1 **Characteristics and agronomic usability of digestates from laboratory digesters**

2 **treating food waste and autoclaved food waste**

3 Elina Tampio[a*], Satu Ervasti[a], Jukka Rintala[b]

4 [a]*Natural Resources Institute Finland (Luke), New business opportunities, Tietotie 2 C,*

5 *FI-31600, Jokioinen, Finland*

6 [b]*Tampere University of Technology, Department of Chemistry and Bioengineering, P.O.*

7 *Box 541, FI-33101 Tampere, Finland*

8 **Abstract**

9 Digestate characteristics such as organic and nutrient content, hygienic quality and

10 stability are valuable measures when evaluating the use of food waste (FW) digestate as

11 organic fertiliser. This study compared the characteristics of FW and autoclaved (160

12 °C, 6.2 bar) FW and their digestates from laboratory-scale reactors. Decreased

13 ammonification and low ammonium nitrogen content were observed in the digestate

14 from an autoclaved FW reactor due to autoclave treatment of FW, which affected the

15 nitrogen-containing molecules by formation of Maillard compounds. The methane

16 potential of autoclaved FW and its digestate was decreased by 40% due to reduced

17 microbial activity as microbes were not able to adapt to the conditions within a reactor

18 fed with autoclaved FW. Both studied materials were suitable for agricultural use in

19 terms of their nutrient content, hygienic quality and stability, and thus the decrease in

20 ammonium nitrogen in digestate from an autoclaved FW reactor supported the use of

21 digestate as soil amendment rather than fertiliser.

[*] Corresponding author. Tel.: +358 29 532 6573, E-mail address: elina.tampio@luke.fi

1. Introduction

It is estimated that globally one third of the food produced for consumption becomes food waste (FW) during production, processing, distribution and consumption (Gustavsson et al., 2011). In Europe the total FW quantity produced each year is 90 million tonnes (180 kg per capita), of which an estimated 38 million tons (76 kg per capita) is generated in households (European Commission, 2010), while in the USA 36 million tons (120 kg per capita) of residential and commercial FW was produced in 2011 (US EPA, 2013). FW composition derived from households varies seasonally and geographically. In a study from Finland, Italy, Portugal, and the UK, the average household food waste consisted mainly of fruit and vegetable waste (> 50%) and to a lesser extent of beverages (coffee filters and tea bags, 9%), meat and fish (6%), bread and bakery (5%) and mixed meals (12%), and had relatively high protein (16-55% of VS, volatile solids) and fat (15-30% of VS) contents (Valorgas, 2011).

In Europe anaerobic digestion (AD) together with composting are increasingly used as treatment methods for organic wastes such as FW due to the EU Waste Framework Directive (2008/98/EC, European Parliament and the Council, 2008), which obligates member states to carry out source segregation and safe treatment of biowastes. With AD, energy- and nutrient-rich organic compounds can be digested to simultaneously produce fertilisers/soil amendments, renewable energy and/or fuel for transport. When used as fertiliser the nutrients in FW digestate can be returned to

45   agriculture to close the nutrient cycle, thereby reducing the need for inorganic fertilisers,

46   and their use as soil amendments improves the physical, chemical and biological

47   properties of the soil. In the EU digestate use in agriculture is regulated by national

48   legislations deriving from EU regulations concerning animal by-products and their

49   digestion residues (European Council, 2011; European Parliament and the Council,

50   2009). In addition to the hygienic quality, the fertilising effect of the mineral and

51   organic forms and plant availability of nutrients are essential when considering the

52   usefulness of the digestate as soil fertiliser/amendment. Determination of the stability –

53   e.g. residual methane potential of digestates, emissions during digestate storage and use

54   – can be minimised and the energy production of AD optimised.

55   In biogas production, pretreatment of food waste affects the characteristics of the

56   FW digestate. The aim of pretreatment is to enhance biodegradability and methane

57   yields and to improve the hygienic quality of the material. Thermal autoclave treatment

58   (130-180 °C) has been observed to lower the methane conversion of high protein-

59   containing substrates (Cuetos et al., 2010, Pinnekamp, 1989) such as FW by 5-10%

60   during semi-continuous mesophilic AD (Tampio et al., 2014). At higher autoclaving

61   temperatures organic material hydrolyses and solubilises; however, toxic (Cuetos et al.,

62   2010) or hardly biodegradable compounds such as Maillard compounds can also be

63   formed through reactions between sugars and amino acids (Bougrier et al., 2008,

64   Monlau et al., 2013), which further affects the AD process and the digestate quality, e.g.

65   decreasing the ammonium nitrogen content of the digestate (Tampio et al., 2014).

66   However, more detailed research about the effects of these compounds and nitrogen

67   transformation during AD of pretreated FW is needed to evaluate the end-use value of

68   the digestate.

69   The aim of this study was to compare the characteristics, quality and agronomic

70   usefulness of FW and autoclaved FW (AFW) digestates. For that purpose digestates

71   from laboratory semi-continuously stirred tank reactors were characterised for hygienic

72   quality, nutrient content as well as residual methane and ammonification potentials.

73   Furthermore, as reference the ammonification and residual methane potentials were

74   compared with digestate from a full-scale AD plant.

75   **2. Materials and methods**

76   **2.1. Origin of food waste and digestates**

77   The FW used in this study was source-segregated domestic FW collected from the

78   South Shropshire Biowaste digestion plant in Ludlow, UK. FW was divided into two

79   portions and subsequently one portion was pre-treated with a novel double-auger

80   autoclave (AeroThermal Group Ltd, UK) at 160 °C and 6.2 bars (referred to as AFW)

81   while the other portion was left untreated (referred to as FW). FW portions were then

82   passed through a macerating grinder (S52/010 Waste Disposer, IMC Limited, UK),

83   frozen and shipped to Natural Resources Institute Finland where the FW samples were

84   melted and stored at 4 °C before use as described in more detail in Tampio et al. (2014).

85   Three different digestates were used in this study. Two digestates were collected

86   from laboratory stirred tank reactors fed with FW (digestate referred to as FW digestate)

87   and AFW (digestate referred to as AFW digestate). The reactors were fed through a

88   feeding inlet tube extended below the digestate surface, and digestate overflowed by

89   gravity through a u-tube trap to prevent gas escape. For this study the digestates were

90   sampled both from the overflow digestate and through the inlet tube from the reactor.

91   The reactors were operated up to 473 days. Organic loading rates were gradually

92 increased from 2 to 6 kgVS/m^{3}d, decreasing the hydraulic retention times from 117 and

93 94 to 39 and 31 days in reactors treating FW and AFW, respectively (Table 1). Starting

94 from runs with organic loading rate of 3 kgVS/m^{3}d the reactors were supplemented with

95 trace elements according to Banks et al. (2012) with element concentrations of Al (0.1

96 mg/l), B (0.1 mg/l), Co (1.0 mg/l), Cu (0.1 mg/l), Fe (5.0 mg/l), Mn (1.0 mg/l), Ni (1.0

97 mg/l), Zn (0.2 mg/l), Mo (0.2 mg/l), Se (0.2 mg/l) and W (0.2 mg/l). Reactor

98 configuration and feeding practices are described in more detail in Tampio et al. (2014).

99 Digestates were stored at 4 °C for a maximum of one week (characterisation and

100 hygiene analysis) or up to 4 weeks (batch assays) before use. Digestates used in

101 characterisation studies were from organic loading rates of 2, 3 and 6 kgVS/m^{3}d (total

102 organic carbon was analysed during organic loading rate 4 kgVS/m^{3}d) while digestate

103 samples for the hygiene analyses were collected during organic loading rates of 4

104 kgVS/m^{3}d (4 samples) and 6 kgVS/m^{3}d (3 samples) (Table 1). The food waste samples

105 were collected simultaneously with the digestates (6 to 7 samples) and thawed and

106 stored in a freezer (4 °C) for 1-5 days prior to analyses.

107 The third digestate (referred to as reference digestate) used in this study originated

108 from a full-scale mesophilic anaerobic digester treating municipal and industrial

109 biowastes (Envor Biotech Ltd, Forssa, Finland).

110 **2.3. Batch assays**

111 The batch assays for biochemical methane potentials and for residual methane and

112 ammonification potentials were performed in duplicate or triplicate 0.5 L bottles with a

113 total liquid volume of 400 ml using automated testing equipment (Bioprocess Control

114 Ltd, Sweden) at 37 °C. The contents were mechanically mixed (84 rpm) for one minute

115 per hour, and CO_2 from the produced biogas was fixed by NaOH prior to automated,

116 liquid displacement-based gas volume measurement.

117   Batch assays were performed with the digestates alone (residual methane

118 potential) and using the digestates as inocula and FW and AFW as substrates

119 (biochemical methane potential, Table 1). In all assays with FW and reference

120 digestates the volume of inoculum was 300 g and the substrate to inoculum ratios on a

121 VS basis 1:1. With AFW digestate assays 340 g of inoculum was used with a VS/VS

122 ratio of 1:2. In all assays distilled water was added to obtain 400 ml liquid volume. pH

123 (if lower than 7.3) was adjusted to around 8 with 3 M NaOH and in the case of the

124 reference digestate, inoculum $NaHCO_3$ (3 g/l) was added as a buffer. Finally, the

125 contents of all bottles were flushed with N_2 to obtain anaerobic conditions.

126 **2.4. Analyses and calculations**

127   From fresh samples, total and volatile solids (TS and VS) were determined

128 according to SFS 3008 (Finnish Standard Association, 1990) and ammonium nitrogen

129 (NH4-N) according to McCullough (1967). Total Kjeldahl nitrogen (TKN) was

130 analysed by a standard method (AOAC, 1990) using a Foss Kjeltec 2400 Analyser Unit

131 (Foss Tecator AB, Höganäs, Sweden), with Cu as a catalyst. For soluble chemical

132 oxygen demand analysis FW samples were diluted 1:10 with distilled water, and

133 agitated for 1 hour. Diluted FW and digestate samples were centrifuged ($2493 \times g$, 15

134 min) after which the supernatant was further centrifuged ($16168 \times g$, 10 min) and stored

135 in a freezer, then thawed before analysis according to SFS 5504 (Finnish Standard

136 Association, 2002). pH was determined using a VWR pH100 pH-analyser (VWR

137 International). Soluble-N was analysed as TKN after 1:15 dilution with distilled water

138   and soluble-P and soluble-K were measured from 1:5 dilution with ICP-OES

139   (inductively coupled plasma optical emission spectrometry).

140   From dried (60 °C) samples, crude protein by Duma's method was analysed with

141   standard methods (AOAC, 1990) using a Leco FP 428 nitrogen analyser (Leco Corp., St

142   Joseph, USA) and by multiplying the N% by a factor of 6.25. Crude fat was analysed

143   with a Soxcap-Soxtec-Analyser (AOAC,1990; Foss Tecator Application Note AN 390).

144   For soluble carbohydrate analyses, samples were inverted with 1 N HCl (50 °C, 12 h)

145   and analysed according to Somogyi (1945). NDF (neutral detergent fibre) was analysed

146   with a filtering apparatus according to Van Soest et al. (1991) and both ADF (acid

147   detergent fibre) and lignin (permanganate-lignin) were determined according to

148   Robertson and Van Soest (1981). Hemicellulose content was calculated from the

149   difference between NDF and ADF while cellulose content was calculated from the

150   difference between ADF and lignin. Total-C was analysed by Duma's method

151   according to manufacturer's instructions with a Leco CN-2000 Elemental Analyser

152   (Leco Corp., St. Joseph, MI, USA). For the analysis of total-P and total-K, samples

153   were digested with HNO_3 (Luh Huang et al., 1985) and analysed with ICP-OES

154   according to manufacturer's instructions.

155   Hygienic quality was analysed using *Escherichia coli*, other coliforms, total

156   coliforms, enterococci, sulphite-reducing clostridia and Salmonella as indicator

157   organisms. Analyses of different coliforms were performed according to Baylis and

158   Patrick (1999) using Harlequin *E. coli* / coliform (LabM) culture medium with 24-48 h

159   incubation time at 37 °C. Enterococci were determined with KF streptococcus agar

160   (incubated 48 h at 44.5 °C) according to SFS-EN ISO 7899 (Finnish Standard

161   Association, 2000) and sulphite-reducing clostridia with sulphite-iron agar (incubated

162 anaerobically 48 h at 37 °C) according to SFS-EN 26461 (Finnish Standard Association,

163 1993). For the qualitative analyses of Salmonella, samples were pre-enriched in

164 buffered peptone water (37 °C, 16-20 h) and incubated in Rappaport-Vassiliadis broth

165 (42 °C, 24 h). Aliquots from the broth were cultured on Salmonella-selective Rambach

166 and xylose-lysine-decarboxylase agars and incubated at 42 °C for 24 h. If growth was

167 observed, colonies were confirmed with triple sugar iron agar, urea-agar and lysine

168 carboxylase broth (37 °C, 24 h) (ISO, 2002).

169   All methane yields were converted into the standard temperature and pressure

170 conditions (0 °C, 100 kPa) according to the ideal gas law using ambient temperature and

171 air pressure. In the ammonification batch assays, the starting NH_4-N, total Kjeldahl

172 nitrogen, TS and VS contents in the bottles were calculated according to the mass

173 balances from the original concentrations of FWs and digestates and the amounts used

174 in the assays.

175 **3. Results and discussion**

176 **3.1. Food waste characteristics**

177   The studied FW had TS of ca 230 g/kgFM, and VS/TS ratio of 93% while AFW

178 had about 10 to 15% lower TS and VS, likely due to dilution by condensed water during

179 the autoclave treatment (Table 2). The FW contained proteins up to 220 g/kgTS while

180 fats and soluble carbohydrates were ca 140 and 120 g/kgTS. Cellulose and

181 hemicellulose contents were around 50 g/kgTS and low lignin content, 6 g/kgTS, was

182 observed. The autoclaving affected the organic composition (per TS) by decreasing the

183 soluble carbohydrates by 50% and hemicelluloses by 40% while increasing the lignin

184 content from 6.6 to 81.6 g/kgTS, whereas the effects on other components were minor.

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185 AFW also had increased SCOD and lowered VFA, likely due to solubilisation,

186 volatilisation and acidification of material during autoclave treatment.

187   The protein (220 g/kgTS) and fat (140 g/kgTS) content in the FW corresponded

188 well with previous studies with FWs from Europe where protein and fat contents in

189 FWs have varied between 100-260 g/kgTS (Table 3). Cellulose and hemicellulose

190 contents were similar in the source-sorted FW in Ludlow, UK, while the present lignin

191 content of 6 kg/kgTS was 60% lower (Table 3, Zhang et al., 2012). The low lignin

192 content of FW as well as the high standard deviations in lignin observed with both FW

193 samples were probably due to the complex nature of lignin, different analysing methods

194 (Hatfield and Fukushima, 2005) and the heterogeneity of the FW material

195 (Papadimitriou, 2010). The autoclave treatment decreased the soluble carbohydrate

196 content, indicating the formation of Maillard compounds (Liu et al., 2012, Monlau et

197 al., 2013) through reactions between sugars and amino acids (Bougrier et al., 2008,

198 Monlau et al., 2013, Pinnekamp, 1989). The reduction in hemicellulose content was

199 most likely due to the branched structure of the hemicellulose, which enables easier

200 hydrolysis during pre-treatment (Papadimitriou, 2010, Pérez et al., 2002).

201 **3.2. Digestate characteristics**

202   The FW digestate had TS and VS of 67.4 and 45.6 g/kg, while the values were

203 slightly higher in the AFW digestate (78.5 and 50.5 g/kg, respectively, Table 2). AD

204 decreased TS, VS, fats and soluble carbohydrates content and increased cellulose and

205 hemicellulose contents (g/kgTS) similarly with both substrates. However, the lignin

206 content increased nearly tenfold in the FW while in the autoclaved digestate lignin

207 content was doubled. Protein content increased by 15% more with the autoclaved

208 material during AD.

209   With AFW digestate the protein content and the hemicellulose, cellulose and

210 lignin contents (g/kgTS) were 25-80% higher than for FW digestate while the NH4-

211 N/TKN ratio was ~30% lower (Table 2). The reduced NH4-N and NH4-N/TKN ratio

212 and higher protein contents in the AFW digestate resulted from formation of Maillard

213 compounds during autoclave treatment, which affected the digestate by decreasing

214 protein degradation and leading to reduced fertiliser value.

215   The content of fibres (cellulose, hemicelluloses and lignin; g/kgTS) increased 30-

216 800% during AD partly due to low biodegradability of the ligno-cellulosic complexes

217 (Pérez et al., 2002), but also indicating some solid material accumulation during the

218 digestion process. The ratio between cellulose (CEL), hemicellulose (HEMI) and lignin

219 (LIGN), CEL+HEMI/LIGN (Eleazer et al., 1997), was used to evaluate the

220 biodegradation of these compounds during autoclaving and AD. For FW, AFW, FW

221 and AFW digestates, the CEL+HEMI/LIGN ratio was 16.3, 1.2, 3.0 and 1.2,

222 respectively. The stable CEL+HEMI/LIGN ratio (1.2) of AFW after AD indicates that

223 the hemicellulose and cellulose had already degraded during autoclaving and could not

224 degrade further during AD. The higher content of hardly degradable cellulose, lignin

225 and proteins in the AFW digestate compared to the FW digestate likely reduced

226 methane production during batch experiments, which supports the results from Tampio

227 et al. (2014) where the methane yield in stirred tank reactors was 5-10% lower with

228 AFW compared to FW.

229 **3.3. Methane and ammonification potentials**

230   First, the residual methane potentials of the FW, AFW and reference digestates

231   were assayed to evaluate the potential recoverable methane and possible emission risk

232   during digestate handling. The FW digestate produced methane more slowly than the

233   AFW and reference digestates; however, it and the reference digestate had higher

234   residual methane potential (around 0.135 m^3CH_4/kgVS) than the AFW digestate

235   (~0.080 m^3CH_4/kgVS). With the FW digestate the cumulative methane potential curve

236   was of a "sigmoid type", indicating some inhibition (Vavilin et al., 2008). During the

237   assay, the NH4-N concentration in the AFW digestate increased by 0.95 g/kgFM while

238   with the other two digestates NH4-N increase was ca 0.3 g/kgFM (Figure 1, Table 4).

239   Secondly, the three digestates were assayed as inocula to digest both FW and

240   AFW to assess the effect of long-term cultivation (>300 days in stirred tank reactors) on

241   micro-organisms' capability to degrade FW and AFW. Both FW and AFW digestate

242   inocula produced 0.451 m^3CH_4/kgVS from FW while from AFW the biochemical

243   methane potential was 10% less with FW digestate and as much as 30% less with AFW

244   digestate as inoculum. With the reference digestate higher methane potentials were

245   observed with both FW and AFW. Both FW and reference digestate inocula degraded

246   ca 50% of VS with both FWs while with the AFW digestate the VS removals were

247   around 37%. However, with the low VS removal AFW digestate produced as much

248   methane from FW using FW digestate as inoculum (0.451 m^3CH_4/kgVS; Table 4).

249   During the assays with digestate inocula and FWs NH4-N concentration increased

250   (inoculum excluded) more with the FW digestate (0.68 and 0.34 g/kgFM) than with the

251   AFW digestate (0.41 and 0.17 g/kgFM) assayed with both FW and AFW, respectively,

252   while the highest NH4-N increases were obtained with reference inoculum (0.73 and

253   0.51 g/kgFM with FW and AFW; Table 4).

254   The lower biochemical methane potentials, VS removals and decreased NH4-N

255   formation with AFW along with low NH4-N starting concentration (~1 g/kg) with AFW

256   digestate were connected to the formation of hardly degradable Maillard compounds

257   during the autoclave treatment of FW, leading to reduced biodegradability of the

258   material (Bougrier et al., 2008, Monlau et al., 2013), which was previously reported to

259   decrease NH4-N concentration in anaerobic digesters (Tampio et al., 2014).

260   Combination of AFW and AFW digestate most likely inhibited the growth of certain

261   microbes due to decreased protein degradation, leading to ca 40% reduced biochemical

262   methane potential. Also the higher initial VS content in the AFW digestate assay bottles

263   (22.6 gVS/bottle versus 13.4 and 9.8 gVS/bottle with FW and reference digestates) may

264   have caused inhibition due to VFA accumulation (Lesteur et al., 2010), decreasing the

265   residual methane potential of the AFW digestate. However, the initial VS

266   concentrations did not correlate with the biochemical methane potential results with

267   FWs.

268   The FW digestate showed good gas production with both FWs studied as did the

269   reference digestate, indicating the capability of microbes to degrade the feed material.

270   However, the AFW digestate showed lower methane production with both FWs, which

271   indicates that the adaptation of the microbial population towards the AFW was not

272   successful. Prior studies have shown that autoclaving of FW changes the microbial

273   populations, especially bacteria, during AD (Blasco et al., 2014) due to the

274   transformation of proteins, leading to further decreases in methane yields during AD.

275   With these batch experiments it was confirmed that the autoclaving of FW affected the

276   ammonification capacity of the digestate, which led to reduced methane formation.

277   **3.4. Hygienic quality**

278   The hygienic quality of the FW (7 samples) and digestate (6 samples) were tested

279   with hygiene indicators *E. coli*, other coliforms, total coliforms, enterococci and

280   sulphite-reducing clostridia and Salmonella (Figure 2). No Salmonella was detected in

281   any of the feed or digestate samples (data not shown). In one of the six FW samples a

282   few colonies of *E. coli* were discovered, while both enterococci (average $2.79 \times 10^4 \pm$

283   2.74×10^4 cfu/g) and clostridia ($2.24 \times 10^3 \pm 1.86 \times 10^3$ cfu/g) were also discovered. In

284   AFW all hygiene indicators were under the detection limit (5 cfu/g). In both digestates,

285   high enterococci concentrations (6.77×10^8 cfu/g $\pm 7.40 \times 10^8$ and $3.71 \times 10^8 \pm 4.64 \times$

286   10^8 in the FW and AFW digestate) were detected, while the clostridia concentration

287   remained lower (6.14×10^2 cfu/g $\pm 4.98 \times 10^2$ and $6.48 \times 10^3 \pm 6.29 \times 10^3$ cfu/g in the

288   FW and AFW digestates, respectively).

289   The absence of coliforms in the studied FW was likely due to the freezer storage

290   time (before preparation as feed and analysis). In fresh FW these indicators have usually

291   been detected in concentrations of $10^4 – 10^5$ cfu/g in biogas plants treating FW as such

292   (Sahlström et al., 2008) and co-digesting FW with manures and animal by-products

293   (Bagge et al., 2005). The present concentration of enterococci was similar to that

294   reported for fresh FW (around 10^4 cfu/g) by Sahlström et al. (2008) due to the resistance

295   of enterococci towards freezing (Geiges, 1996). Similarly high concentrations of

296   sulphite-reducing clostridia were detected as these spore-forming organisms are also

297   resistant to freezing (Geiges, 1996).

298   The results show that the studied autoclave treatment effectively reduced all the

299   hygiene indicator concentrations in AFW due to high temperature and pressure, which

300   are widely used for sterilisation. However, the observed increase in concentrations of

301   enterococci and clostridia (up to 8 logs) in the AFW digestate clearly indicates the

13

302 potential of hygienised material for microbial growth. The increase was apparently due

303 to growth of indicator organisms in the stirred tank reactors, originating from the sludge

304 with which the reactors were inoculated or possibly from contamination of the AFW

305 samples. Absence of coliforms in the studied digestates indicates that either there were

306 no coliforms in the original inoculum or the microbes were not able to survive due to

307 competition of microbial communities while the conditions were favourable for

308 clostridia and enterococci.

309 Altogether, according to the EU's Animal By-Product regulations (European

310 Council, 2011; European Parliament and the Council, 2009) digested FW and digested

311 autoclaved FW were both hygienically suitable for land application as the concentration

312 of *E. coli* was under the threshold value 1000 cfu/g and no Salmonella was detected.

3.5 Agronomic usefulness of digestates

314 The total and soluble nutrient composition of the FWs and digestates was studied

315 to evaluate the agronomic usefulness of the digestates (Table 2). The total nutrient

316 levels of nitrogen (31 gN/kgTS), potassium (11 gK/kgTS) and carbon 470-487

317 (gC/kgTS) were similar between the studied FWs, and thus the AFW had a higher total-

318 P content (~7 g/kgTS in AFW, 4 g/kgTS in FW). The soluble P and K contents in both

319 FWs were around 1.7 and 9 g/kgTS while the soluble-N concentration increased from

320 10 to 16 g/kgTS after autoclaving. When digestates were compared the AFW digestate

321 had 20% lower total Kjeldahl nitrogen and 44% lower soluble-N levels compared to the

322 FW digestate. The C/N ratios were relatively low with both studied digestates (3.3-4.5),

323 which was due to the mineralisation of carbon during AD.

324   The total nutrient concentration in FWs correlated well with different European

325   (UK, Finland, Italy) food wastes, where total-N concentrations varied between 24-34

326   g/kgTS, total-P between 2.7-6.4 and total-K between 8.6-14.3 g/kgTS (Valorgas, 2011).

327   Only total-P was observed in slightly higher concentrations in the AFW where some

328   additional phosphorus could have dissolved from the autoclaving apparatus due to P

329   impurities in steel. Soluble N increase after autoclaving was probably due to

330   solubilisation of nitrogen into other compounds than NH_4-N, e.g. to soluble Maillard

331   compounds.

332   In the digestates the NPK-ratios (per TS) were 100:17:38 in the FW digestate and

333   100:17:33 in the AFW digestate, which were similar to the results obtained with source-

334   sorted FW in the UK (NPK 100:11:41; Zhang et al., 2012). Compared to available

335   commercial fertilisers (~20 %N) the N content in the FW digestate was low but the

336   proportion of K and P was higher, and thus it was considered to be a suitable fertiliser

337   for leguminous plants (Israel, 1987) and plants at reproductive state (Clemens &

338   Morton, 1999). However, when considering the low NH4-N/TKN ratio of the AFW

339   digestate (26%) compared to the FW digestate (52%), the AFW digestate was evaluated

340   to be more suitable for use as soil amendment than fertiliser (Nkoa, 2013). The 10-15%

341   lower N-tot and K-tot concentrations (per FM) would also increase the volume of AFW

342   digestate needed for fertilising in similar quantities. The TS contents of the studied

343   digestates were 67 g/kgFM (FW digestate) and 79 g/kgFM (AFW digestate), which are

344   similar to those of manure used as fertiliser in agriculture (Amon et al., 2006), enabling

345   the spreading of digestates with similar machinery as manure.

346   Calculated with the values obtained from this study the FW produced in Europe

347   (38 million tonnes; European Commission, 2010) accounts for approximately 296 000

348   tonnes of N, 46 200 tonnes of P and 108 000 tonnes of K. These calculated values

349   represent 2.8, 4.5 and 5.0% of the manufactured fertilisers consumed in the EU (10.4 Mt

350   of N, 1.0 Mt of P, 2.2 Mt of K; Eurostat, 2013). With European FW, approximately 1.74

351   million hectares of field could be fertilised, using an assumed N fertilisation rate of 170

352   kg/ha.

353   **4. Conclusions**

354   Anaerobic digestion of high protein-containing FW produces digestates with

355   relatively high NH4-N (4 g/kgFM), which supports its use as a fertiliser in agriculture.

356   Also the hygienic quality, nutrient concentrations (NH4-N, P, K), TS content and low

357   residual methane emission potential facilitate fertilisation use.

358   Anaerobic digestion of autoclaved FW results in digestate with higher undegraded

359   protein and lower ammonium content than without autoclaving, leading to reduced

360   microbial activity and decreased methane yield in batch assays. This increases the

361   volumes needed to achieve the desired fertilising effect by approximately 10-15%

362   compared to FW; this, coupled with its low ammonium content, supports the use of

363   autoclaved FW digestate in soil amendment practices.

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513 Table 1. Source of digestates used for characterisation, hygiene analyses and batch

514 assays. Organic loading rate (OLR) and hydraulic retention time (HRT) of the reactors

515 and supplementation of trace elements (TEs) are shown for time of sampling as well as

516 the sampling procedures. FW=food waste, AFW=autoclaved food waste.

Digestate	OLR	HRT	TE	Sampling feeding inlet	Sampling overflow
FW	2	117	-	Characterisation	-
AFW		94	-	Characterisation	-
FW	3	78	+	Characterisation	-
AFW		58	+	Characterisation	-
FW	4	63	+	Hygiene	Batch assays
AFW		47	+	Hygiene	-
FW	6	39	+	Hygiene	-
AFW		31	+	Characterisation, hygiene	Batch assays
Reference	N/A	N/A	-	-	Batch assays

-, no trace elements addition or sampling

+, trace element addition

N/A, not available

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526   Table 2. Characteristics of food waste (FW) and autoclaved food waste (AFW) as well

527   as FW and AFW digestates. Averages and standard deviations are shown, feed N=3-4,

528   FW digestate N=2, AFW digestate N=3, if not otherwise stated.

Parameter	Unit	Feed		Digestate	
		FW	AFW	FW	AFW
General characteristics					
pH	-	5.2 ± 0.22	5.2 ± 0.21	8.0 ± 0.02	7.7 ± 0.05
TS	g/kgFM	248.6 ± 2.86	215.5 ± 8.66	67.4 ± 0.07	78.5 ± 5.12
VS	g/kgFM	231.1 ± 1.93	198.8 ± 7.50	45.6 ± 2.96	60.5 ± 6.53
VS/TS	%	92.6 ± 0.29	92.5 ± 0.29	67.7 ± 4.33	77.0 ± 3.72
TKN	g/kgFM	7.62 ± 0.33	6.9 ± 0.27	7.8 ± 0.59	7.3 ± 0.52
NH4-N	g/kgFM	0.4 ± 0.14	0.4 ± 0.03	4.07 ± 0.25	1.9 ± 0.41
NH4-N/TKN	%	4.7 ± 1.71	5.3 ± 0.53	52.2 ± 0.66	25.7 ± 7.18
SCOD	g/kgFM	101.7 ± 12.55	112.8 ± 16.19	13.1 ± 1.51	15.3 ± 1.28
VFA	g/kgFM	3.5 ± 0.41	2.2 ± 0.21	0.3 ± 0.01	0.2 ± 0.03
Organic characteristics					
Crude protein	g/kgTS	218.9 ± 17.51	208.6 ± 26.96	311.2 ± 31.82	443.4 ± 36.08
Crude fat	g/kgTS	141.7 ± 9.48	142.5 ± 6.22	56.7 ± 3.39	46.1 ± 6.75
Soluble carbohydrate	g/kgTS	122.7 ± 17.94	59.7 ± 5.38	5.2 ± 0.00	5.2 ± 0.64
Cellulose	g/kgTS	51.5 ± 6.94	62.5 ± 9.62	66.4 ± 16.69	123.5 ± 23.20
Hemicellulose	g/kgTS	56.2 ± 6.97	35.9 ± 8.14	81.6 ± 12.37	108.2 ± 8.07
Lignin	g/kgTS	6.6 ± 8.29	81.6 ± 10.72	40.8 ± 2.47	192.9 ± 12.15
(CEL+HEMI)/LIGN	-	16.32	1.21	3.63	1.20
Total nutrients					
Total-C[a]	g/kgTS	469.1	486.6	386.1	415.4
TKN	g/kgTS	30.7 ± 1.68	32.1 ± 1.62	115.6 ± 8.38	93.2 ± 3.23
C/N		15.3	15.2	3.3	4.5
Total-P	g/kgTS	3.8 ± 0.06	6.5 ± 1.31	19.9 ± 3.63	16.2 ± 2.63
Total-K	g/kgTS	11.4 ± 1.57	10.31 ± 0.41	44.1 ± 8.64	30.7 ± 1.73
Soluble nutrients					
Soluble-N	g/kgTS	9.6 ± 0.52	16.3 ± 0.44	74.9 ± 6.75	42.2 ± 4.33
Soluble-P	g/kgTS	1.7 ± 0.75	1.7 ± 0.28	2.6 ± 1.09	1.4 ± 0.55
Soluble-K[a]	g/kgTS	9	9	22.6	26.3

[a] N=1

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531　Table 3. Characteristics of food wastes in various European countries. Organic fraction

532　of municipal solid waste (OFMSW), restaurant waste (RW), household waste (HW),

533　food waste (FW), autoclaved food waste (AFW), source-sorted (ss), mechanically

534　recovered (mr).

Waste	Country	Protein (g/kgTS)	Fat (g/kgTS)	Cellulose (g/kgTS)	Hemicellulose (g/kgTS)	Lignin (g/kgTS)	Reference
ss-OFMSW	Denmark	105-171	102-177	N/A	N/A	N/A	Hansen et al., 2007
RW	Spain	275	288	N/A	N/A	N/A	Garcia et al., 2005
HW	Spain	163	113	N/A	N/A	N/A	Garcia et al., 2005
FW	Finland	169	175	N/A	N/A	N/A	Valorgas, 2011
FW	Italy	233	215	N/A	N/A	N/A	Valorgas, 2011
FW	UK	161-172	194-257	N/A	N/A	N/A	Valorgas, 2011
ss-FW	UK	257	165	55	42	18	Zhang et al., 2012
mr-OFMSW	UK	204	108	397	82	289	Zhang et al., 2012
FW	UK	219	142	52	56	7	Present study
AFW	UK	209	143	60	34	82	Present study

N/A, not available

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538   Table 4. Initial TS, VS and NH4-N (g/kgFM) during batch assays with different inocula

539   (FW, AFW and reference digestates) and food waste (FW) and autoclaved food waste

540   (AFW) as substrates. Residual methane potentials (RMPs) and biochemical methane

541   potentials (BMPs) are shown with standard deviations (N=2-3).

Inoculum	FW digestate			AFW digestate			Reference digestate		
Added substrate	-	FW	AFW	-	FW	AFW	-	FW	AFW
Characteristics									
TS initial (g/kg)	49.8	85.7	86.1	69.9	99.6	99.5	38.6	65.1	65.2
TS final (g/kg)	43.9	53.0	54.7	61.9	67.9	68.8	32.2	41.8	45.0
VS initial (g/kg)	33.4	66.8	66.8	56.6	84.1	84.1	24.4	48.9	48.9
VS final (g/kg)	27.5	33.6	35.4	48.3	52.5	53.4	19.1	24.03	26.3
VS removal (%)	17.6	49.8	47.1	14.6	37.5	36.6	21.9	50.8	46.2
TKN initial (g/kg)[a]	6.08	7.17	7.45	5.48	7.11	7.00	N/A	N/A	N/A
NH4-N initial (g/kg)[a]	3.02	3.09	3.11	1.03	1.05	1.07	1.31	1.35	1.35
NH4-N final (g/kg)	3.31	4.07	3.75	1.98	2.41	2.19	1.64	2.41	2.19
NH4-N increase (g/kg)	0.3	0.98	0.64	0.95	1.36	1.12	0.33	1.06	0.84
NH4-N increase, inoculum excluded (g/kg)	N/A	0.68	0.34	N/A	0.41	0.17	N/A	0.73	0.51
RMP or BMP measured (m^{3}CH$_4$/kgVS)	0.132 ± 0.002	0.452 ± 0.001	0.411 ± 0.002	0.079 ± 0.003	0.451± 0.004	0.307 ± 0.003	0.139 ± 0.007	0.501 ± 0.020	0.445 ± 0.001

-, no FW added

N/A, not available/applicable

[a] calculated value
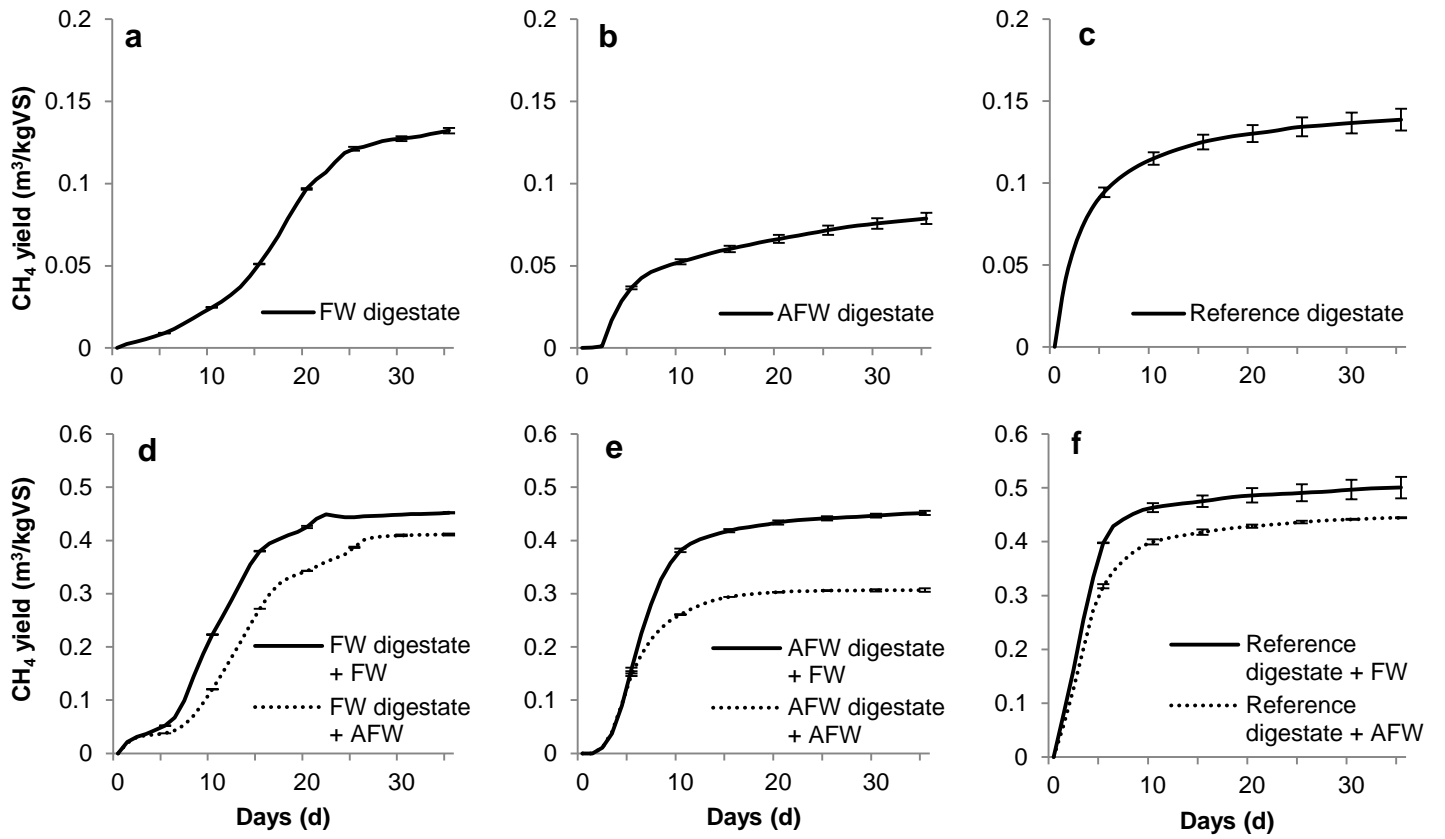
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548   **Figure Captions**

549   **Figure 1.** Residual methane potentials (RMP) of food waste (FW, a), autoclaved FW

550   (AFW, b) and reference (c) digestates. Biochemical methane potentials (BMPs) of FW

551   and AFW digested with FW digestate (d), AFW digestate (e) and reference digestate (f)

552   (inoculum RMP subtracted). Error bars represent standard deviations and are plotted in

553   five-day intervals, N=2-3.

554   **Figure 2.** Hygienic quality of food waste (FW), autoclaved food waste (AFW) and FW

555   and AFW digestates. Averages and positive standard deviations are shown, N=6-7.
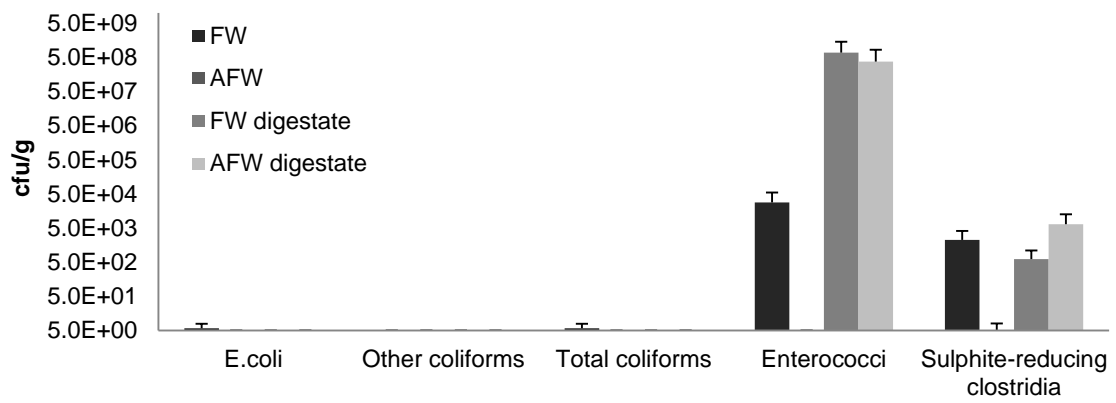
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557   Fig.1.

571   Fig. 2.

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