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Methane and hydrogen production from potato wastes and wheat straw under dark fermentation

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Batch dark fermentation of wheat straw and boiled potato wastes at volatile suspended solids (VSS) 5 g VSS/L are examined and compared. Investigations on dark fermentation of potato wastes and wheat straw were carried out at different pH and OFR (oxygen flow rate) values and inoculum pretreatment. The obtained hydrogen yield from waste potato was 70 mL/g VSS, while for hydrolysed wheat straw it amounted to 80 mL/g VSS. The optimum conditions for potato dark fermentation are acidic pH 6.0 and OFR 1.0 mL/h, while for the wheat straw, optimal conditions are pH 6.4 and OFR 4.6 mL/h. The comparison revealed a significant difference in hydrogen production due to the type of substrate, inoculum stressing and DF conditions applied.

Keywords: dark fermentation; hydrogen production; wheat straw; boiled potato wastes; micro-aeration; inoculum stressing

1. INTRODUCTION

Demand for methane and hydrogen grows rapidly with depletion of fossil fuels, increased role of renewable energy sources and their imminent variation. So, methane and hydrogen (or related e-fuels) can help to secure provision when energy is needed. To cover the demand, biomethane and biohydrogen produced from biowaste are interesting options. Potatoes and wheat are basic for global food system, thus they constitute a major source of biowaste. In Poland, potato and wheat straw wastes amount to 7.4×10^6 Mg and 5.8×10^6 Mg, contributing 14.3% and 16.5% of the European Union wastes, respectively (De Cicco and Jeanty, 2017; Sołowski et al., 2019a). Production of both crops generates enormous masses of wastes, so, the relevant issue is its proper utilization.

Dark fermentation (DF) is one of the potential methods for conversion of simple organic compounds (usually sugars or fats) into hydrogen, carbon dioxide, and organic acids. It is a special case of anaerobic digestion (AD), terminated at acetogenesis, i.e. the bacterial conversion of organic wastes (hydrolysed to acids, sugars, fats and proteins) into volatile acids, hydrogen, CO_2, etc. (García Depraect et al., 2020; Hawkes et al., 2007). During DF methane is not formed or its concentration is minimal (Gallipoli et al., 2020). The optimal for DF conditions are, in general, acidic – pH around 5.0 (Aly et al., 2018), but in the case of cotton stalk best pH equals 8.5 (Li et al., 2018), or neutral in the case of potato wastes – pH 7.8 (Sekoai et al., 2019). Hydrogen production is usually conditioned by pretreatment of bacterial

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community (inoculum stressing), such as heat shock, sudden pH change, etc. (Chaganti et al., 2012). However Dessě et al. (2017) claimed that inoculum stressing is not necessary, see e.g. gangrene case (Chi et al., 1995).

The substrate selection and minimal pretreatment costs are relevant factors for an economically profitable DF process. The common agricultural waste such as wheat straw or potato wastes are investigated under different process conditions – see e.g. (Kumar et al., 2015) fermentation of fries. Potato wastes contain 16 – 22% of dry matter including: starch 10 – 16%, total sugars 0.3 – 0.6%, total protein 1.7 – 2.3%, lipids 0.10 – 0.12%, dietary fibres 2.0 – 2.3% and ash 1.0 – 1.2% (Leszczyński, 2000).

Wheat straw contains mainly lignocellulose – a combination of cellulose, hemicellulose and lignin (Patel et al., 2015). A comparison of dark fermentation of both wastes under similar conditions shows the influence of starch and a different fraction of residual lignin in the feed before fermentation. Moreover, a comparison of economic efficiency of hydrogen production from these two substrates helps in DF substrate selection.

In the case of lignocellulosic substrates the initial stage of dark fermentation is related to its hydrolyses to simple sugars like pentoses and hexoses (Sołowski et al., 2020b). Bartacek et al. (2007) and Woodward et al. (2000) pointed out that there are three thermodynamically possible DF pathways from hexoses: acetate (1), butyrate (2) and acetate-ethanol pathway (3):

$$C_6H_{12}O_6 + 4H_2O \rightarrow 2CH_3COO^- + 2HCO_3^- + 2CO_2 + 4H_2 + 2H^+, \qquad \Delta G^0 = -48 \text{ kJ mol}^{-1} \tag{1}$$

$$C_6H_{12}O_6 + 2H_2O \rightarrow CH_3CH_2CH_2COO + 2HCO_3^- + 2CO_2 + 2H_2 + 3H^+, \qquad \Delta G^0 = -137 \text{ kJ mol}^{-1} \tag{2}$$

$$C_6H_{12}O_6 + 3H_2O \rightarrow CH_3COO^- + 2HCO_3^- + 2CH_3CH_2OH + 3H_2, \qquad \Delta G^0 = -97 \text{ kJ mol}^{-1} \tag{3}$$

The acetate pathway is the one with the highest theoretical hydrogen yield: 4 moles of H_2 from 1 mole of hexose. The most efficient according to Hawkes et al. (2007) is the acetate pathway (1) but the most probable is the butyrate one (2). Alkaline pretreatment supports acetate fermentation pathway.

Besides, there were some over optimistic chemical pathways proposed with the complete decomposition of 1 mole of glucose to 12 moles of hydrogen: [$C_6H_{12}O_6$+ $6H_2O \rightarrow 12H_2$+ $6CO_2$] – see e.g. (Słupek et al., 2019). However, the process which could theoretically yield 12 moles of hydrogen from 1 mole of glucose:

$$C_6H_{12}O_6 + 12H_2O \rightarrow 6HCO_3^- + 6H_2 + 6H^+, \quad \Delta G^0 = 241 \text{ kJ mol}^{-1} \tag{4}$$

is thermodynamically unobtainable due to required positive value of Gibbs free energy.

Another important issue is a choice of the proper nutrients to enhance DF efficiency. Han et al. (2015) examined the ability and stimulation range of some nutrients e.g. whether the selected nutrient enhances DF for every substrate, or only particular types.

The DF studies of sour cabbage (Sołowski et al., 2019b), cotton wastes (Sołowski et al., 2020b) and glycerol (Paillet et al., 2019) proved that micro-aeration can increase the process efficiency.

The aim of this paper is:

- to verify the viability of wheat straw and potato wastes as a substrate for DF,
- to verify the effect of inoculum stressing,
- to determine the dependence of fermentation process on micro-aeration.

2. MATERIALS AND METHODS

2.1. Inoculum

The inoculum used for the experiments came from a mesophilic digester at Darżyno (Pomerania), treating mainly maize silage and manure. Before experiments, the inoculum was stored for about two weeks to minimize its biogas production and sieved to remove large particles. The inoculum without any pretreatment (stressing) was denoted as "raw inoculum". The stressed inoculum was prepared by boiling it for 30 minutes at 105 °C, following the heat shock procedure described by (Nasirian et al., 2011) and (Hernández et al., 2019).

2.2. Substrates

Potato wastes were taken from the local restaurant "3 Smaki" in Gdańsk in July 2019. The potato wastes and the wheat straw were milled mechanically using a mincer Royal Catering Model RCFW 120 PRO with a sieve size of 10 mm. The characteristics of the final products are presented in Table 1.

2.3. Physical analyses

The total solids (TS) [%FM] and volatile solids (VSS) [%TS] in the inoculum and the substrate were determined using the Standard Methods for characterization of biomass properties (Moriarty, 2013), see Table 1.

Table 1. Characteristics of inoculums, wheat straw and boiled potatoes wastes

Material	pH	TS [%FM]	VSS [%TS]
Raw inoculum	8.24	1.1%	45.4 ± 1.0
Wheat Straw	6.5	92.4%	98.3% ± 1.2%
Boiled Potatoes Wastes	5.83	24.4%	93.0% ± 1.0%

2.4. Acid pretreatment of wheat straw

Some of the wheat straw milled (WSM) to the size of 2 mm was hydrolysed to obtain wheat straw hydrolysed (WSH); hydrolysis was performed at pH 2.4 during 3 hours using 0.1 M HCl solution as in (Nasirian et al., 2011) then diluted with water (4 litres of water per 100 mL of hydrolyzate) to pH 5.5. This replaces more expensive sulphuric acid (3 €/L of acid) with hydrochloric acid procedure – 0.23 €/ L of HCl.

2.5. Batch fermentation

Wheat straw and potato waste fermentations were placed in 2 dm^3 glass reactors with a working volume of 1.2 dm^3. The WSM and WSH dark fermentation was investigated. Potato wastes were just milled. Stressed (boiled) and raw inoculums were used to ferment wheat straw and potato wastes under VSS load of 5 g/L. The fermentation process was performed under mesophilic conditions with temperatures ranging from 36 °C to 40 °C.

During wheat straw and potato waste dark fermentation, an effect of micro-aeration was studied for oxygen flow rate (OFR) from 0 to 4.6 mL/h and OFR 0–1 mL/h, respectively. The addition of oxygen was accomplished by syringe twice per day. The pH was measured and stabilized once per day, using 0.1M HCl and 0.1M NaOH solutions.

2.6. Biogas measurement

The gas assessment was performed and determined in two stages. During the first one, the gas composition was determined by a portable biogas analyser (GA5000, Geotech), when the volume of collected biogas in the measuring cylinder (see Fig. 1) was at least 0.45 dm^3. The analyser poses ATEX II 2G Ex ib IIA T1 Gb (Ta: −10 °C ... +50 °C), IECEx, and CSA quality certifications, as well as UKAS ISO 17025 calibration certificate. The equipment allowed the measurement of CH_4, CO_2, O_2, H_2, and H_2S in the ranges 0–100%, 0–100%, 0–25%, 0–1000 ppm, and 0–5000 ppm, respectively. Calibration of the device was performed twice a week. During the second stage, when hydrogen concentration was above 1000 ppm, the gas was measured by a gas chromatograph (GC) with a thermal conductivity detector (TCD), argon was used as a carrier gas (gas flow rate was 0.6 mL/h). A Silica packed column (Restek®) with characteristics of 2 m / 2 mm ID 1/8" OD Silica was used. All the experiments were triplicated, and the mean values for methane and hydrogen measurements are reported.

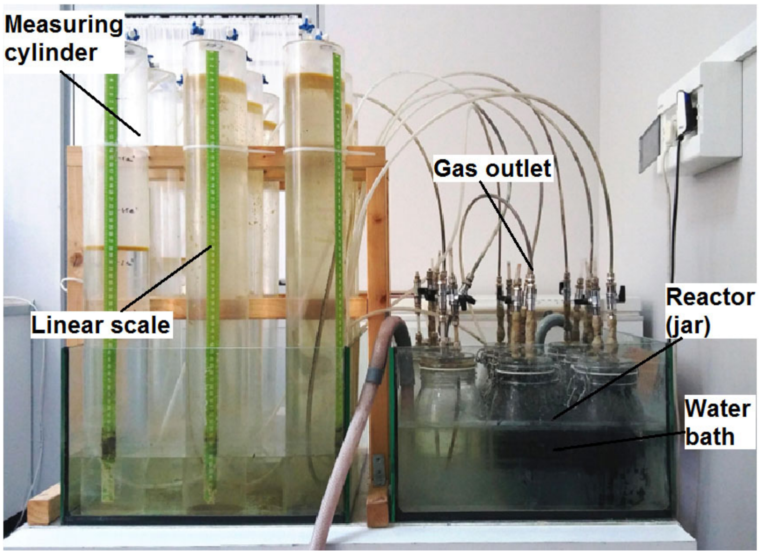

Fig. 1. Photo of the experimental setup

3. RESULTS AND DISCUSSION

Both, hydrogen and methane were present in biogas produced during studied DF under load of 5 g VSS/L. However, for clarity reasons, the results for hydrogen and methane production are discussed separately in 3.1 and 3.2 subchapters, respectively.

3.1. Hydrogen production

The hydrogen production is discussed taking into account various substrates, role of inoculum stressing and micro-aeration. Figure 2 presents cumulative hydrogen production from different substrates versus fermentation time. It demonstrates also the difference between application of raw and boiled (stressed) inoculum.

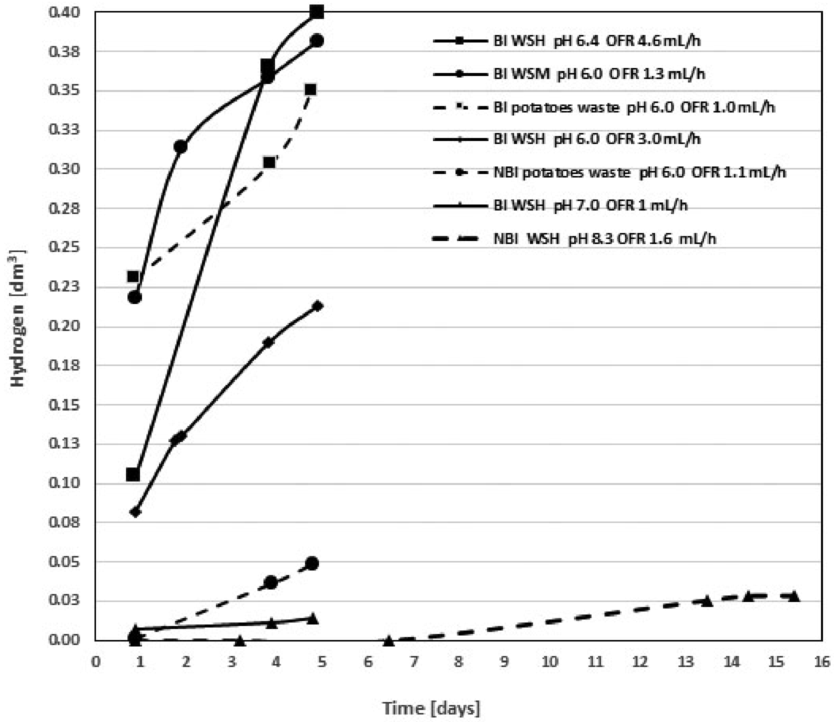

Fig. 2. Cumulative DF hydrogen production from hydrolysed wheat straw and potato wastes under load 5 g VSS/L using boiled (BI) and not boiled (raw) inoculum (NBI)

3.1.1. Substrates verification

It is seen (Fig. 2) that during first 5 days accumulative DF hydrogen production from wheat straw (WSM and WSH) and boiled potatoes are similar (ranging from 0.35–0.40 dm^3) when boiled (stressed) inoculum is used. However, it should be pointed out that in the case of WSH significantly larger OFR was applied.

The fermentation of potato waste requires only limited pretreatment e.g. milling, low pH of the process, and boiled inoculum. The hydrochloric pretreatment (hydrolysis) of wheat straw results in higher hydrogen production (and costs). Such pretreated wheat straw could be as efficient hydrogen source as key-lime or cabbage (Sołowski et al., 2019b).

3.1.2. Role of inoculum stressing

The inoculum stressing leads to increased hydrogen production and to decreased duration of dark fermentation process. The highest hydrogen production in WSH fermentation applying raw inoculum was found at an OFR of 1.6 mL/h and pH 8.3, i.e. 28 mL, while applying boiled inoculum leads to hydrogen production of 400 mL at OFR 4.6 mL/h and pH 6.4. So, the hydrogen production was almost 700 times higher and for 2.5 shorter times of fermentation using boiled inoculum. In the case of the boiled potato waste 367 mL of hydrogen was produced during DF (at pH 6.0 and OFR 1.0 mL/h)with boiled inoculum, while for raw inoculum only 48 mL of hydrogen was obtained (at pH of 6.0 and OFR 1.1 mL/h).

The decrease of DF fermentation time in boiled inoculum in potato waste and wheat straw was already reported in (Sołowski at al., 2020a). In the case of wheat straw dark fermentation with raw inoculum hydrogen is produced 11 days longer but the process continuation did not increase significantly final cumulative volumes. In the case of cotton waste 16 days process prolongation was observed Sołowski et al., 2020b).

It was found that application of stressed (boiled) inoculum leads also to an increase of hydrogen concentration in biogas. In the case of potato wastes, application of raw inoculum results in 4% hydrogen concentration, while use of boiled inoculum led to increased H_2 concentration, equal to 48%.

3.1.3. Role of micro-aeration

Micro-aeration enables significant increase of hydrogen production as was pointed out by (Sołowski et al. 2020a). It was found that significant hydrogen production is possible only for a limited range of oxygen flow rates and an optimal OFR value was found both for DF of sour cabbage and cotton wastes.

In the case of WSH fermentation with the untreated, not boiled inoculum hydrogen production was observed for OFR up to 4 mL/h. For the boiled inoculum, hydrogen appeared during DF with OFR even above 4.6 mL/h. In the case of WSH DF with OFR of 4.6 mL/h (pH 6.4), the largest hydrogen production was found, i.e. 400 mL after 5 days of fermentation, while for OFR 3.0 mL/h (pH 6.0), it falls to 213 mL.

3.2. Methane production

During experiments discussed here, methane production far exceeded hydrogen production (even 5 times), e.g. in the case of potato waste fermentation using boiled inoculum under OFR 1 mL/h and pH 6.0 cumulative methane production 1,85 dm^3 was registered on 5^{th} day, while hydrogen production was only of the order 0.35 dm^3. The registered methane yields from potato wastes were 370 mL CH_4/ gVSS. Achinas et al., (2019) reported a lower methane yield from potato peels of 217.8 mL CH_4/g VSS. The analysis of methane production as a competitive process to hydrogen production is indispensable to draw sound conclusions of DF results (Si et al., 2016).

Wheat straw was a less effective source of methane production than potato waste and requires more complex pretreatment, so it is better to use potato waste as fermentation substrate than wheat straw (see Fig. 3).

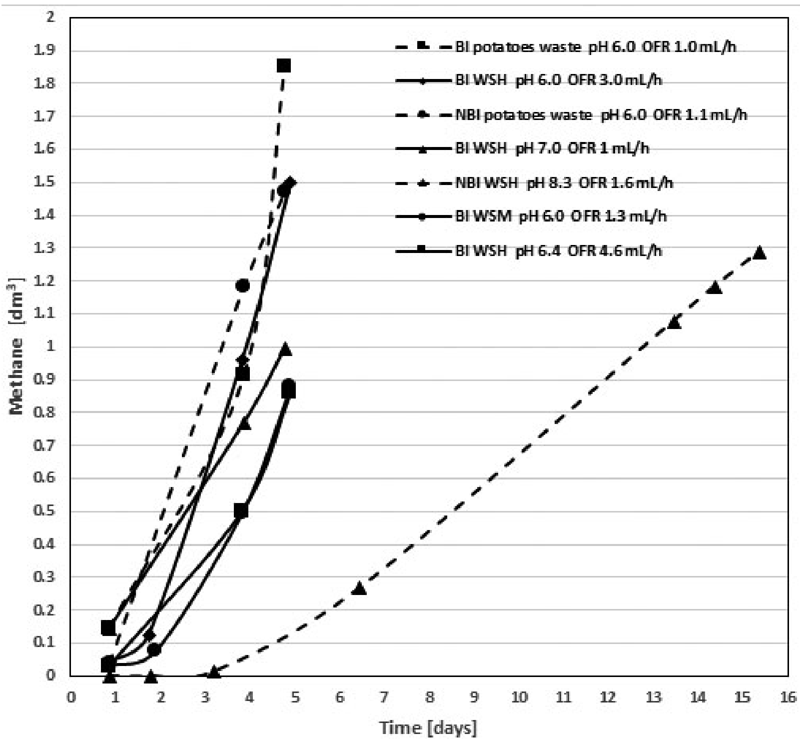

Fig. 3. Cumulative methane production from potatoes and wheat straw for 5 g VSS/L in boiled (BI) and raw (NBI) inoculum and results for potato fermentation under OFR 1.1 mL/h, pH 6.0 (Sołowski et al., 2020a)

Also fermentation of acid-pretreated WSH gives better results (more biomethane) than using only milled WSM.

However, strangely enough use of raw inoculum leads to lower methane production, both in the case of potato waste and WSH. This may point to the fact that the inoculum thermal stressing(boiling) applied here (after Nasirian et al., 2011) does not inhibit methanogens substantially. These might have affected also the hydrogen yields discussed above.

Comparing the results of WSH fermentation under increased OFR (from 3.0 to 4.6 mL/h) one concludes that methane production is gradually inhibited by increasing micro-aeration at least for OFR > 3 mL/h.

Surprisingly, methane production at acidic pH 6.02 was more efficient than at higher pH, although usually neutral or basic pH is preferred (Garcia-Bernet et al., 2017).

3.3. Efficiency of hydrogen production

The process mass and economic efficiency of hydrogen production depends strongly on the kind of substrate. Potato wastes is a cheaper substrate for DF than wheat straw (but not so abundant) with potentially higher hydrogen and methane productivity (see Figs. 2 and 3). Besides it is known from literature – see (Bundhoo, 2019), that wheat straw for efficient hydrogen production needs to be delignified (this increases costs), and hydrolysis of hemicellulose and cellulose requires strict control to obtain high efficiency. The estimations for hydrogen production efficiency from wheat straw and potato wastes are presented in Table 2.

Table 2. Comparison of hydrogen production from wheat straw and potato wastes

Substrates / Process parameters	Potato wastes	Wheat Straw hydrolysed (WSH)
Hydrogen yield in experiment	0.07 L of H_2/g VSS	0.08 L of H_2 /g VSS
Highest hydrogen yield published	0.3 L of H_2 /g VSS (Sekoai et al., 2019)	0.148 L of H_2 /g VSS (Nasirian et al., 2011)
Substrate pretreatment in this study	milling with sieve 10 mm	milling with sieve 10 mm (knives must be changed every 2 h of milling) HCl acid-pretreatment
Process requirements	stressing (boiling) of inoculum, pH ~6.0	stressing (boiling) of inoculum, pH ~6.4

The hydrogen yields obtained here were lower than that obtained by Sekoai et al. (2019), i.e. 0.3 L H_2/g VSS. Sekoai et al. (2019) used special salts and performed their experiments in higher working volumes. Our results were better than those obtained by Laurinavichene et al. (2010), i.e. 0.05 L of H_2/g VSS. Finally, it should be stressed that the estimation of hydrogen production presented here, show that potato waste (when available) is the preferable substrate. However, it is important to determine the effects of a substrates and nutrients blending (Aly et al., 2018).

Besides, substrates other important issue is pH control during fermentation process. For example, Fig. 4 presents the results of cotton-waste (lignocellulose) and sour cabbage DF under controlled pH 6 and without pH control condition. The control of low pH conditions may significantly increase hydrogen production.

One can also see the microaeration effect – fermentation of sour cabbage with oxygen flow rate (OFR) equal 0.6 mL/h results in increase of hydrogen production of about 15%.

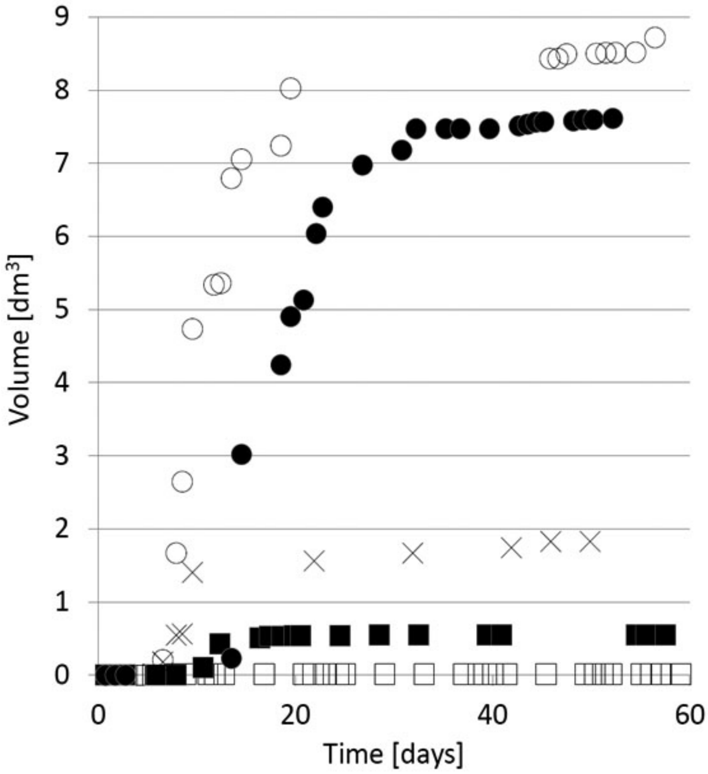

Fig. 4. Cumulative hydrogen production from: cotton with pH 6 ■ or without pH control □ and from sour cabbage with pH 6 (OFR = 0) • and (OFR = 2 mL/h) ○ and without pH control ×; VSS 40 g/L

4. CONCLUSIONS

Dark fermentation of boiled potato wastes is more efficient for hydrogen production than DF of wheat straw. Dark fermentation of wheat straw without delignification requires micro-aeration. Similarly, dark fermentation of potato waste depends on micro-aeration (enhancing the process yields) and varies with pH. Also, methane production from boiled potatoes is more efficient than from wheat straw hydrolysed (WSH).The stressing (boiling) of inoculum dramatically increases hydrogen yield and shortens fermentation time in comparison to process with raw inoculum. Dark fermentation optimization demands: careful choice of potential substrates and process parameters such as OFR and pH as well as their control during process continuation. The phenomena require further research.

ABBREVIATIONS

AD	Anaerobic Digestion
ATEX	Appareils destinés à être utilisés en ATmosphères Explosives
CSA	Canadian Standards Association
VSS	Volatile Suspended Solids
OFR	Oxygen Flow Rate
DF	Dark Fermentation

IECEx	International Electrotechnical Commission System for Certification of Standards Relating to Equipment for Use in Explosive Atmospheres
FM	Fresh Mass
GA	Gas Analyzer
GC	Gas Chromatography
TCD	Thermal Conductivity Detector
TS	Total Solids
UKAS	United Kingdom Accreditation Service
WSM	Wheat Straw Milled
WSH	Wheat Straw Hydrolyzed

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