmark$		13.2	0.57
7	20		$\checkmark$		$\checkmark$		22.0	0.58
Mix 1: NS22109, NS22177 Mix 2: NS87014, Spirizyme								

Table 1. Pilot trials and results of bioethanol production from source-separated food waste.

From Table 1 it is evident that the increase of solids loading leads to a reduction of ethanol yield. Nevertheless, this corresponds to higher ethanol concentrations which are advantageous in technoeconomic terms. Moreover, the use of enzymatic mix 1 (NS22109 and NS22177) resulted in ethanol yields that ranged from 0.27 to 0.48, while the ethanol yields with mix 2 (NS87014 and Spirizyme) were higher (0.57-0.58). In addition, the enzymatic formulations proved efficient even in the lower temperature of 35 °C, well below their optimum range. Thus, SSF could be considered as a promising solution.

After 7h of SSF process with 15% and 20% solids loading, 13.2 g/L ( $Y_{EtOH}=0.57$ ) and 22.0 g/L (or  $Y_{EtOH}=0.58$ ) ethanol respectively was obtained by the application of enzymatic mix 2. The degradation of cellulose was approximately 62% and the degradation of starch ranged from 75 to 80%. Furthermore, more than 84% of produced ethanol was recovered via the distillation unit.

# Conclusions

The production of bioethanol by applying two fermentation modes; SHF and SSF, was examined at pilot scale. It was revealed that the enzymatic formulations can operate effectively at lower temperature (35°C) which is also suitable for the yeast. Hence, SSF could stand as a viable fermentation mode for the production of bioethanol from source-separated food waste.

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