

1 **Identification of elite local accessions of Roselle (*Hibiscus sabdariffa*) through the  
2 evaluation of morpho-phenological characters.**

3

4 **Summary**

5 In Côte d'Ivoire, the available data on the diversity and genetic structuring of Guinea sorrel  
6 remain limited and not very popularized, thus making research work and varietal  
7 improvement efforts particularly difficult. To overcome this problem, a survey carried out in  
8 12 localities has made it possible to have an in-situ collection of 80 accessions. Twenty-nine  
9 (29) accessions from this collection were evaluated in this study. The objective was to identify  
10 elite accessions, potentially candidates for an improvement program. Nineteen (19)  
11 quantitative traits were analyzed on a Fisher block device. Descriptive analysis revealed a  
12 high morphological variability between accessions. The coefficient of variation showed a  
13 strong heterogeneity between accessions for certain traits. The Principal Component Analysis  
14 revealed 70.07% of the total variability. The classification made it possible to distinguish  
15 three groups of accessions according to their similarity. The classification by the k-means  
16 method made it possible to classify accessions into three groups and to identify the most  
17 efficient ones according to the stages of development. Thus, the accessions HSKO 037, HSFE  
18 031, HSBK 004 and Bangolo stood out respectively at the 2-leaf, vegetative, flowering, and  
19 fruiting stages by presenting the highest values for the traits evaluated.

20 **Keywords** :Identification, local elite accessions, *Hibiscus sabdariffa*, Roselle, morpho-  
21 phenological evaluation.

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23 **Introduction**

24 *Hibiscus sabdariffa L.*, commonly known as roselle, is a species belonging to the Malvaceae  
25 family (**Bakasso et al., 2013**). It is an annual or biennial plant of tropical and subtropical areas  
26 that adapts to all climatic conditions (**Kone et al., 2018**). According to **Grubben and Denton**  
27 (**2004**), it was initially domesticated in Africa, probably in Sudan, about 6000 years ago,  
28 before spreading to Asia and America. Other authors such as **Duke (1993)** and (**Abu-**  
29 **Tarboushet et al., 1997**) suggest an origin in eastern India or Saudi Arabia. Sudan is the main  
30 producer of sorrel in Africa (**Ternoyet et al., 2006**). The annual area under cultivation varies  
31 between 11,000 ha and 57,000 ha, depending on rainfall and prices. In 1995, Sudan exported  
32 about 32,000 tonnes of Guinea sorrel calyx (**Mc Clintock et al., 2011**). According to the  
33 Central Bank of Sudan (2012), the quantity of dry calyxes exported was 18,531 tonnes in

34 2011 and 15,656 tonnes in 2012 for a net gain of US\$17.59 million and US\$14.09 million,  
35 respectively (Ibrahim *et al.*, 2013). Senegal and Mali are the main producers of sorrel in West  
36 Africa. However, much of their production is for family consumption or sold in local markets  
37 (McClintock *et al.*, 2011). In Senegal, the average annual income from the sale of sorrel  
38 leaves ranges from US\$41 to US\$500 (Diouf *et al.*, 2007). In addition, in Burkina Faso,  
39 Benin, Côte d'Ivoire, Mali, Mauritania and Senegal, a 30 to 50 cl ice pack is sold for between  
40 25 and 50 CFA francs, while a 1-litre bottle for between 1,000 and 1,500 CFA francs  
41 (McClintock, 2004).

42 In Côte d'Ivoire, most research on *H. sabdariffa* is mainly carried out at the biochemical level;  
43 little information is available on the morphological, agronomic, and molecular characteristics  
44 of *H. sabdariffa* cultivars. Research activities on regeneration from cuttings (Sié *et al.*, 2008)  
45 and agromorphological characterizations (Sié *et al.*, 2009) made it possible to evaluate the  
46 diversity of 159 seeds from seeds collected on the markets of Korhogo (Côte d'Ivoire). The  
47 research of Anzaraet *et al.* (2023) made it possible to have 80 accessions collected in 12  
48 localities in Côte d'Ivoire.

49 The evaluation of accessions collected in 2023 identified 29 elite accessions. Our study  
50 proposed to evaluate the morpho-phenological characteristics of these accessions to identify  
51 the best ones. The aim was to identify the elite accessions at each stage of development and to  
52 identify the elites in a global way by cross-referencing the best at each stage.

### 53 **Material and Method**

#### 54 **Study site**

55 The study was conducted at the experimental site of the botanical garden of the Peleforo GON  
56 COULIBALY UNIVERSITY (UPGC). The experimental plot is located at longitude 5°38'  
57 West and Latitude 9°26'.

#### 58 **Plant material**

59 The plant material consists of 29 *Hibiscus sabdariffa* accessions from different localities in  
60 Côte d'Ivoire (Table I). These accessions are in the form of seeds and are the result of a  
61 survey conducted in 2023 (Anzaraet *et al.*, 2023).

#### 62 **Method**

##### 63 **Experimental design and data collection**

64 The trial was conducted using the randomized Fisher block device with three (3) replicates.  
65 The parameters measured concerned 450 plants due to 15 plants per accession.

66 Quantitative variables (**Table II**) were measured during (4) life stages: the 2-leaf stage, the  
67 vegetative stage, the flowering stage, and the fruiting stage (**Figure 1**).  
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71 **Table I.** List of accessions studied, their origin and number

| Area             | Collection location    | Accession Code    |
|------------------|------------------------|-------------------|
| Centre (05)      | Bouaké (05)            | HSBK 003          |
|                  |                        | HSBK 004          |
|                  |                        | HSBK 005          |
|                  |                        | HSBK 002          |
|                  |                        | HSBK 006          |
|                  |                        |                   |
| Centre-West (01) | Daloa (01)             | HSDA 001          |
| North (17)       | Ferkessédougou (03)    | HSBO 023          |
|                  |                        | HSBO 024          |
|                  |                        | HSBO 026          |
|                  |                        | HSFE 028          |
|                  |                        | HSFE 031          |
|                  |                        | HSFE 032          |
| Northeast (01)   | Korhogo (09)           | HSKO 037          |
|                  |                        | HSKO 040          |
|                  |                        | HSKO 042          |
|                  |                        | HSKO 043          |
|                  |                        | HSKO 044          |
|                  |                        | HSKO 046          |
|                  |                        | HSKO 047          |
|                  |                        | HSKO 052          |
|                  |                        | HSKO 053          |
| West (04)        | Niakara (02)           | HSNI 054          |
|                  |                        | HSNI 058          |
| South (01)       | Agnibilékrou (01)      | HSAG 080          |
|                  | Bangolo (01)           | Bangolo           |
| West (04)        | Bangolo campement (01) | Bangolo campement |
|                  | Duékoué (01)           | HSDU 022          |
|                  | Ziagolo (01)           | Ziagolo           |
|                  | Abobo-baoulé (01)      | Abobo-Baoulé      |
|                  | Koumassi (01)          | HSAB 015          |

72 **Source :**Prospecting data (**Anzara et al., 2023**)

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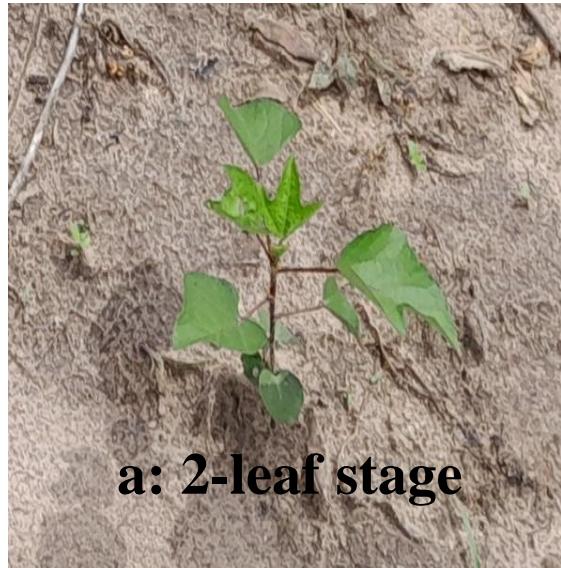
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81 **Table II.** Quantitative parameters studied during the study

| <b>Stage of development</b> | <b>Descriptors (unit)</b>                | <b>Code</b> |
|-----------------------------|--|-------------|
| Seedling (2-leaf stage)     | Germination time                         | TeGe        |
|                             | Sheetlength (cm)                         | LoFe        |
|                             | SheetWidth (cm)                          | LaFe        |
|                             | Diameter from Stem to Collar (cm)        | DiTc        |
| Vegetative stage            | ShaftDiameter (cm)                       | DiTi        |
|                             | Number of Separated Lobes                | NoLs        |
|                             | LeafPetioleLength (cm)                   | LoPf        |
|                             | LeafPetioleWidth (cm)                    | LaPf        |
|                             | Blade length                             | LoLe        |
|                             | Blade width (cm)                         | LaLe        |
|                             | Plant Height (cm)                        | HaPl        |
| Flowering stage             | First flower appearance time (days)      | TaF         |
|                             | Flower diameter (cm)                     | DiF         |
|                             | Flower PeduncleLength (cm)               | LpF         |
| Fruiting stage              | Number of Fruits per Plant               | NoFr        |
|                             | Opening date of the first capsules (day) | DoCa        |
|                             | Number of fruiting branches              | NoBf        |
|                             | Weight of 100 seeds                      | Seln        |
|                             | Number of Seeds per Fruit                | NgFr        |

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**a: 2-leaf stage**



**b: Vegetative stage**



**c: Flowering stage**



**d: Fruiting stage**

**Figure 1.** Different stages of development

## **120 Statistical processing of data**

121 Descriptive analysis at a 95% confidence interval showed the variability of the measured  
122 traits. After checking for normality, the Kruskal-wallis test was performed to check if there

123 was a difference between the accessions according to the parameters studied. When there was  
124 a difference, Dunn's post-hoc test made it possible to group them together and classify the  
125 groups obtained. A Pearson correlation test was performed, to see the link between the  
126 parameters taken two by two.

127 With K=3, the Classification by the dynamic cloud method (k-means) made it possible to  
128 divide the accessions into three different groups, namely the performers, the average  
129 performers, and the worst performers. The F-value of the ANOVA coupled with the k-means  
130 method made it possible to test the significance of the established groups at a 95% confidence  
131 level.

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## 133 **Results and Discussion**

### 134 **Results**

#### 135 **Variability of quantitative traits at each stage**

136 Descriptive analysis coupled with the Kruskal-wallis test at each stage of development  
137 revealed significant differences ( $P<0.05$ ) between accessions for all variables studied at the  
138 5% threshold except for five variables including leaf length at the 2-leaf stage, stem diameter,  
139 leaf petiole width and leaf blade length at the vegetative stage and weight of 100 seeds at the  
140 stage of fruiting.

141 At the 2-leaf stage (**Table III**), the germination time varies between 3 and 6 days with an  
142 average of  $3.87 \pm 0.95$  days, for a coefficient of variation (CV) of 25%. The average leaf  
143 length is  $2.61 \pm 0.85$  cm (CV = 33%), while the average width is  $2.41 \pm 0.64$  cm with CV =  
144 26%. As for the diameter of the stem at the collar, it shows an average of  $0.40 \pm 0.13$  cm and a  
145 coefficient of variation CV of 33%, reflecting a moderate diversity within the accessions. In  
146 the vegetative stage (**Table IV**), the stem diameter increases to an average of  $0.807 \pm 0.201$   
147 cm with a coefficient of variation of 25%, which remains relatively stable. On the other hand,  
148 the number of separated lobes varies greatly between accessions (0 to 5), with a mean of  
149  $3.393 \pm 1.329$  and a high coefficient of variation of 39.1%. The average leaf petiole length is  
150  $7.473 \pm 1.741$  (CV = 23.3%), while the leaf petiole width is highly variable and has a high  
151 coefficient of variation of 32.7%. The length and width of the blade show averages of  $10.947$   
152  $\pm 1.939$  cm (CV = 17.7%) and  $9.728 \pm 2.072$  cm (CV = 21.3), respectively. The height of the  
153 plants varies greatly, with an average of  $31,560 \pm 10,607$  cm and a coefficient of variation of  
154 33.5% showing high variability in growth between individuals. At the flowering stage (**Table**  
155 **V**), the appearance of the first flower varies from 74 to 192 days with an average of  $146.705 \pm$

156 24.809 days, for a coefficient of variation of 16.9%. While the mean flower diameter is 5.242  
 157  $\pm 1.089$  cm (CV = 20.7%), indicating a significant average variability between accessions.  
 158 The length of the flower peduncle has an average of  $1.097 \pm 0.509$  cm with a coefficient of  
 159 variation CV = 46.3%, this translates into a high variation.  
 160 Finally, at the fruiting stage (**Table VI**), the number of fruits per plant shows a high variability  
 161 with an average of  $28,898 \pm 12,080$  fruits and a high coefficient of variation of 89.4%.  
 162 Similarly, the number of fruiting branches shows a significant variation (CV = 71.5%).  
 163 However, parameters such as the date of opening of the first capsules and the weight of 100  
 164 seeds show little variation, with coefficients of variation CV = 12.7% and 18.4% respectively.  
 165 The number of seeds per fruit shows moderate variability (CV = 21.2%) with a mean of  
 166  $28.384 \pm 6.039$

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 168 **Table III.** Mean value  $\pm$  standard deviation, minimum, maximum and coefficient of variation  
 169 (CV) of the quantitative traits analyzed at the 2-sheet stage

| Statistics  | Min  | Max  | Mean $\pm$ Standard Deviation | CV (%) | F     | Pr > F   |
|-------------|------|------|-------------------------------|--------|-------|----------|
| <b>TeGe</b> | 3    | 6    | $3,87 \pm 0,95$               | 25     | 2,681 | < 0.0001 |
| <b>LoFe</b> | 1    | 5,1  | $2,61 \pm 0,85$               | 33     | 1,309 | 0,147    |
| <b>LaFe</b> | 1    | 5    | $2,41 \pm 0,64$               | 26     | 1,927 | 0,005    |
| <b>DiTc</b> | 0,11 | 0,94 | $0,40 \pm 0,13$               | 33     | 3,189 | < 0.0001 |

170 *TeGe* : Germination time, *LoFe* : Leaf length, *LaFe* : Leaf width, *DiTc* : Diameter from stem to collar

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 172 **Table IV.** Mean value  $\pm$  standard, minimum, maximum deviation and coefficient of variation  
 173 (CV) of quantitative traits analysed at the vegetative stage

| Statistics  | Mean $\pm$ Standard |       |                     |        |       |          |
|-------------|---------------------|-------|---------------------|--------|-------|----------|
|             | Min                 | Max   | Deviation           | CV (%) | F     | Pr > F   |
| <b>DiTi</b> | 0,35                | 1,69  | $0,807 \pm 0,201$   | 25,00  | 1,218 | = 0,217  |
| <b>NoLs</b> | 0,00                | 5,00  | $3,393 \pm 1,329$   | 39,1   | 6,134 | < 0.0001 |
| <b>LoPf</b> | 2,40                | 13,40 | $7,473 \pm 1,741$   | 23,3   | 1,938 | = 0,005  |
| <b>LaPf</b> | 0,20                | 9,34  | $0,443 \pm 0,146$   | 32,7   | 0,862 | = 0,667  |
| <b>LoLe</b> | 6,10                | 16,20 | $10,947 \pm 1,939$  | 17,7   | 1,059 | = 0,390  |
| <b>LaLe</b> | 4,50                | 19,50 | $9,728 \pm 2,072$   | 21,3   | 1,674 | = 0,023  |
| <b>HaPl</b> | 8,20                | 71,50 | $31,560 \pm 10,607$ | 33,5   | 5,011 | < 0.0001 |

174 *DiTi* : Diameter of the stem, *NoLs* : Number of separate lobes, *LoPf* : Length of the leaf petiole, *LaPf* :  
 175 Width of the leaf petiole, *LoLe* : Length of the blade, *LaLe* : Width of the blade, *HaPl* : Height of the  
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184 **Table V.** Mean value  $\pm$  standard deviation, minimum, maximum and coefficient of variation  
185 (CV) of quantitative traits analyzed at the flowering stage

| <b>Statistics</b> |            |            | <b>Mean</b>          | $\pm$ | <b>Standard</b> | <b>CV (%)</b> | <b>F</b> | <b>Pr &gt; F</b> |
|-------------------|------------|------------|----------------------|-------|-----------------|---------------|----------|------------------|
|                   | <b>Min</b> | <b>Max</b> | <b>Deviation</b>     |       |                 |               |          |                  |
| <b>TaF</b>        | 74,00      | 192,00     | 146,705 $\pm$ 24,809 |       |                 | 16,9          | 31,720   | < 0.0001         |
| <b>DiF</b>        | 3,168      | 7,736      | 5,242 $\pm$ 1,089    |       |                 | 20,7          | 11,986   | < 0.0001         |
| <b>LpF</b>        | 0,450      | 2,700      | 1,097 $\pm$ 0,509    |       |                 | 46,3          | 13,152   | < 0.0001         |

186 *TaF: First Flower Onset Time, DiF: Flower Diameter, LpF: Flower Peduncle Length*

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188 **Table IV.** Mean value  $\pm$  standard, minimum, maximum deviation and coefficient of variation  
189 (CV) of quantitative traits analysed at the fruiting stage

| <b>Statistics</b> |            |            | <b>Mean</b>          | $\pm$ | <b>Standard</b> | <b>CV (%)</b> | <b>F</b> | <b>Pr &gt; F</b> |
|-------------------|------------|------------|----------------------|-------|-----------------|---------------|----------|------------------|
|                   | <b>Min</b> | <b>Max</b> | <b>Deviation</b>     |       |                 |               |          |                  |
| <b>NoFr</b>       | 5          | 155        | 28,898 $\pm$ 12,080  |       |                 | 89,4          | 2,623    | < 0.0001         |
| <b>DoCa</b>       | 109        | 217        | 187,347 $\pm$ 23,780 |       |                 | 12,7          | 29,289   | < 0.0001         |
| <b>NoBf</b>       | 1          | 18         | 4,625 $\pm$ 3,311    |       |                 | 71,5          | 1,820    | = 0,010          |
| <b>NgFr</b>       | 12,6       | 41,4       | 28,384 $\pm$ 6,039   |       |                 | 21,2          | 7,444    | < 0.0001         |
| <b>Selen</b>      | 2          | 4          | 2,161 $\pm$ 0,396    |       |                 | 18,4          | 1,375    | = 0,108          |

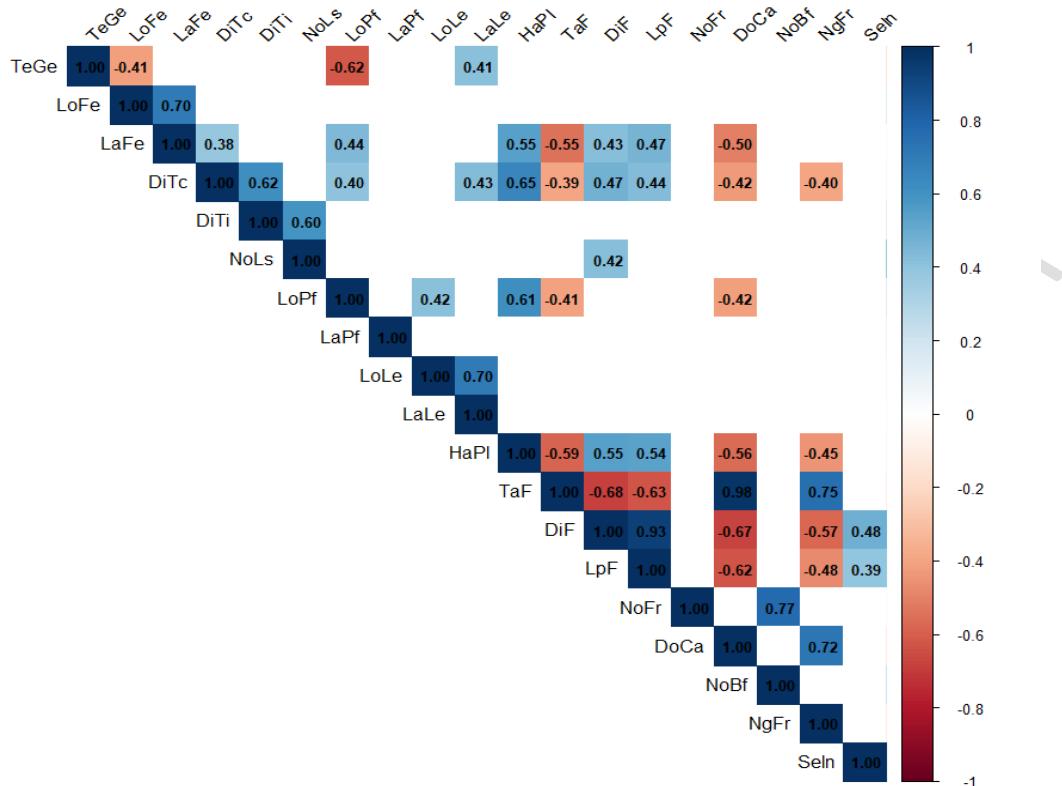
190 *NoFr: Number of Fruit per plant, DoCa: Date of opening of the first capsules, NoBf: Number of fruiting  
191 branches, Selen: Weight of 100 seeds, NgFr: Number of Seeds per fruit*

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### Relationship between the parameter traits studied

194 The Pearson correlation matrix between quantitative traits is shown in **Figure 2**. It revealed  
195 significant relationships between the traits studied, with a strong structuring of vegetative,  
196 floral and productive traits. Leaf traits are positively correlated with each other, including leaf  
197 length and width (**r = 0.70**), and leaf blade length and width (**r = 0.70**), reflecting proportional  
198 vegetative growth. Plant height is positively associated with flower diameter (**r = 0.55**) and  
199 flower peduncle length (**r = 0.54**), indicating that vigorous plants develop larger floral organs.  
200 On the other hand, the time of appearance of the first flower is negatively correlated with  
201 flower diameter (**r = -0.68**), flower peduncle length (**r = -0.63**) and number of fruits per plant  
202 (**r = -0.56**), suggesting that late flowering penalizes the expression of yield traits. The  
203 production parameters are strongly related to each other, with a very high positive correlation

204 between flower diameter and the number of fruits per plant (**r = 0.93**), as well as between the  
 205 number of fruits per plant and the number of fruiting branches (**r = 0.77**) and the number of  
 206 seeds per fruit (**r = 0.72**).



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 208 *TeGe* : Germination time, *LoFe* : Leaf length, *LaFe* : Leaf width, *DiTc* : Diameter of stem at collar, *DiTi* :  
 209 Diameter of stem, *NoLs* : Number of Separate Lobes, *LoPf* : Length of leaf petiole, *LaPf* : Width of leaf petiole,  
 210 *LoLe* : Blade length, *LaLe* : Width of leaf blade, *HaPl* : Height of plant, *TaF*: First Flower Appearance Time,  
 211 *DiF*: Flower Diameter, *LpF*: Flower Peduncle Length, *NoFr*: Number of Fruit per Plant, *DoCa*: Date of  
 212 Opening of First Capsules, *NoBf*: Number of Fruiting Branches, *Seln*: Weight of 100 Seeds, *NgFr*: Number of  
 213 Seeds per Fruit

214 **Figure 2.** Pearson correlation matrix between quantitative traits  
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## 216 Structure of the morpho-phenological diversity of accessions

217 Table **VII** presents the results of the Principal Component Analysis performed on quantitative  
 218 traits. This analysis made it possible to identify five main axes that explain 70.07% of the  
 219 total variability observed. Axis 1 accounted for 30.39% of the total variability. The variables  
 220 that contributed most to the formation of this component were the time of appearance of the  
 221 first flower (TaF), the date of opening of the first capsules (DoCa) and the number of seeds  
 222 per fruit. All these variables were positively correlated with axis 1. The variables leaf width  
 223 (LaFe), stem diameter at crown (DiTc), leaf petiole length (LoPf), plant height (HaPl), flower  
 224 diameter (DiF), and flower peduncle length (LpF) were negatively correlated with this axis,  
 225 this axis can be referred to as the vegetative vigour axis. Axis 2, which represents 14.75% of

226 the total variance, combining germination time (TeGe), stem diameter (DiTi), blade length  
 227 (LaLe) and number of fruits per plant. These variables were positively correlated with axis 2.  
 228 This axis can be considered as an axis of potential yield and robustness. Axis 3, representing  
 229 12.20% of the total inertia, positively associates the variable limb length (LoLe). The variable  
 230 number of separate lobes is negatively correlated with this axis. Axis 4, showing 10.53% of  
 231 the total variability, positively associates the variable number of fruiting branches (NoBf).  
 232 Axis 5 (7.21%) was formed, primarily, from leaf length.

233 **Table VII.** Eigenvalues, percent variance and correlation between morphophenological traits  
 234 and the five main PCA factors

|                        | <b>Axis 1</b>                             | <b>Axis 2</b> | <b>Axis 3</b> | <b>Axis 4</b> | <b>Axis 5</b> |
|------------------------|---|---------------|---------------|---------------|---------------|
| <b>Eigenvalue</b>      | 5,772                                     | 2,802         | 2,319         | 2,001         | 1,370         |
| <b>Variability (%)</b> | 30,378                                    | 14,745        | 12,204        | 10,532        | 7,210         |
| <b>Cumulative %</b>    | 30,378                                    | 45,123        | 57,327        | 67,859        | 75,070        |
| <b>Variable</b>        | Correlation between variables and factors |               |               |               |               |
| <b>TeGe</b>            | 0,377                                     | <b>0,538</b>  | -0,313        | -0,439        | -0,008        |
| <b>LoFe</b>            | -0,279                                    | -0,486        | 0,216         | 0,216         | <b>0,561</b>  |
| <b>LaFe</b>            | <b>-0,688</b>                             | -0,321        | 0,278         | -0,078        | 0,333         |
| <b>DiTc</b>            | <b>-0,655</b>                             | 0,481         | 0,173         | -0,172        | 0,060         |
| <b>DiTi</b>            | -0,440                                    | <b>0,479</b>  | -0,096        | -0,129        | <b>0,472</b>  |
| <b>NoLs</b>            | -0,160                                    | 0,599         | <b>-0,649</b> | 0,155         | 0,292         |
| <b>LoPf</b>            | <b>-0,612</b>                             | -0,085        | <b>0,562</b>  | 0,142         | -0,071        |
| <b>LaPf</b>            | 0,086                                     | -0,018        | <b>0,401</b>  | -0,166        | 0,313         |
| <b>LoLe</b>            | -0,109                                    | <b>0,506</b>  | <b>0,725</b>  | -0,103        | 0,054         |
| <b>LaLe</b>            | -0,131                                    | <b>0,684</b>  | 0,317         | -0,476        | 0,062         |
| <b>HaPl</b>            | <b>-0,770</b>                             | 0,064         | 0,344         | -0,025        | -0,274        |
| <b>TaF</b>             | <b>0,860</b>                              | 0,149         | 0,166         | 0,085         | 0,269         |
| <b>DiF</b>             | <b>-0,843</b>                             | 0,136         | -0,319        | 0,164         | 0,014         |
| <b>LpF</b>             | <b>-0,809</b>                             | -0,030        | -0,281        | 0,221         | 0,028         |
| <b>NoFr</b>            | 0,156                                     | <b>0,650</b>  | 0,153         | <b>0,553</b>  | -0,260        |
| <b>DoCa</b>            | <b>0,848</b>                              | 0,114         | 0,181         | 0,097         | 0,255         |
| <b>NoBf</b>            | -0,076                                    | 0,345         | 0,210         | <b>0,804</b>  | -0,190        |
| <b>NgFr</b>            | <b>0,706</b>                              | 0,047         | 0,245         | 0,410         | 0,196         |
| <b>Selen</b>           | -0,270                                    | 0,139         | -0,217        | 0,433         | 0,409         |

235 *TeGe* : Germination time, *LoFe* : Leaf length, *LaFe* : Leaf width, *DiTc* : Diameter of stem at collar;  
 236 *DiTi* : Diameter of stem, *NoLs* : Number of Separate Lobes, *LoPf* : Length of leaf petiole, *LaPf* : Width  
 237 of leaf petiole, *LoLe* : Blade length, *LaLe* : Width of leaf blade, *HaPl* : Height of plant, *TaF*: First  
 238 Flower Appearance Time, *DiF*: Flower Diameter, *LpF*: Flower Peduncle Length, *NoFr*: Number of  
 239 Fruit per Plant, *DoCa*: Date of Opening of First Capsules, *NoBf*: Number of Fruiting Branches, *Seln*:  
 240 Weight of 100 Seeds, *NgFr*: Number of Seeds per Fruit

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248 **Morphophenological groups of Guinea sorrel accessions**

249 *At the 2-leaf stage*

250 **Class 1** includes accessions with longer germination times (TeGe=3.961 days), longer leaves  
251 (LoFe=2.871 cm) and widest leaves (LaFe=2.587 cm), reflecting good morphological  
252 development from this early stage.

253 **Class 2** is distinguished by intermediate values for most of the measured traits. The  
254 germination time is the shortest (TeGe=3.441 days), but the leaves are significantly smaller  
255 than those of class 1. This class is then composed of the moderately growing accessions, but  
256 with faster germination.

257 **Class 3** is characterized by a high value of germination time, showing a longer time for  
258 seedling emergence, however it has the lowest values for other traits such as leaf length, leaf  
259 width and stem diameter at the collar, suggesting a slower initial development despite late  
260 germination. Only the length of the leaves (LoFe) made it possible to clearly differentiate the  
261 three groups formed (**Table VIII**).

262 **Table VIII.** Average characteristics of K-Means classes at the 2-leaf stage

| Characters          | Class 1             | Class 2            | Class 3            | Pr > F(Model)     |
|---------------------|---------------------|--------------------|--------------------|-------------------|
| TeGe                | 3,961 <sup>ab</sup> | 3,441 <sup>b</sup> | 4,286 <sup>a</sup> | <b>0,007</b>      |
| LoFe                | 2,871 <sup>a</sup>  | 2,548 <sup>b</sup> | 2,163 <sup>c</sup> | <b>&lt;0.0001</b> |
| LaFe                | 2,587 <sup>a</sup>  | 2,371 <sup>a</sup> | 2,100 <sup>b</sup> | <b>&lt;0.0001</b> |
| DiTc                | 0,42                | 0,412              | 0,362              | <b>0,132</b>      |
| <b>2-leaf stage</b> | HSKO 044_A          | Ziagolo_S          | HSDA 001_A         |                   |
|                     | HSFE 031_S          | HSAB 015_S         | HSBK 006_A         |                   |
|                     | HSKO 047_S          | HSFE 032_A         | HSNI 058_S         |                   |
|                     | HSAG 080_A          | HSBO 024_A         | HSBK 003_A         |                   |
|                     | Bangolocampement_S  | HSBK 002_S         | HSKO 040_A         |                   |
|                     | HSFE 028_A          | HSKO 052_S         | HSKO 042_S         |                   |
|                     | Bangolo_S           | HSBO 026_S         | HSNI 054_S         |                   |
|                     | HSKO 043_A          |                    | Abobo-Baoulé_A     |                   |
|                     | HSKO 037_A          |                    | HSBO 023_A         |                   |

HSBK 004\_S  
HSKO 053 A

HSKO 046\_A

263 *TeGe* : Germination time, *LoFe* : Leaf length, *LaFe* : Leaf width, *DiTc* : Diameter from stem to collar

264

265 *In the vegetative stage*

266 **Class 1** is characterized by a high plant height (HaPl=37.74 cm), a large blade length  
267 (Lole=11.19 cm) and a noticeable blade width (LaLe=9.97 cm). These accessions therefore  
268 show good vegetative development and have a high growth potential.

269 **Class 2** is composed of accessions with a higher number of separated lobe (NoLs=4), a length  
270 and width of the blade comparable to that of Class 1, but a slightly lower plant height.

271 **Class 3**, on the other hand, has the lowest values for many of the traits studied. (**Table IX**).  
272 Only leaf petiole length (LoPf) and plant height (HaPl) significantly differentiated the three  
273 classes. However, only the height of the plant made it possible to differentiate them distinctly.

**Table IX.** Average characteristics of K-Means classes at the vegetative stage

| Characters       | Class 1             | Class 2             | Class 3             | Pr > F(Model)      |
|------------------|---------------------|---------------------|---------------------|--------------------|
| DiTi             | 0,797               | 0,402               | 0,772               | <b>0,643</b>       |
| NoLs             | 2,682               | 3,838               | 3,449               | <b>0,071</b>       |
| LoPf             | 7,970 <sup>a</sup>  | 7,540 <sup>a</sup>  | 6,681 <sup>b</sup>  | <b>0</b>           |
| LaPf             | 0,429               | 0,41                | 0,47                | <b>0,701</b>       |
| LoLe             | 11,194              | 11,203              | 10,573              | <b>0,162</b>       |
| LaLe             | 9,977               | 9,94                | 9,588               | <b>0,743</b>       |
| HaPl             | 37,742 <sup>a</sup> | 30,624 <sup>b</sup> | 23,853 <sup>c</sup> | <b>&lt; 0.0001</b> |
|                  |                     |                     |                     |                    |
| Vegetative stage | HSBK 006_A          | HSAG 080_A          | HSDA 001_A          |                    |
|                  | Bangolocampement_S  | HSBO 024_A          | HSKO 047_S          |                    |
|                  | HSAB 015_S          | HSKO 042_S          | HSBO 023_A          |                    |
|                  | HSBK 004_S          | HSKO 040_A          | HSBK 002_S          |                    |
|                  | HSFE 032_A          | HSBO 026_S          | HSKO 053_A          |                    |
|                  | HSKO 043_A          | HSKO 044_A          | HSKO 046_A          |                    |
|                  | Ziagolo_S           | HSNI 054_S          | HSFE 028_A          |                    |
|                  | HSKO 037_A          |                     | HSKO 052_S          |                    |
|                  | Bangolo_S           |                     | HSNI 058_S          |                    |
|                  | HSBK 003_A          |                     | Abobo-Baoulé_A      |                    |
|                  |                     |                     |                     |                    |
|                  |                     |                     |                     |                    |
|                  |                     |                     |                     |                    |

275 *DiTi* : Diameter of the stem, *NoLs* : Number of separate lobes, *LoPf* : Length of the leaf petiole, *LaPf* : Width of  
276 the leaf petiole, *LoLe* : Length of the blade, *LaLe* : Width of the blade, *HaPl* : Height of the plant  
277

278

279

280 ***In the flowering stage***

281 **Class 1** occupies an intermediate position, with late flowering and medium-sized flowers  
282 (*DiF*=5.8 cm).

283 **Class 2** has an early flowering (*TaF*=108.26 days), large diameter flowers (*DiF*=6.14 cm) and  
284 relatively longer peduncles. The accessions of this class are the earliest to flower with large  
285 fruits with long stalks.

286 **Class 3** is characterized by the highest time of appearance of the first flowers (*TaF*=161.38  
287 days), associated with the smallest flower diameter (*DiF*=4.69 cm) and the length of the  
288 flower peduncle (*LpF*=0.837). This shows that the accessions of this class are characterized  
289 by a late flowering accompanied by smaller flowers. (**Table X**)

290 **Table V.** Average characteristics of K-Means classes at the flowering stage

| Characters             | Class 1              | Class 2              | Class 3              | Pr > F(Model) |
|------------------------|----------------------|----------------------|----------------------|---------------|
| TaF                    | 143,609 <sup>b</sup> | 108,268 <sup>c</sup> | 161,703 <sup>a</sup> | < 0.0001      |
| DiF                    | 5,800 <sup>a</sup>   | 6,138 <sup>a</sup>   | 4,660 <sup>b</sup>   | < 0.0001      |
| LpF                    | 1,381 <sup>a</sup>   | 1,469 <sup>a</sup>   | 0,811 <sup>b</sup>   | < 0.0001      |
| <hr/>                  |                      |                      |                      |               |
| <b>Flowering stage</b> |                      |                      |                      |               |
|                        | HSKO 046_A           | Ziagolo_S            | HSKO 044_A           |               |
|                        | HSAB 015_A           | Bangolocampement_S   | HSDA 001_A           |               |
|                        | HSBO 026_S           | HSBK 004_S           | HSKO 047_S           |               |
|                        | HSKO 040_A           | HSFE 031_S           | HSKO 042_S           |               |
|                        |                      | Bangolo_S            | HSFE 028_A           |               |
|                        |                      | HSKO 037_A           | HSFE 032_A           |               |
|                        |                      | HSKO 043_A           | HSBO 023_A           |               |
|                        |                      |                      | HSBO 024_A           |               |
|                        |                      |                      | HSKO 053_A           |               |
|                        |                      |                      | HSBK 006_A           |               |
|                        |                      |                      | HSAG 080_A           |               |
|                        |                      |                      | HSBK 002_S           |               |
|                        |                      |                      | HSKO 052_S           |               |
|                        |                      |                      | HSNI 054_S           |               |

HSNI 058\_S  
HSBK 003\_A  
Abobo-Baoulé\_A

291 *TaF: First Flower Onset Time, DiF: Flower Diameter, LpF: Flower Peduncle Length*

292

293 ***At the fruiting stage***

294 **Class 1** shows an intermediate position, with an average number of fruits (25.51) and seeds  
295 (30.11), but with a longer capsule opening time (DoCa) (201.97 days), which could slow  
296 down the availability of seeds at maturity.

297 **Class 2** brings together the best performing accessions in terms of productivity with the  
298 highest number of fruits per plant (46,046) and a high number of seeds per plant (31,27). It  
299 also has the highest number of fruiting branches (6.31).

300 **Class 3** is characterized by the lowest values in number of fruits per plant (20.86) and number  
301 of seeds per fruit. (**Table XI**)

302 **Table XI.** Average characteristics of K-Means classes at the fruiting stage

| Characters     | Class 1              | Class 2              | Class 3              | Pr > F(Model) |
|----------------|----------------------|----------------------|----------------------|---------------|
| NoFr           | 25,514 <sup>b</sup>  | 46,046 <sup>a</sup>  | 20,864 <sup>b</sup>  | < 0.0001      |
| DoCa           | 201,971 <sup>a</sup> | 191,117 <sup>a</sup> | 158,476 <sup>b</sup> | < 0.0001      |
| NoBf           | 4,198 <sup>a</sup>   | 6,308 <sup>a</sup>   | 4,230 <sup>b</sup>   | 0,008         |
| NgFr           | 30,269 <sup>a</sup>  | 31,269 <sup>a</sup>  | 22,958 <sup>b</sup>  | < 0.0001      |
| Selen          | 2,115                | 2,156                | 2,190                | 0,54          |
| Fruiting stage | HSBO 023_A           | HSKO 037_A           | Ziagolo_S            |               |
|                | HSKO 042_S           | HSFE 031_S           | HSBK 006_A           |               |
|                | HSAG 080_A           | HSBK 004_S           | HSBO 026_S           |               |
|                | HSBK 002_S           | HSKO 046_A           | Bangolocampement_S   |               |
|                | HSBO 024_A           | Bangolo_S            | HSAB 015_S           |               |
|                | HSKO 040_A           |                      | HSKO 044_A           |               |
|                | Abobo-Baoulé_A       |                      |                      |               |
|                | HSKO 047_S           |                      |                      |               |
|                | HSKO 053_A           |                      |                      |               |
|                | HSNI 058_S           |                      |                      |               |
|                | HSNI 054_S           |                      |                      |               |
|                | HSFE 028_A           |                      |                      |               |
|                | HSFE 032_S           |                      |                      |               |
|                | HSKO 043_A           |                      |                      |               |
|                | HSBK 003_A           |                      |                      |               |
|                | HSDA 001_A           |                      |                      |               |
|                | HSKO 052_S           |                      |                      |               |

303 *NoFr: Number of Fruit per plant, DoCa: Date of opening of the first capsules, NoBf: Number of fruiting*  
304 *branches, Seln: Weight of 100 seeds, NgFr: Number of Seeds per fruit*

305

306

307

308

309 **Selection of elite accessions by stage of development**

310 The k-means classification made it possible to select the best ones at each stage of  
311 development. At the 2-leaf stage, Class 1, shows the highest values for leaf length, leaf width  
312 and stem diameter at the collar, reflecting good morphological development from this early  
313 stage. The elite accessions at this stage are **HSKO 044, HSFE 031, HSKO 047, HSAG 080,**  
314 **Bangolo camp, HSFE 028, Bangolo, HSKO 043, HSKO 037, HSBK 004, HSKO 053.** At  
315 the vegetative stage, the accessions (**HSBK 006, Bangolocampement, HSAB 015, HSBK**  
316 **004, HSFE 032, HSKO 043, Ziagolo, HSKO 037, Bangolo, HSBK 003, HSFE 031**)  
317 belonging to class 1 appear to be the most promising in terms of vegetative vigour. Class 2  
318 from the flowering stage has the most favourable characteristics early flowering, large  
319 flowers, long peduncle, which is often sought after in varietal improvement. The elite  
320 accessions belonging to this class are **Ziagolo, Bangolo camp, HSBK 004, HSFE 031**  
321 **Bangolo, HSKO 037, HSKO 043.** Class 2 at the fruiting stage, whose elite accessions are  
322 **HSKO 037, HSFE 031, HSKO 004, HSKO 046, Bangolo,** was distinguished by its high  
323 yield potential, characterized by a high number of fruits per plant, many seeds per fruit. Some  
324 accessions are consistently found in the best performing classes. These accessions (**HSKO**  
325 **037, HSFE 031, HSBK 004, Bangolo**), which can be described as elite accessions, have good  
326 vigour at the 2-leaf stage, balanced vegetative growth, abundant flowering and a high yield of  
327 fruit and calyx at maturity.

328

329 **Discussion**

330 The morpho-phenological evaluation of the different accessions at each stage of development  
331 carried out based on quantitative parameters, revealed a heterogeneity between accessions, as  
332 indicated by the Kruskal-wallis test. High values of coefficients of variation of traits such as  
333 leaf length, stem diameter at collar, number of separated lobes, leaf petiole width, plant  
334 height, peduncle length, number of fruits per plant, number of fruiting branches. According to  
335 **Aljane and Ferchichi (2007)**, a high value of the coefficient of variation (30%) reflects the  
336 heterogeneity of the material studied. The mixing of several morphotypes within the same

337 accession and the spontaneous shattering at maturity of certain accessions, which leads to  
338 seed losses, can also explain these results. Regarding morpho-phenological performance, the  
339 leaves varied from 4.5 cm to 19.5 cm (width) and from 6.1 cm to 16.20 cm (length) with  
340 petioles with an average length of 7.47 cm. According to **McClintock *et al.* (2011)**, the leaf  
341 blade can reach 9 to 15 cm long and 9 to 20 cm wide in Senegalese varieties, and 10 to 16 cm  
342 long and 10 to 20 cm wide for Thai varieties (**Bakasso, 2010; Hien, 2012**). The blade is  
343 connected to the stem by a petiole 4 to 12 cm long. The average number of fruits per plant  
344 (28.38 capsules) remains low compared to the results of **Bakasso (2010)**, which could be  
345 attributed to temperature variation and an early cessation of rains, resulting in flower bud  
346 drop. The accessions collected have a flowering date between 74 and 192 days. Our results  
347 differ from those of **Bakasso (2010)** and **Satyanarayana *et al.* (2017)** which found  
348 respectively a cycle of 65 to 97 days for 50% flowering for genotypes grown in Niger and 153  
349 to 163 days for 50% flowering for genotypes grown in India. According to **Islam *et al.***  
350 (**2008**), the flowering date of *Hibiscus sabdariffa* depends not only on environmental  
351 conditions and genotypes but also on the sowing date. **Hacket and Carolene (1982)** reported  
352 that *Hibiscus sabdariffa* was sensitive to day length (i.e., it was a short-day plant that  
353 flowered when day length shortened).

354 The correlations observed between the variables, whether positive or negative, could be  
355 exploited in a breeding program. For example, positive correlations are observed between leaf  
356 blade width and blade length ( $r = 0.70$ ), petiole length and blade width ( $r = 0.58$ ), plant height  
357 and flower diameter ( $r = 0.39$ ), and petiole length and leaf blade width ( $r = 0.58$ ) show that  
358 more vigorous plants produce more leaves and fruit. These results are like those obtained by  
359 **Islam *et al.* (2008)** and **Bakasso (2010)**. Similarly, the positive and significant correlation  
360 between the date of opening of the first capsules and the time of appearance of the first  
361 flowers ( $r = 0.88$ ), the length of the peduncle and the diameter of the flower ( $r = 0.82$ ) and  
362 between the number of fruiting branches and the number of fruits per plant ( $r = 0.81$ ), shows  
363 that certain morphological characteristics are directly related to reproductive performance and  
364 yield. In addition, negative correlations between certain variables, such as plant height and  
365 first flower appearance time ( $r = -0.47$ ), first flower appearance time and flower diameter ( $r =$   
366  $-0.60$ ), show that floral earliness is often associated with better vegetative performance.

367 Analysis by the classification of dynamic swarms applied to the different stages of  
368 development has made it possible to highlight three homogeneous groups ranked according to  
369 the stage of development. At the 2-leaf stage, Class 1, shows the highest values for leaf  
370 length, leaf width and stem diameter at the collar, reflecting good morphological development

371 from this early stage. Also at the vegetative stage, accessions belonging to class 1 appear to be  
372 the most promising in terms of vegetative vigour. Class 2 from the flowering stage has the  
373 most favourable characteristics early flowering, large flowers, long peduncle, which is often  
374 sought after in varietal improvement. Class 2 at the fruiting stage was distinguished by a high  
375 yield potential, characterized by a high number of fruits per plant, many seeds per fruit. The  
376 analysis shows that the most successful accessions are not always consistent from one stage to  
377 another. Some early accessions at the 2-leaf stage showed average vegetative vigour  
378 thereafter, while others that were slower at the start expressed better agronomic performance  
379 at maturity. However, a few accessions stood out for their regularity in the most successful  
380 classes at all stages observed. This phenotypic stability suggests good adaptability and high  
381 genetic potential. These accessions, considered elite, represent priority candidates for varietal  
382 improvement programs.

383

## 384 Conclusion

385 The goal of plant breeding is to create cultivars. It requires both elite accessions for their  
386 characteristics and a good understanding of them.

387 The objective of our study was to identify elite accessions of Guinea sorrel through their  
388 morpho-phenological evaluation during four (4) stages of development.

389 The results obtained based on the quantitative traits revealed a high variability for several  
390 traits related to growth and yield, such as plant height, leaf length, stem diameter and the  
391 number of fruits per plant. The structuring of the morpho-phenological diversity of accessions  
392 revealed three groups, regardless of the geographical origin of accessions. This study also  
393 highlighted important correlations between several variables related to yield and plant cycle.  
394 The comparison of accessions over the four stages of development made it possible to  
395 identify, at each stage, elite accessions. Some of them, such as **HSKO 037, HSFE 031,**  
396 **HSBK 004** and**Bangolo**, have shown high stability and performance over the entire cycle, and  
397 could thus be used as starting material for a Guinea sorrel breeding programme.

398

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