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Native stinging nettle (*Urtica dioica* L.) growing spontaneously under short rotation coppice for phytomanagement of trace element contaminated soils: Fibre yield, processability and quality

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19 **Abstract**

20 This work assesses the potential of stinging nettle (*Urtica dioica* L.) growing on trace
21 element contaminated soils to produce fibres for material applications. The nettles studied
22 in this work grew spontaneously and dominated the vegetation cover in poplar short
23 rotation coppices planted for the phytomanagement of lands contaminated by trace
24 elements. Two sites were studied, contaminated by Hg for the first one and a mix of As,
25 Cd, Pb and Zn for the second one. Results show that, for the considered soils, the
26 contaminant contents in nettle bast fibres were at low levels, comparable to those
27 collected at unpolluted control areas. It makes it possible to consider this biomass for
28 material use. The measured matter yield was lower than those obtained with traditional
29 fibre crops cultivated in Europe on agricultural lands. However, the tensile properties of
30 the bast fibres mechanically extracted **without field retting or prior alkaline** treatment
31 were equal to or better than those of industrial hemp and flax, making spontaneous nettles
32 an interesting supplement to traditional European fibre crops for material applications.

33

34 **Key words:** Nettle fibre, phytomanagement, contaminated soils, fibre yield, tensile
35 properties

36

37 1 Introduction

38 Environmental contamination is a threat to global sustainable development. The **United Nations**
39 **(UN) Sustainable Development Goals (SDGs)** have a strong focus on reducing and managing
40 environmental pollution. **Among the various soil** phytoremediation options, phytostabilisation is
41 a sustainable and profitable use of marginalized lands (Pilon-Smits, 2005) that can remediate soil
42 layers in contact with the roots and at the same time provide tree biomass (Robinson et al., 2006),
43 which can be used to reach the targets for the use of renewable energy sources (Ciadamidaro et
44 al., 2017; Mench et al., 2010). It creates value from contaminated land while minimising
45 environmental risks and even rendering **ecosystemic services by promoting biodiversity,**
46 **improving the carbon storage and limiting soil erosion.** Interestingly, it has been observed
47 simultaneous growth of poplars under short rotation coppice (SRC) cropping systems and
48 spontaneous dioecious nettle in phytomanaged land sites, thus constituting a multilevel canopy.
49 Biomass from nettle could be used to extend the productivity of the marginal land from SRC
50 alone, whilst still maintaining a functioning phytomanagement system of land rehabilitation. This
51 nettle biomass could constitute a relevant fibre source and thus contribute to answer the increasing
52 demand in plant fibres for material application, by mitigating at the same time the land-use
53 conflict between the needs of food and non-food production. Indeed, recent literature underlines
54 that the use of marginal land is a sustainable method to expand purpose-grown biomass and
55 essential for meeting the emerging massive requirement for biomass in the future (Mohanty et al.,
56 2018).

57 *Urtica dioica* L., often called common nettle or stinging nettle, is a herbaceous perennial
58 flowering plant in the family *Urticaceae*, living up to 10 to 15 years and growing up to 2 m in
59 height (Gravis, 1885). Each stem is formed of a succession of nodes and internodes containing
60 bast fibres in the bark. At each node, two opposite leaves and four cauline stipules are inserted.
61 As for all the fibre crops, the fibre features depend on the living conditions, the plant maturity,
62 the position in the stem (Gravis, 1885). The morphological features of nettle bast fibres were
63 accurately characterised in historical and encyclopaedic works (Gravis, 1885; Vétillart, 1876) and

64 more recently in the scientific literature (Di Virgilio, 2013). The fibres, in limited amount, are
65 distributed in small numbers in the parenchyma in an irregular manner. Most of the fibres are oval
66 in shape, with a diameter generally comprised between 20 and 40 μm . They have a relatively thin
67 wall and thus a very large inner cavity. The fibre wall is mainly composed of cellulose,
68 hemicellulose and lignin, in amount generally comprised in the following ranges 53-86, 4-12.5
69 and 2-10% respectively, depending on the plant age, maturity, growing and processing conditions
70 (Bacci et al., 2010; Di Virgilio, 2013; Di Virgilio et al., 2015). **When nettle is cultivated, dry stalk**
71 **yields could be comprised between 3 and 12 t.ha⁻¹ (Bacci et al., 2009; Dreyer et al., 1996; Vogl**
72 **and Hartl, 2003).** As the two other European fibre crops (flax and hemp), nettle (*Urtica dioica* L)
73 has a long history as a textile fibre and its potential as reinforcement fibres in composite
74 applications was also investigated more recently (Bacci et al., 2010; Di Virgilio et al., 2015;
75 Dreyer et al., 2002; Franck, 2005; Harwood and Edom, 2012; Vogl and Hartl, 2003). Indeed, its
76 bast fibres have remarkable tensile strength and fineness, along with a low density. Average
77 tensile strength and rigidity up to 1600 MPa and 90 GPa have been reported in literature (Bodros
78 and Baley, 2008; Davies and Bruce, 1998; Lanzilao et al., 2016). The stalks and shives can also
79 be used to manufacture fibreboards (Akgül, 2013). As underlined by Di Virgilio et al. (Di Virgilio
80 et al., 2015), despite a high market potential, the products made from nettle are currently more a
81 result of curiosity rather large-scale industrial production, mostly due to lack in crop and post-
82 harvest management.

83 The present study is part of a wider research project, which main objective is the multi-
84 purpose valorisation of wood and plant fibres produced on marginal lands, and focused more
85 precisely on the phytomanagement of trace element (TE) contaminated lands. It aims at
86 investigating the potentialities of nettle fibres growing spontaneously under SRC grown poplar at
87 two phytomanagement sites, for material application. **Hg is the main contaminant at the first site**
88 **due to the chlor-alkali process (Durand et al., 2017), whereas As, Cd, Pb and Zn are the main**
89 **contaminants at the second site due to contamination of deposit sediments from the canal Escaut.**
90 **(Ciadamidaro et al., 2017). This work addresses several issues such as the potential accumulation**

91 of TE in the bast fibres. The potential influence of contamination on the plant and fibre yields and
92 on the physical properties of the bast fibres are also studied by comparing the data obtained on
93 the contaminated sites to data from literature collected on non-contaminated soils. Regarding TE
94 accumulation in plants, previous studies (revealed that nettle is a poor TE accumulating specie
95 when grown on contaminated soils, as compared to woody accumulating or hyperaccumulating
96 species (Phanthavongsa et al., 2017). Indeed, in our previous work, Zn and Cu contents in nettle
97 leaves collected at a sediment landfill were within physiological ranges (<100 mg/kg DWt for Zn
98 and <10 mg/kg DWt for Cu), and Cd content was < 10 mg/kg DWt) (Phanthavongsa et al., 2017);
99 (Codling and Rutto, 2014; Tack and Verloo, 1996). However, there is no clear conclusion
100 regarding the content in stems and bast fibres. For other fibre crops, such hemp, it was previously
101 demonstrated that bast fibres and shives were not affected by TE contamination (Linger et al.,
102 2002). The fibre features (fineness, strength) were also not altered by the contamination making
103 this type of crop a relevant candidate for phytostabilisation option.

104

105 **2 Materials and methods**

106 2.1 Site description

107 The experimental sites are located in Tavaux (France, Bourgogne Franche Comté region, lat. 47°
108 5' 5.98'' N, 5° 19' 44.0322'' E) and Fresnes-sur-Escaut (France, Hauts-de-France region, lat. 50°
109 25' 4'' N, 3° 35' '' E). Briefly, the Tavaux site (S1)(Fig.2-S1) investigated in the present study
110 was exploited from the 1950s to 2003 as a storage area for sediments originated from effluents
111 produced during the electrolytic processes associated with a Hg cell chlor-alkali activity. The
112 Fresnes-sur-Escaut (S2)(Fig.2-S2) site was a deposit site of sediments from the canal Escaut. Due
113 to transport of metal ores in the past, sediments were contaminated by TE such as Cd, Zn, As and
114 Pb. Soils at both sites differed in their composition. The full description of the Tavaux and
115 Fresnes-sur-Escaut sites and soils are provided in (Durand et al., 2017) and (Ciadamidaro et al.,
116 2017), respectively. The main meteorological parameters during the growth period at the two sites

117 are summarized in table 1. At both sites, nettle grew spontaneously under the woody cover (Fig.
118 1).

119 Two other uncontaminated site (control) were used as reference to compare contamination values.
120 The control site in France is a natural area located at Courcelle-les-Montbéliard, 140 km east of
121 the site S1, not directly influenced by the point-source. (Yung et al., 2019) gives a description of
122 this site. The other control site is located in Belgium at Ghent region at 120 km north-east of the
123 site S2. A description of this site is given in (Tack and Verloo, 1996).

124 2.2 Experimental design

125 Within the BIOFILTREE project (Ciadamidaro et al., 2017), four type of field plots were set-up
126 in March (S2) and April (S1) 2011. The four type of plots consisted in i) poplar (Skado clone)
127 monocultures, ii) poplar (I214 clone) monocultures, iii) Skado-alder associations, iv) I214-alder
128 associations, with or without inoculation. Only the poplar monocultures have been considered in
129 the present study. On the two sites, non rooted poplar cuttings (1.5 m length) of the clones Skado
130 (*P. trichocarpa x P. maximowiczii* section Tacahamaca) and I214 (*P. deltoides x P. nigra* section
131 Aigeiros) were used, with a final tree density of 2.200 plants.ha⁻¹, each plot therefore having a
132 surface of 28 x 18 m for S1 and 36 x 12 m for S2. Both clones were selected for their growth yield
133 and low leaf TE accumulation capacities, from the previous clonal field trial PHYTOPOP (Pottier
134 et al., 2015). Figure 2 represents the implantation of the two described sites. Full description of
135 the sites is provided in (Phanthavongsa et al., 2017).

136 2.3 Nettle harvesting

137 During the first year (Y1), half of each plot area (approximately 250 m²) was harvested for three
138 selected plots (T.I1, T.I2 and T.I3) on S1 (Fig. 2, Fig.3a, b and c) on the 28th of September 2017.
139 The second year (Y2), an area of 50 m² was harvested in the plot TS of S1 on the 8th of June 2018,
140 and an area of 156 m² in the plots FS, P4, P5 and P30 on S2 on the 18th of July 2018. The
141 harvesting strategy was mainly driven by observations of the nettle development as well as the
142 competing species. At each harvest, stems were cut at soil level using scythes and shears. The
143 aerial parts of the cut plants were then immediately picked up by hand and baled, without prior

144 **field retting**. The stems were kept aligned in the **bundles** (Fig. 3d). The **bundles** were picked up
145 the same day and stored in indoor spaces for a minimal period of **3 months**.

146 2.4 Fresh and dry matter yields

147 The nettle bales were weighed in the field just after harvesting to determine the mass of fresh
148 plant materials (M_{fresh}), and then weighed again after air-drying (at least 4 months of conditioning
149 in indoor spaces) to determine the mass of dried plant materials (M_{dry}). The leaves were then
150 manually picked off. The remaining stems were further weighed to determine the mass of dried
151 stems (M_{stem}). The yields were then estimated by calculating the ratio between the masses and the
152 plot areas. The yields in fresh matter, dry matter and dry stems are noted η_{fresh} , η_{dry} , η_{stem} ,
153 respectively.

154 2.5 Stem length and diameter

155 Stems were analysed in terms of dimensions by selecting randomly 30 stems in each harvested
156 batch. Stem diameters were measured for each stem in three positions along the stem height
157 (top, middle and base parts of the stem) using a calliper with a precision to 0.01 mm. For each
158 position, two measurements were done according to the two main directions of the transversal
159 cross-section area. The average diameter for this position was then calculated from these two
160 measurements. The mean diameters measured in these respective positions are noted D_{top} ,
161 D_{mid} , D_{low} . The length (L) was measured using a graduated steel rule with a precision of 0.1
162 cm.

163 2.6 Stalks processing and fibre extraction.

164 The stems of the Y2-S1 batch were mechanical processed using a Laroche (France) Cadette 1000
165 “all fibre” extraction device. This machine is a three-drum opener with a 1000 mm wide feed belt
166 (Fig. 4a). At the entrance of each of the three modules, the feeding of the fibrous material was
167 provided by a pair of rolls, one smooth and the other grooved (made of rubber) (Fig. 4b). Then,
168 each module was equipped with a cylinder with nails, *i.e.* the extracting roller (or fibre extraction
169 roller) (Fig. 4c). Its distance from the metal trough located at the module inlet is adjustable
170 (distance A in Fig. 4b), as well as its rotational speed (from 750 to 1800 rpm). Under the extracting

171 roller, a trap door allowed the evacuation of shives by gravity. The trap door opening is also
172 adjustable and, for this study, it was set at its maximum to favour the shives removal. At the end
173 of the module, there was also a perforated cylinder at which ventilation was applied. The latter
174 had three distinct functions: (i) the de-dusting of the material, (ii) the formation of the fibre lap,
175 and (iii) its transfer to the next module or the outlet. Each de-dusting fan was equipped with a
176 motor having a 2865 rpm maximum rotation speed.

177 During the experiments, the operating parameters used were inspired by two previous studies
178 dealing with the mechanical extraction of technical fibres from oleaginous flax (Ouagne et al.,
179 2017) and hemp (Grégoire et al., 2019) straws, respectively. From modules 1 to 3, distance A
180 were 1.9 mm, 1.4 mm and 1.1 mm, respectively. The inlet flow rates of nettle stems were
181 approximately 125 kg/h, corresponding to a 2.5 m/min feed belt speed. The transmission speed
182 of the lap from module 1 to module 2 was 2.2 m/min, and it was 2.1 m/min from module 2 to
183 module 3. Finally, a 1.7 m/min speed was used for the output belt. In each of the three modules,
184 a 750 rpm rotation speed was chosen for the extracting roller. Looking at the de-dusting fans, the
185 rotation speed of the motor was 1500 rpm for module 1, and 2000 rpm for the next two modules.

186 **Since the nettle straws were not field retted, three different conditionings before processing were**
187 **tested to evaluate their influence on the nettle fibre yield and quality. Prior to these treatments,**
188 **the straws were stored during 3 months.**

189 - Condition 1 (C1): as-received. **The nettle** straw was processed as-received after the 3
190 months of storage. Its average moisture content was approximately 10.4%.

191 - Condition 2 (C2): moistened. **The nettle** straw was sprayed with water to re-moisturise
192 the matter **(during 15 hours)**. Its average moisture content was thus approximately 22.0%.

193 - Condition 3 (C3): soda treatment. **The nettle** straw was immersed for 2 h and at 60 °C in
194 a soda solution (2%) and then thoroughly rinsed with water until reaching a pH value
195 **comprised between 7 and 8. The nettle straw was then oven-dried inside a ventilated oven**
196 **at 50 °C and during 16 h.** The moisture content before mechanical processing was 16.5%.

197 After the mechanical processing step, the fractions of the resulting matters were determined.
198 These matters were divided into three categories: a fibre lap containing the extracted bast fibres,
199 shives and dust. The content of impurities inside the fibre lap (*i.e.* remaining shives and dust) was
200 also evaluated. Indeed, the lap collected at the outlet of the “all fibre” extraction device consisted
201 not only in bast fibres but also in some remaining shives and dust, both trapped inside the lap.
202 From each of the three generated laps, a 50 g sample was collected to determine the content of
203 impurities inside the lap, and thus the real bast fibre content. A mechanical sieving was performed
204 on the lap sample for 5 min in order to drop shives and dust. Then, the residual shives that
205 remained trapped were collected manually.

206 Moisture content of the fibrous materials at the inlet of the “all fibre” extraction device and out
207 of the process was determined according to standard ISO 665:2000.

208

209 2.7 Fibre diameter

210 **In order to characterize the morphology of the bast fibres and determine their diameters, fresh**
211 transverse cross-sections were cut from the stems using razor blades at different locations and
212 different development and maturity stages. Cytochemical staining of lignin was achieved on
213 sections using the Wiesner reagent (phloroglucinol-HCl) that colored lignin into purple-red and
214 assist in the definition of the fibre wall outlines. The observations were made on a microscope
215 Keyence VHX 5000 series with a magnification of 20.

216 2.8 Single fibre tensile properties

217 Single fibre tensile tests were performed on **nettle** fibres isolated by hand from the batch Y2-TS
218 and Y2-FS, and from the mechanically processed batch Y2-TS. The fibres were first glued on a
219 paper frame and then examined using an optical microscope to determine their external width.
220 The average width of each fibre was computed by obtaining ten measurements equally spaced
221 along its length. A Bose Electroforce 3230 machine was used to perform the monotonic tensile
222 tests up to failure. The paper frame supporting each fibre was clamped onto the testing machine

223 and then cut prior to the beginning of each test. The clamping length was 10 mm. Fibres were
224 tested at a constant crosshead displacement rate of 0.01 mm.s⁻¹. The applied force was measured
225 with a load sensor of 22 N with a resolution of approximately 10 mN, and the displacement was
226 measured using an LVDT with a resolution of approximately 0.5 µm. The sample elongation
227 and the load were recorded continuously. The strain was calculated as the elongation divided by
228 the initial fibre length, taking into account the compliance of the loading frame.

229 The effective cross-section was determined using the mean external width, assuming
230 that the fibre was perfectly cylindrical and the lumen area neglected. The stress was calculated
231 using the applied force and the evaluated initial cross-section mean value of the fibre. The
232 variation of the cross-section during the tensile loading was neglected. The tensile strength (σ_R)
233 and the strain at failure (ϵ_R) were determined, and the apparent tangent modulus of rigidity (E)
234 was computed on a strain range comprised between 0.2% and the strain at failure. During the
235 fibre preparation, and using only optical microscopy, it is sometimes difficult to discriminate
236 single fibres against small bundle of fibres. As a result, some small bundles of nettle fibres were
237 also tested. This can be easily detected on the shape of the resulting stress-strain curve. In a
238 strain-controlled situation, the force drops abruptly when an individual fibre breaks. Thus, a
239 criterion was introduced to identify during the data processing fibre bundles. This one is based
240 on the detection of sudden stress drop (set at 5 MPa) between two successive measure points.

241 2.9 Determination of TE content

242 Before analyses, dried nettle stems and leaves from S1 and S2 and non contaminated soil (control)
243 were separated, air-dried and ground into a homogenous powder in a Mixer Mill, for 4 min at 30
244 Hz and 7 min at 30 Hz for leaves and stems, respectively. We also isolated nettle fibres for Hg
245 analysis. Cd, Cu, Hg, Pb and Zn concentrations were measured in the soil and different parts of
246 nettles. Soil samples were ice-dried and sieved (0.2 mm particle size). The total element
247 concentrations (Al, Cd, Fe, Mn, Pb, S, Zn) in the soil from S1, S2 and control were measured
248 following the acid digestion of 500 mg of sample (DigiPREP system, SCP Sciences, Courtaboeuf,
249 France) using a mix of 2 mL of 67% nitric acid, 6 mL of 34% hydrochloric acid and 2 mL of 48%

250 hydrofluoric acid (Ciadamidaro et al., 2017). CaCl₂-extractable fractions of TE were determined
251 in soil samples after shaking 2.5g of dried soil in 25 ml of a 0.01 M solution of CaCl₂ during 3 hr
252 (140 g / min⁻¹) (Houba et al., 1996). The soil mixtures were filtered on filtration units at 0.45 μm
253 (Millipore). Eluates were acidified to a pH < 2 for preservation. TE in plant and soil samples were
254 analysed using inductively coupled plasma atomic emission spectrometry (ICP-AES, Thermo
255 Fischer Scientific, Inc., Pittsburgh, USA). Hg was measured using the atomic absorption
256 spectrophotometry AMA-254 cold vapor atomic absorption (CV-AAS) Hg analyser (Altec Co.,
257 Czech Republic), using standard conditions (45 s drying, 150 sheating, 45 s cooling) and the
258 certified reference material (CRM), i.e. Oriental Basma Tobacco leaves (INCT-OBTL-5). All the
259 results are expressed as μg/kg of the dry weight (DW).

260

261 2.10 Statistical analysis

262 The dimensional and mechanical data presented in this paper are the mean values, standard
263 deviations and the parameters of a log-normal distribution law identified by inverse method.

264 The best distribution function for each property was identified using the EasyFit software by
265 minimising the criterion of distances from the distribution diagram. The distribution functions
266 were selected among the classical ones. The log-normal distribution ($LogN(\mu, \lambda)$)(Eq. 1) was
267 found to be more realistic and is preferred to the other distributions. This distribution is a two-
268 parameter family of continuous probability distributions. μ and λ are the expectation and standard
269 deviation of x and $\ln(x)$, respectively.

$$270 \quad - \quad LogN(\mu, \lambda) : f(x) = \frac{1}{x} \frac{1}{\lambda \sqrt{2\pi}} e^{-\frac{1}{2} \left(\frac{\ln(x) - \mu}{\lambda} \right)^2} \quad (1)$$

271

272 3 Results and discussion

273 3.1 Trace element content of soils and harvested biomasses

274 The Fresnes-sur-Escaut soil was highly contaminated by Cd, Pb and Zn, and Cu to a lesser extent,
275 which confirms our previous studies (Ciadamidaro et al., 2017; Phanthavongsa et al., 2017).
276 However the nature of the soil rendered these TE poorly available, with CaCl₂ extractable fraction
277 represented 0.2, 0.4, 0.02 and 0.04% of the total fraction for Cd, Cu, Pb and Zn, respectively
278 (Table 2). Cu, Zn and Pb contents in nettle collected at the Fresnes-sur-Escaut site ranged within
279 physiological levels (Kabata-Pendias, 2011; Kalra et al., 1998), and did not significantly differed
280 between tissues (Table 2). Cd contents in the various nettle tissues were within the ranges found
281 in plants grown in agricultural soils, and always much lower than Cd concentrations measured in
282 poplar or salix leaves (from 2 to 14 mg/kg DW) collected at the same site (Phanthavongsa et al.,
283 2017). Additionally, the TE concentrations in nettle found in our study were within the ranges of
284 concentrations measured in nettles collected at uncontaminated sites in Belgium **near the location**
285 **of the site S2**. (Tack and Verloo, 1996). These TE concentrations were also well below the critical
286 levels set up for plant tissues (Kabata-Pendias, 2011), at which growth depression may be
287 observed, i.e. 10-20 and 100-500 mg/kg DW for Cd and Zn, respectively.

288 The Tavaux soil was contaminated by Hg, with concentrations around 6 mg/kg DW, as
289 previously found (Durand et al., 2017; Maillard et al., 2016). At the Tavaux site, translocation of
290 Hg to nettle tissues remained low, as Hg contents in leaves did not exceed 0.01 mg/kg DW (Table
291 2). These concentrations were comparable to those measured at our French control site (mean
292 0.006 mg/kg DW, Table 2). Hg content of **nettle** fibres was below the limit of quantification. The
293 Hg concentrations in nettle leaves were far below the threshold set to 0.2 mg/kg for concentrations
294 tolerable in agronomic crops and within the ranges measured in food plants (0.006-0.139 mg/kg
295 DW) in various countries (Kabata-Pendias, 2011). The Hg content of food plants grown in chlor-
296 alkali areas were in the range 0.032-0.59 mg/kg DW (Kabata-Pendias, 2011). These authors also
297 set the toxic Hg content in crop leaves within the range 1-3 mg/kg DW.

298 Overall, our dataset confirmed the limited translocation of TE in nettle biomass, as
299 compared to woody species grown on the same plots, which indicate that fibres from nettle grown
300 at these sites may be used without restriction in industrial processes.

301 3.2 Fresh biomass and straw yields

302 Nettle bales were weighed just after harvesting to determine the yields of fresh matter (Table 3).
303 During the first year, only the experimental site of Tavaux (S1) was harvested at the end of
304 summer. Relatively low yields were measured on each plot and were comprised between 0.293
305 and 0.817 t.ha⁻¹ for fresh matter, and between 0.028 and 0.091 t.ha⁻¹ for dry stems. This disparity
306 between each plot and particularly for I3 was due to the presence of plant species (*Galium aparine*
307 *L.*, *Sambucus ebulus L.*, *Calystegia sepium L.*) which are able to compete with nettle (Strutek and
308 M, 1998), and also to differences in the quantity of incident light (Yung L et al.). These yields
309 values were lower than those mentioned in the literature (Table 4, (Bacci et al., 2009; Bredemann
310 and Garber, 1959; Dreyer et al., 1996; Francken-Welz et al.; Köhler et al., 1999; Lehne et al.,
311 2001; Meirheage, Mars 2011; REXEN, 2002; Schmidtke et al., 1998; Vetter et al., 1996) which
312 were in the range 1 - 11.5 t.ha⁻¹ for dry stem in the context of traditional agronomic practices. The
313 second year, at the beginning of the summer, two plots (TS and FS) located at the edge of the
314 poplar coppices were harvested in Tavaux (S1) and Fresnes-sur-Escaut (S2) respectively. Yields
315 in fresh matter were 4 to 10 times higher than those measured under the poplar coppices (Table
316 3). A new weighing was made, after 4 months of air-drying. The dry biomass yield reached a
317 value of approximately 2 t.ha⁻¹. The dry stem yields were also measured after leaf removal. The
318 obtained values varied between 0.55 and 1.38 t.ha⁻¹, as a function of the harvesting site.

319 These yields were comparable to the lower values reported in the literature in the context
320 of traditional agronomic practices. Indeed, literature (Bacci et al., 2009; Dreyer et al., 1996; Francken-
321 Welz et al.; Hartl and Vogl, 2009) reported dry matter yields comprised between 2.3 to 15.4 t.ha⁻¹
322 depending on the fertilization conditions. The yields obtained in our site conditions were
323 nevertheless extremely interesting since, in this agro-forestry system, nettle grows naturally and

324 spontaneously, without chemical inputs. The savings associated to the lack of planting / sowing,
325 crop maintenance and inputs may motivate harvesting lower yielding plots.

326 The fraction of leaves consisted in approximately 35 to 60% of the dry biomass,
327 depending of the year and localisation of harvest. reported leaf fractions around 7%, which
328 emphasizes the differences that can be obtained between the spontaneous nettle and cultivars.

329 Figure 5 represents the statistical distributions of the stem features for each harvested
330 plots. The features measured for the Tavaux (Fig.5a) and Fresnes-sur-Escaut (Fig.5b) sites show
331 that the length or the diameters of the stems were higher for nettle growing at the edge of the
332 poplar coppice (Y2-S1-TS and Y2-S2-FS) than under the coppice. The mean length was
333 approximately 1.14 m at the edge of the coppice and 0.87 m under poplars. These values of native
334 nettle length were 33 to 50% lower than those measured by (Bacci et al., 2009) (1.7m) for a
335 cultivar using traditional practices.

336 We further estimated the diameter of the nettle fibres for the plots Y2-S1-TS and Y2-S2-
337 F, with average values of 37 ± 11 and 34 ± 12 μm , respectively. These values are in agreement
338 with the mean diameters measured by others authors (Di Virgilio, 2013) for nettle cultivars (20-
339 $47\mu\text{m}$). On the other hand, the thickness of the fibre wall was very different between the two
340 batches. The batch Y2-S2-FS presented an average wall thickness of 6.5 ± 2 , as compared to a
341 thickness of 12.5 ± 3 μm for the Y2-S1-TS batch. Figure 6 shows transverse cross-sections of
342 nettle stems harvested in the second year that are representative of each site, S1 (Fig. 6a) and S2
343 (Fig. 6b), respectively. It can be observed that the nettle bast fibres presented a quite large lumen
344 area for the two batches, when compared to other European crops such as hemp and flax. For the
345 spontaneous nettle, the harvesting time was not selected as a function of a specific maturity stage.
346 However, it is well known for other crop fibres that the harvest time strongly influences the fibre
347 yield and the fibre filling level (Goudenhooff et al., 2018; Musio et al., 2018). So, the influence
348 of the harvest time of the nettle fibre features needs to be investigated in further studies.

349 3.3 Continuous fibre extraction using the “all fibre” extraction device

350 From the standard settings **described in section 2.5**, the “all fibre” extraction device was able to
351 extract the textile fibres contained in the nettle stalks, leading to the separate production of a fibre-
352 enriched mat (*i.e.* the fibre lap), shives and dust (Figure 7). Shives were conveyed separately via
353 the waste evacuation belt, and dust was removed thanks to the de-dusting fans. The three
354 extracting conditions (C1, C2 and C3) showed significant differences in the quality of the **nettle**
355 fibres obtained in the outgoing mat. Visually, the C1 condition, corresponding to the use of the
356 as-received nettle stalks, resulted in the cleanest and the finest fibres obtained, with extracted bast
357 fibres up to 10 cm in length.

358 The material balance of the three trials conducted is mentioned in Table 5. Firstly, it
359 systematically showed a slight decrease in the moisture content of **nettle** fibres from the inlet to
360 the outlet, which was very likely explained by the use of the de-dusting fans. Moreover, these
361 fans allowed a suitable elimination of dust generated during the fibre extraction (2.2-5.0% of the
362 mass of nettle stalks processed), in priority at the level of module 1 and, to a lesser extent, at the
363 level of the two following modules. Depending on the extracting condition, the fibre lap collected
364 at the outlet of the Cadette machine represented between 11 and 48% of the incoming mass. The
365 C2 samples (*i.e.* straw re-moistened before extraction) had the greatest proportion of fibre lap.

366 However, this mass proportion must be corrected by the content of impurities inside the
367 fibre lap (Table 5). The latter was estimated inside the three laps produced, impurities
368 corresponding to (i) shives trapped inside the mat, (ii) dust not properly removed by aspiration,
369 and also (iii) the few fibres not yet separated from shives. The content of impurities was minimal
370 (18%) for the C1 condition (control sample), whereas it doubled for the C2 condition (re-
371 moistened stalk) and reached a particularly high value for the C3 one (stalk pre-treated with soda
372 before mechanical extraction).

373 In conclusion, the as-received nettle straw (*i.e.* control sample, C1) undoubtedly appeared
374 as the most promising raw material, combining (i) the lower impurity (shives) content inside the
375 fibre lap (18% in weight) and (ii) the finest extracted fibre bundles, some of them reaching about

376 10 cm in length. For this sample, the bast fibres contained in the outgoing mat represented 9.1%
377 of the initial mass of nettle stalks introduced. Before any further textile step (carding), it would
378 still be necessary to clean this mat to remove impurities (shives, dust) trapped between the
379 extracted fibres. For future experiments, such operations could be performed using an additional
380 sieving step for the fibre mat. This nettle fibre yield obtained for the Y2-S1-TS batch is in
381 accordance with the literature, which reports nettle fibre yields between 3 and 16% (Table 4).

382 3.4 Mechanical properties of fibres

383 Figure 8 shows the stress/strain curves obtained from single fibre tensile tests, for three different
384 batches that comprised from 32 to 50 fibres. On Figure 8a, results concern nettle fibres from the
385 Fresnes-sur-Escout site, manually extracted. Figures 8b and 8c present the results obtained for
386 the nettle fibres obtained on the Tavaux site after manual extraction and after mechanical
387 extraction, respectively. The mean value of the stress at failure for each batch is represented by a
388 dotted line. A large scattering in the response was observed, which is typical of plant fibres
389 (Bourmaud et al., 2018). A significant non-linearity could be observed for most of the tested nettle
390 fibres, which contrasts with the results of Bodros and Baley (Bodros and Baley, 2008) who
391 reported a linear response for nettle fibres manually extracted from retted stems. Table 6
392 synthesises the mean tensile properties for each batch.

393 *Influence of the harvesting site*

394 The average properties, presented in Table 6 (tensile strength, strain at failure and apparent
395 rigidity), of the nettle fibres extracted manually are equal to 711 MPa, 1.37%, 53 GPa and 812
396 MPa, 2.14%, 36 GPa for Y2-S2-FS and Y2-S1-TS respectively. These values are lower than those
397 reported in the literature for native nettle cultivars (Bodros and Baley, 2008; Lanzilao et al., 2016).
398 These differences can be due to several factors such as the harvesting time or related to the fibre
399 preparation. If the strength was quite similar for the two tested batches, a significant difference
400 could be observed for the strain at failure and the apparent rigidity. Y2-S1-TS nettle fibres had a
401 mean strain at failure 1.5 time greater than the Y2-S2-FS nettle fibres and an apparent rigidity 1.5
402 time lower.

403 This difference in apparent rigidity can be attributed to the difference in the average fibre
404 wall. It was above-mentioned that the wall thickness of the Y2-S2-FS nettle fibres was twice
405 higher than for Y2-S1-TS. The cross-section area (CSA) of the fibres was calculated considering
406 the fibre cylindrical and by neglecting the lumen area. This clearly led to a significant over-
407 estimation of the CSA and then an under-estimation of the apparent rigidity and strength. This
408 under-estimation was more important when the lumen area was large. To ensure a more reliable
409 comparison, additional tests taking into account the real CSA of the nettle fibres are necessary.

410 *Influence of the fibre extraction method*

411 The impact of the extraction mode (mechanical or manual) was also evaluated on the Y2-S1-TS
412 batch. An analysis of variance (ANOVA) was realised on the different values. While the impact
413 on the stress at failure was not statistically representative (p-Value=0.258), a strong influence was
414 observed on the apparent rigidity and on the strain at failure. Interestingly, we can observe a
415 significant difference also in the diameters of the tested nettle fibres, with means of 35 and 28
416 μm , measured for the tested nettle fibres obtained after manual extraction and after mechanical
417 extraction, respectively. This difference could be attributed to the better fibre separation achieved
418 with the mechanical extraction. The fibre individualisation is a particularly difficult step when
419 made by hand, which is confirmed during the post-treatment of the tensile tests. Using the
420 introduced criterion, it is observed that the bundles of nettle fibres represented approximately 6%
421 of the tested fibres after mechanical extraction against 30% for the manual extraction. For bundle
422 of fibres, a lower rigidity and strength were generally measured (Placet et al., 2017). This was
423 due to the weakness of layer between the individual fibres. So, based on this reasoning, it could
424 be expected to measure a higher stress at failure for the mechanically processed batch. This
425 potential positive effect induced by the improvement of the fibre individualization can also be
426 counterbalanced by the fact that mechanical extraction also generally induces damages and thus
427 a lowering of the properties at failure (Ouagne et al., 2017) .

428 Finally, the properties obtained for nettle after mechanical processing are promising when
429 compared to the best results obtained for the other European fibre crops, such as flax (see Table

430 6). (Bensadoun et al., 2017) reported 57 GPa and 791 MPa for the tensile rigidity and strength of
431 industrial flax fibres, respectively, when determined using single fibre tensile test. It is also
432 important to emphasize that for this spontaneous nettle fibres, the harvest time was not optimized
433 and, thus, an improvement of the tensile properties can be expected by adjusting the harvest time
434 to the fibre maturity. In this work, we only considered the apparent properties, which are clearly
435 under-estimated when considering the large lumen area of the tested fibres. By taking into account
436 the real CSA, the effective properties of the fibres could be determined.

437 **4 Conclusions**

438 This work focused on the sustainability of spontaneously stinging nettle grown on trace-
439 element contaminated soil. Our results demonstrate that the use of trace-element contaminated
440 soils is a relevant option to expand the material purpose-grown biomass and at the same time
441 mitigate the land-use conflict between the needs of food and the increasing demand for plant
442 based fibre raw materials. **Trace-element contents in nettle fibres are far below the tolerable**
443 **threshold for agronomic crops.** Tensile properties obtained after an extraction made with an “all
444 fibre” industrial extraction device and without field retting or prior alkaline pre-treatment are
445 comparable to the properties of the best industrial flax fibres.

446 Our set of results is promising in view of a material use. Indeed, the savings associated to
447 the lack of planting/sowing, crop maintenance and inputs may motivate harvesting lower yielding
448 plots. The question of the mechanization of the harvesting, collecting and transportation and thus
449 of the economic balance are however still opened, **as well as the generalization of the results to**
450 **other phytomanagement sites and configurations.**

451

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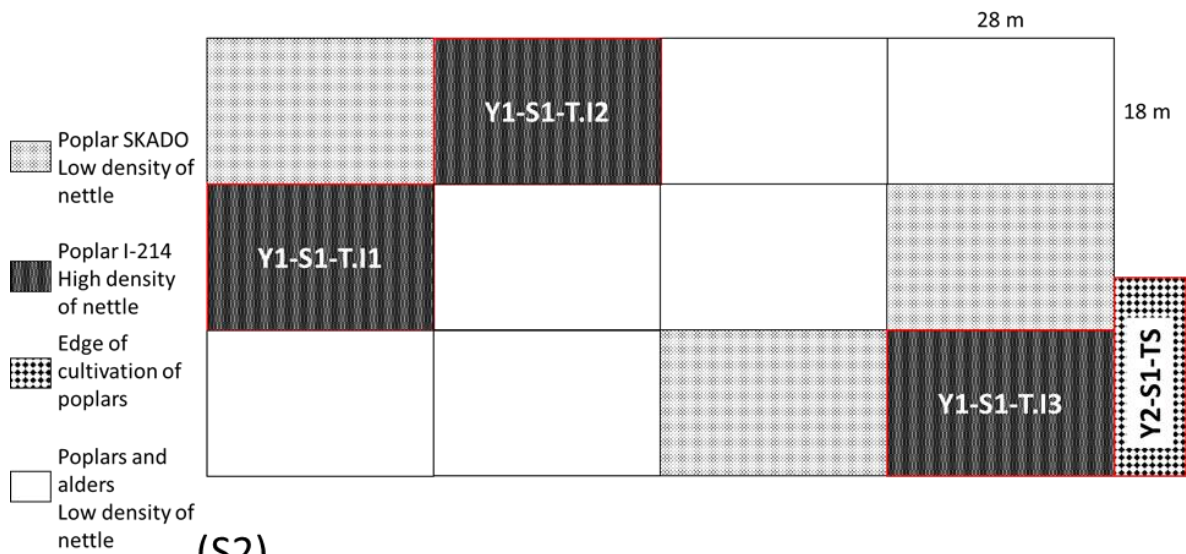
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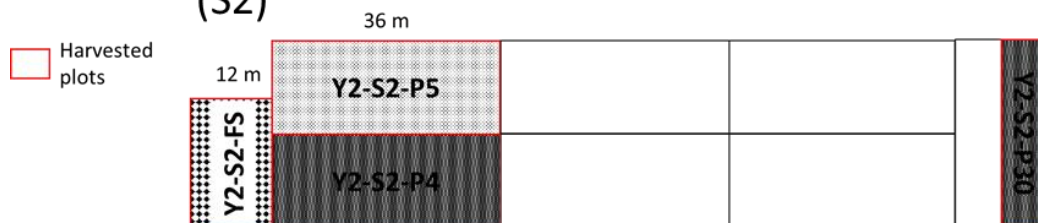
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582 **Figure 1: Photograph of stinging nettle growing spontaneously and in a prevalent manner under short rotation**
 583 **coppice for phytomanagement of trace elements contaminated soils.**

(S1)

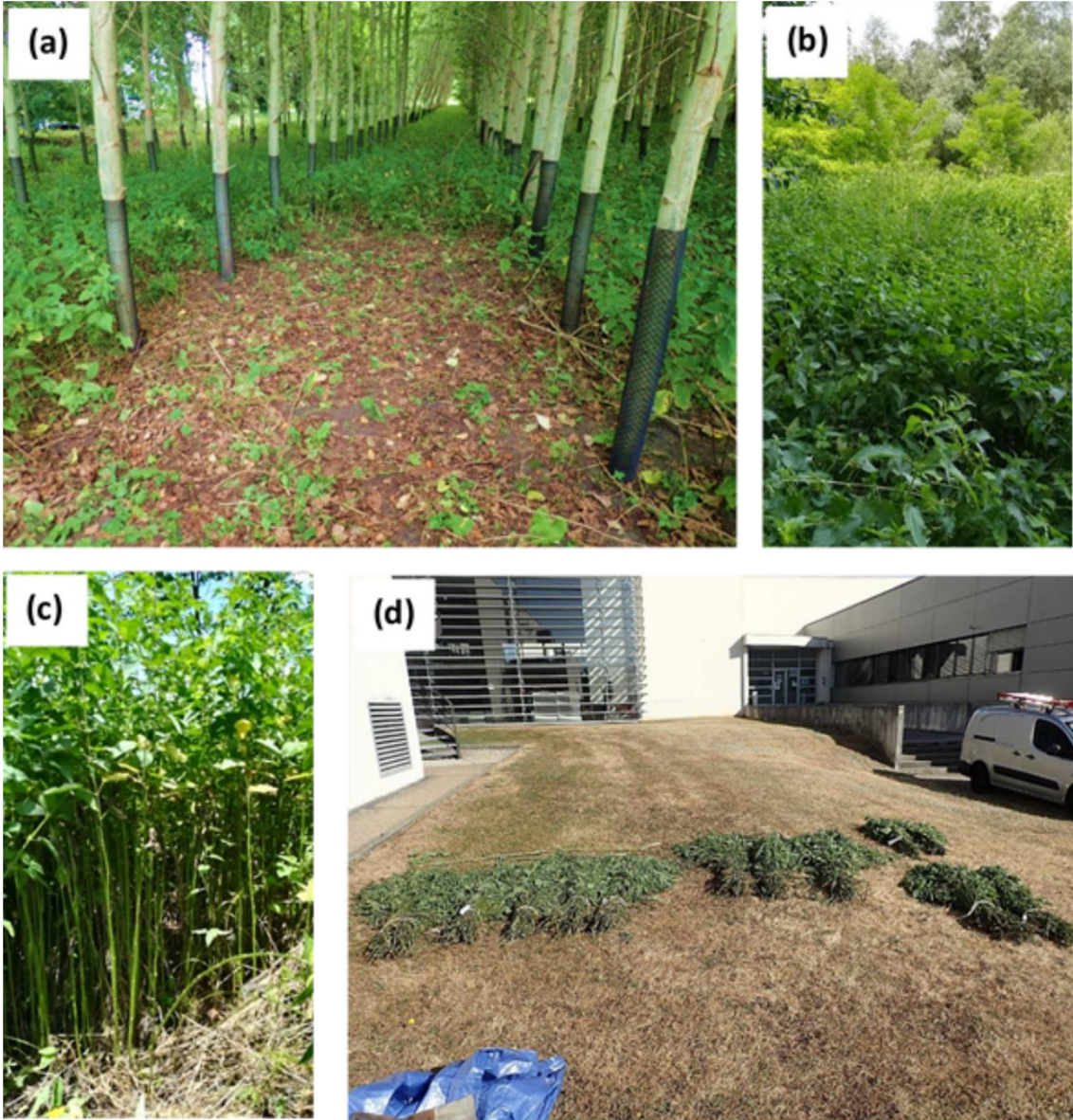


(S2)



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585 **Figure 2: Experimental designs at the two phytomanaged field sites, Tavaux (S1) and Fresnes-sur-Escaut (S2).**
 586 **The sampled plots are in dark grey (I214 plots) and light grey (Skado), on which nettles were collected. Full**
 587 **description of the sites is provided in (Phanthavongsa et al., 2017). Nettles were also collected in the edge of the**
 588 **poplar plots.**

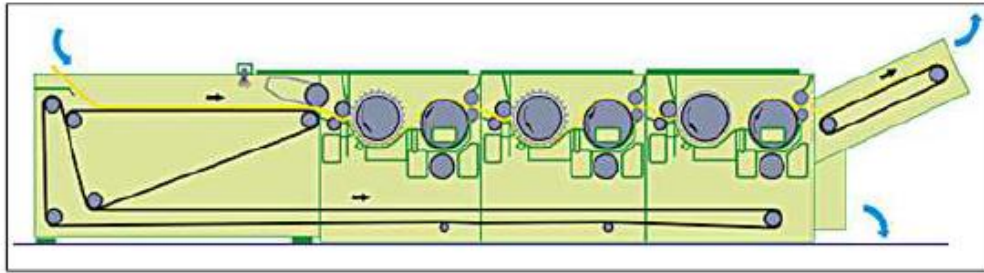


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590 **Figure 2: Photograph the plots and the crops. (a) Plot No. Y1-S1-T.I3 after harvest, (b ,c) Nettles in the plot**
591 **No. Y1-S2-TS, at the wood-edge, and (d) Crop of Y2-S2-(FS, P4, P5, P30).**

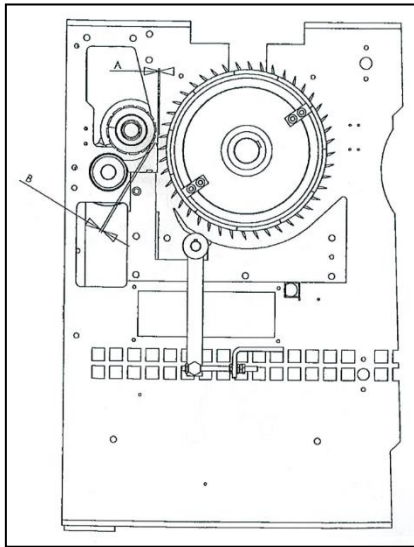
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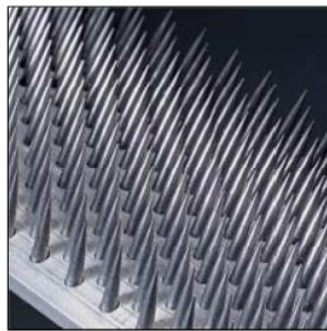
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(a)



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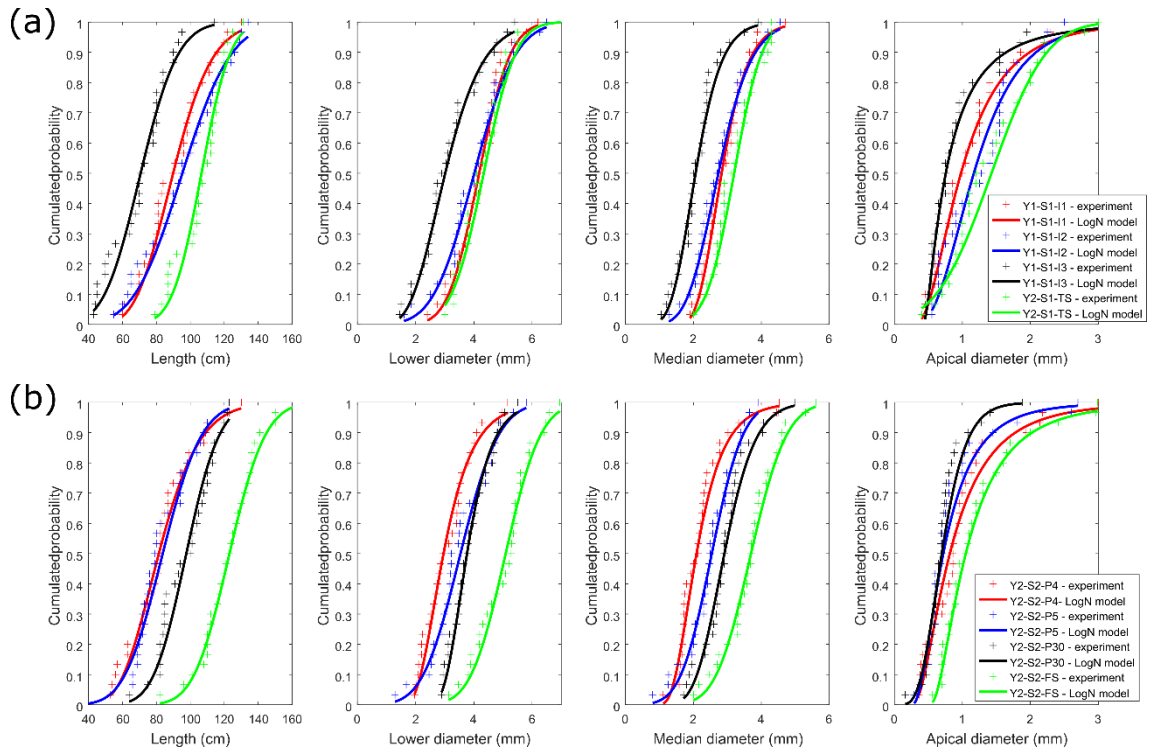
(b)



(c)

596 **Figure 3: Laroche (France) Cadette 1000 “all fibre” extraction device used in this study (from Laroche**
 597 **company website). (a) Material path along the opener (in yellow on the drawing), from the feed belt (left) to**
 598 **the exit belt (right). (b) Detailed view (from left to right) of the pair of rolls needed for feeding (smooth roll**
 599 **down and grooved roll up), and the fibre extraction roller (cylinder equipped with nails), all three located at**
 600 **the module inlet. (c) Photograph of the nails of the extracting roller in module 1.**

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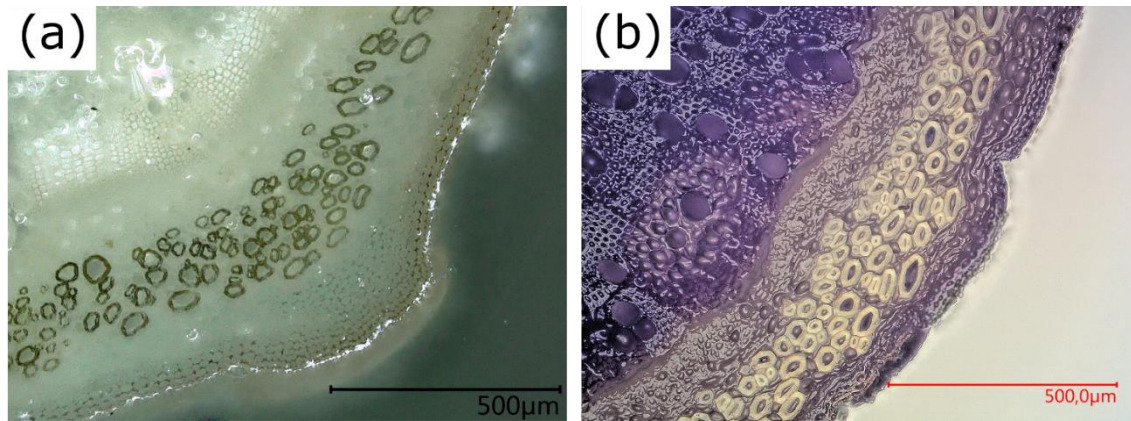
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603 **Figure 4: Statistical distribution of the nettle stem features (length and diameter) as a function of the location**
 604 **in the plant and of the harvesting site: S1 (a) and S2 (b).**

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609 **Figure 5: Micrograph of the transverse cross-section of nettle stems. Nettle stems were harvested in second**
 610 **year on S1 (a), and S2 (b) site, respectively.**

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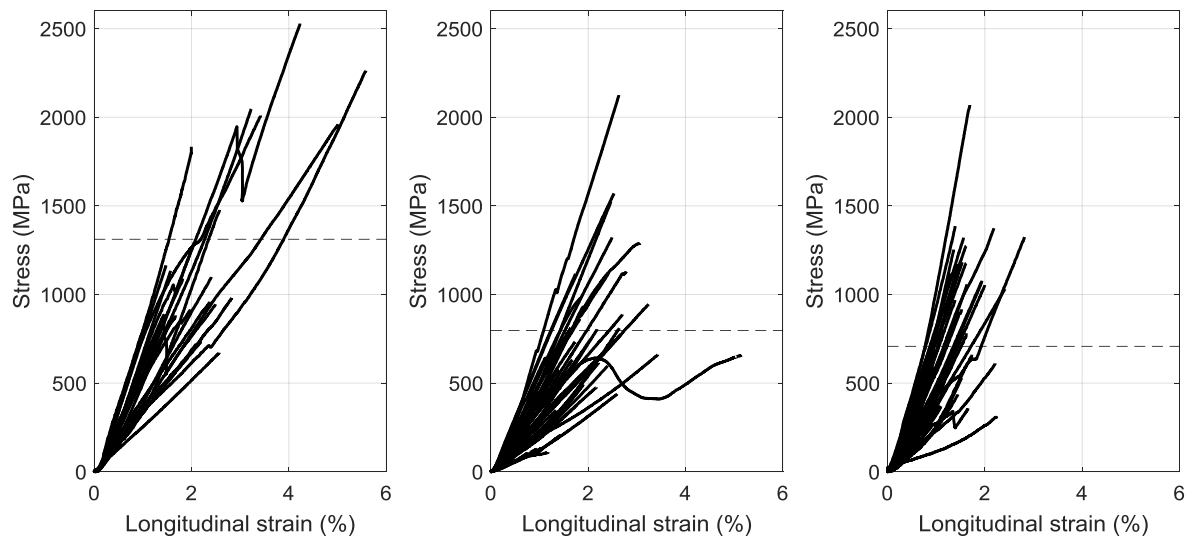


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616 **Figure 6: Pictures of the nettle matters after mechanical processing.**

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620 **Figure 7: Tensile stress-strain curves of nettle fibres from the batches Y2-S2-FS manually extracted (a), Y2-**
621 **S1-TS with manual extraction (b) and Y2-S1-TS with mechanical extraction (c).**

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Table 1: Main meteorological conditions during the growth period at the two sites.

	S1-Y1	S1-Y2	S2-Y1	S2-Y2
Annual average temperature (°C)	11.9	13.1	10.8	11.3
Total annual sunshine (h)	2068	2122	1290	1699
Total annual rainfall (mm)	758	718	543	632

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Table 2: Trace element concentrations (mg/kg DW) in soil and nettle samples from the two sediment landfills. For comparison, the TE background concentrations, from natural sites in Belgium were obtained from Tack and Verloo (1996) for Cd, Cu, Pb and Zn, and from our control site located in the East of France for Hg.

Trace element (mg/kg DW)		Cd	Cu	Pb	Zn	Hg
Industrial sediment landfills		Fresnes sur Escaut				Tavaux
Soil	total	15 ± 19	79 ± 33	541 ± 1397	1506 ± 856	6 ± 1
	(pH = 6.6) CaCl ₂ ext.	0.03 ± 0.01	0.33 ± 0.06	0.13 ± 0.57	0.71 ± 0.003	NA
Nettle	stems	1.0 ± 0.7	3.3 ± 0.6	1.9 ± 0.4	24.0 ± 6.3	0.01 ± 0.004
Nettle	fibres	1.1 ± 0.4	2.9 ± 1.0	1.3 ± 0.5	47.5 ± 5.0	NA
Nettle	leaves	0.9 ± 0.8	9.4 ± 0.53	2.0 ± 0.2	31.9 ± 1.8	0.03 ± 0.01
Control sites		Belgium				France
Soil	aqua regia ext.	0.5 ± 0.4	12 ± 5	28 ± 17	54 ± 13	0.6 ± 0.1
Nettle	leaves & stems	0.4 ± 0.2	14 ± 5	34 ± 19	113 ± 82	0.006 ± 0.003

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633 **Table 3: Nettle stem and bast fibre yields (t.ha⁻¹) determined at the two contaminated sites.**

	Fresh biomass yield	Dry biomass yield	Dry stem yield
Y1-S1-T-I1	0.73	0.15	0.059
Y1-S1-T-I2	0.817	0.154	0.091
Y1-S1-T-I3	0.293	0.059	0.028
Y2-S1-TS	3.5	-	0.55
Y2-S2-P30	2.95	0.91	0.60
Y2-S2-P4	2.38	0.77	0.34
Y2-S2-P5	2.2	1.01	0.67
Y2-S2-FS	3.76	1.98	1.38

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Table 4: Stem and bast fibre yields reported in the literature for fibrescrops cultivated on agricultural lands

	Cultivar	Straw yield (t.ha⁻¹)	Bast fibre yield (t.ha⁻¹)	References
Nettle	/	3 – 4.1	/	(Bredemann and Garber, 1959)
	/	4.4 – 7.3	/	(Vetter et al., 1996)
	Clone 13	4.88 – 11.52	0.14 – 1.28 (2.9 – 11%)	(Dreyer et al., 1996)
	Clone 13	2.19 – 4.93	0.3 – 0.6 (13.6 – 12%)	(Schmidtke et al., 1998)
	/	2.66 -5.52	0.21 - 0.49 (7.9 – 8.9%)	(Köhler et al., 1999)
	/	6.71 - 8.12	1.09 - 1.22 (16.2 – 15%)	(Francken-Welz et al.)
	Clone 13	4.4	0.53 (12%)	(Lehne et al., 2001)
	Clone 5-9	2.3 - 9.7	0.3 - 1.02 (13 – 10.5%)	(Hartl and Vogl, 2009)
	/	1 - 10 (3.4 mean)	/	(Rexen, 2002)
	clone 13	15.4	1.696	(Bacci et al., 2009)
Hemp	/	7	2 – 2.2 (28.6 – 31.4%)	(Meirheage,
Flax	/	6.9	0.7 – 1.7 (10 – 24.6%)	Mars 2011)
Oleaginous flax	/	2	0.5 (25%)	

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Table 5: Material balance for the three trials conducted, including nettle bast fibre content and impurities level in nettle fibre lap, after mechanical processing

Sample	Control sample (C1)	Moistened (C2)	Soda treatment (C3)
Moisture content at the inlet (%)	10.4 ± 0.2	22.0 ± 0.5	16.5 ± 0.3
Moisture content out of the process (%)	9.6 ± 0.2	18.9 ± 0.3	10.3 ± 0.2
Fibre lap (%)	11.0	48.4	15.4
Shives (%)	86.8	46.6	82.3
Dust (%)	2.2	5.0	2.3
Content of impurities in fibre lap (%)	17.6	36.5	51.0
Real content of bast fibres inside the lap (%)	82.4	63.5	49.0

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645 **Table 6: Tensile properties of nettle bast fibres**

Mechanical properties of nettle fibres	Y2-S2-FS	Y2-S1-TS	Y2-S1-TS	(Bodros and Baley, 2008)	(Lanzilao et al., 2016)
Fibre extraction	Manual	Mechanical	Manual	Manual	Manual
Diameter (μm)	27.8 \pm 6.8	28.2 \pm 6	35.4 \pm 9	19.9 \pm 4	24.0 \pm 15.9
Stress at failure (MPa)	1314 \pm 552	711 \pm 427	812 \pm 451	1594 \pm 640	2196 \pm 801
Elastic modulus (GPa)	54 \pm 17	53 \pm 24	36 \pm 19	87 \pm 28	79 \pm 29
Strain at failure (%)	2.62 \pm 1.16	1.37 \pm 0.53	2.14 \pm 0.81	2.11 \pm 0.91	2.8 \pm 0.9

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