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19 **Effects of stand composition and soil properties on water repellency and hydraulic**
20 **conductivity in Mediterranean forests**

21

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37 **Running ahead:** Water repellency and hydraulic conductivity in Mediterranean forests

38

39 **Keywords:** Organic matter, mixed forest, soil texture, water infiltration,
40 hydrophobicity.

41 **Abstract**

42

43 This study evaluates soil hydraulic conductivity (SHC) and water repellency (SWR) in
44 three mixed forest stands in relation to site plant and soil characteristics. The studied
45 forest stands were: i) *Pinus nigra* Arn. ssp *salzmannii* and *Quercus ilex*; ii) *Pinus nigra*
46 Arn. ssp *salzmannii* and *Juniperus Thurifera*; iii) *Pinus nigra* Arn. ssp *salzmannii*,
47 *Quercus ilex* and *Juniperus Thurifera*. A 100-120 years old *Pinus nigra* Arn. ssp
48 *salzmannii* stand, not subjected to any management practice was also chosen as control.
49 The hydrological variables, physico-chemical properties and surface characteristics of
50 soils were surveyed. Soil water infiltration was higher in *Pinus* + *Juniper* mixed forest,
51 and *Pinus* + *Quercus* + *Juniper* mixed forests compared to unmanaged *Pinus* stands.
52 None of the studied forest stands shows a high level of repellency. Only a slight
53 repellency (in unmanaged stands dominated by pines) or moderate repellency (in soils
54 with *Pinus* and *Juniper*) were evident, while soils with *Pinus* and *Quercus* were not
55 repellent. Differences in SHC among the forest species were driven primarily by the soil
56 texture and associated structure , and secondarily by soil organic matter and associated
57 SWR. The latter was mainly due to organic matter content of the soils, but others of the
58 soil physico-chemical properties and covers analysed were found as influencing
59 parameters to discriminate SWR among mono-specific and mixed forest stands. While
60 SHC at the studied forest stands could be predicted using organic matter as well as sand
61 and clay contents of the soil, SWR is the result of several hydrological and physico-
62 chemical parameters of Mediterranean forest soils.

63

64 **Keywords:** forest stand; water infiltration; soil structure; organic matter; forest
65 biodiversity; soil properties.

67 **List of abbreviations**

- 68 SHC = soil hydraulic conductivity
69 SWR = soil water repellency
70 PN+QI = forest stand of *Pinus nigra* Arn. ssp *salzmannii* and *Quercus ilex*
71 PN+JT = forest stand of *Pinus nigra* Arn. ssp *salzmannii* and *Juniperus thurifera*
72 PN+QI+JT = forest stand of *Pinus nigra* Arn. ssp *salzmannii*, *Quercus ilex* and
73 *Juniperus Thurifera*
74 PNUM = unmanaged forest stands of 100-120 years old *Pinus nigra* Arn. ssp
75 *salzmannii*
76 RC = rock cover of soil
77 VC = vegetation cover of soil
78 BSC = bare soil cover
79 DWC = dead wood cover on soil
80 SaC = sand content of soil
81 SiC = silt content of soil
82 CIC = clay content of soil
83 SWC = soil water content
84 BD = bulk density of soil
85 EC = electrical conductivity of soil
86 OM = organic matter content of soil
87 TN = total nitrogen of soil
88 WDPT = Water Drop Penetration Test
89 PERMANOVA = Multivariate Permutational Analysis Of Variance
90 ANOSIM = Analysis Of Similarities
91 NMDS = Non-metric Multi-Dimensional Scaling
92 DISTLM = Distance-Based Linear Modeling
93 dbRDA = distance-based Redundancy Analysis

94 **1. Introduction**

95

96 The Mediterranean climate is characterized by frequent and intense rainstorms,
97 often generating high-magnitude flash floods (Fortugno et al., 2017). Moreover, the
98 soils of the Mediterranean Basin are generally shallow with low aggregate stability and
99 organic matter and nutrient contents (Cantón et al., 2011). The combination of these
100 climate and soil characteristics can make Mediterranean areas prone to excessive runoff
101 and high soil erosion rates (Zema et al., 2020a; Zema et al., 2020b). Since soil hydraulic
102 conductivity (SHC) can be low in these soils, the hydrological processes generating
103 runoff and erosion on Mediterranean soils tend to be dominated by the infiltration-
104 excess mechanism (Lucas-Borja et al., 2018). The hydrology of Mediterranean soils
105 may also be affected by soil water repellency (SWR) (Cerdà & Doerr, 2007), which is
106 primarily determined by the accumulation of long-chain organic compounds in/around
107 soil particles. SWR can stimulate land degradation processes by reducing the affinity of
108 soil and water, thereby triggering a reduction in soil fertility and increasing soil and
109 water losses (Keesstra et al., 2017). SWR can reduce the infiltration capacity of soils
110 and increase surface runoff, resulting in soil erosion and the transport of entrained
111 pollutants (e.g. Imeson et al., 1992; Ritsema et al., 1993; Ritsema et al., 1997;
112 Shakesby et al., 1993). SWR can, thereby, reduce SHC (e.g. Imeson et al., 1992; Plaza-
113 Álvarez et al., 2019; Van Dam et al., 1990), causing Hortonian infiltration-excess
114 overland flow during intense storms (Doerr et al., 2000).

115 Due to the synergistic effects of high SWR and low SHC, infiltration rates of
116 Mediterranean soils can be several orders of magnitude lower than would be expected
117 (DeBano, 1971; Doerr et al., 2003; 2000), with heavy environmental on-site (e.g., soil
118 loss, landslides) and off-site impacts (e.g., transport of entrained pollutants, damage of

119 urban infrastructures). Therefore, the relationship between SWR and SHC is very
120 important to control and mitigate the hydrological risks and the related environmental
121 hazards (Lucas-Borja et al., 2018; Plaza-Álvarez et al., 2018; Di Prima et al., 2018).
122 Moreover, soil hydraulic and hydrological properties in forests are noticeably
123 influenced by disturbances (Bormann & Klaassen, 2008; Lucas-Borja et al., 2019;
124 Lucas-Borja et al., 2019; Di Prima et al., 2020), such as human management.
125 Understanding SWR and SHC dynamics is thus of fundamental importance in the
126 Mediterranean forest ecosystems, which are subject to many disturbances (e.g., wildfire,
127 biodiversity lost, drought) in addition to the hydrological risks.

128 Variability in SHC across forest ecosystems can be substantial ($1-10^3 \text{ mm h}^{-1}$,
129 Carlyle-Moses et al., 2018, and references therein); moreover, forest soils have been
130 observed to span the full range of SWR, from negligible to extreme repellency (Mao et
131 al., 2019). Generally, forest species (including their litter), and the physico-chemical
132 and microbial community characteristics of the soil appear to drive SWR and SHC
133 variability (Neary et al., 1999). These characteristics and the related physical, chemical
134 and biological processes have been widely investigated across Mediterranean
135 ecosystems using various approaches (e.g., Martín & Vila, 2012; Pausas et al., 2004;
136 Sardans & Peñuelas, 2013). Especially in Mediterranean forests, SHC and SWR have
137 been deeply explored under a variety of pedological, climatic and management
138 conditions (e.g., Inbar et al., 2014; Neris et al., 2013; Wittenberg et al., 2011). Less
139 attention has been paid to the interactive influence of forest species and soil
140 characteristics on SWR and SHC in Mediterranean forests. An improved knowledge of
141 these vegetation-soil-infiltration relationships may yield valuable insights into the
142 hydrological processes of these delicate ecosystems and help improve management
143 strategies for runoff control and soil conservation.

144 To fill this gap, this study evaluates SHC and SWR in four 100-year-old forest
145 stands, containing tree species that are common in the Mediterranean region -*Pinus*
146 *nigra* Arn. ssp *salzmannii* and *Quercus ilex* (PN+QI); *Pinus nigra* Arn. ssp *salzmannii*
147 and *Juniperus thurifera* (PN+JT); *Pinus nigra* Arn. ssp *salzmannii*, *Quercus ilex* and
148 *Juniperus thurifera* (PN+QI+JT), and in a 100-120 years old *Pinus nigra* Arn. ssp
149 *salzmannii* stand of Castilla-La Mancha (Central-Eastern Spain) - not subject to any
150 management operations - in relation to the plant and soil characteristics. The study aims
151 at identifying the soil properties that mainly drive the SHC and SWR in the investigated
152 forest species. To these aims, we hypothesize that soil texture and organic matter may
153 play a fundamental role in driving these important hydrological parameters of forest
154 soils.

155

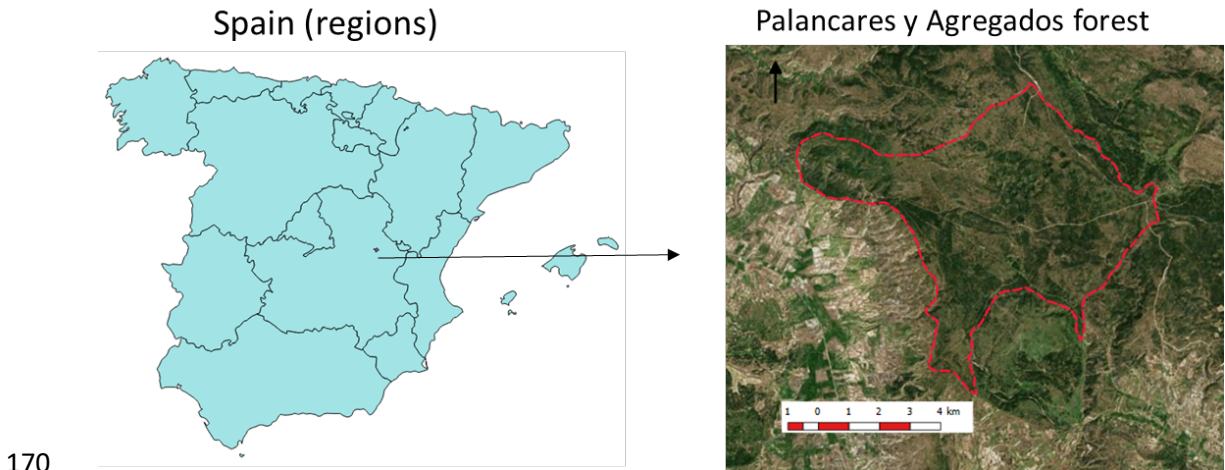
156 **2. Material and methods**

157

158 *2.1. Study area*

159

160 The study area is located in “Los Palancares y Agregados” forest, inside the largest
161 natural reserve of Castilla-La Mancha (18,078 ha, Central-Eastern Spain, Figure 1)
162 (Lucas-Borja et al., 2012; 2016). This forest covers about 4900 ha (40°01'50''N;
163 1°59'10''W) at an average elevation of 1200 m above sea level and consists of 85
164 management units. According to the Köppen-Geiger classification, the climate of the
165 study area is Mediterranean humid (Csa). The mean annual temperature is 11.9 °C, with
166 a mean lowest temperature of the coldest month equal to -0.5 °C and a mean highest
167 temperature of the hottest month equal to 30.5 °C. The mean annual precipitation is 595
168 mm, of which only 99 mm occurring in summer.



170

171 Figure 1 - Location of the “Los Palancares y Agregados” forest (Castilla La Mancha,
172 Spain).

173

174 The soils of this area are classified as Entisols, according to the USDA soil
175 taxonomy (Soil Survey Staff, 1999), with clay loam or loam texture. The study area is
176 dominated by mixed natural forests of Spanish black pine (*Pinus nigra* Arn. ssp
177 *salzmannii*), Holm oak (*Quercus ilex* L.), Portuguese oak (*Quercus faginea* Lam) and
178 Spanish juniper (*Juniperus thurifera* L.). The herbaceous and shrub vegetation is
179 characterized mainly by *Juniperus oxicedrus* L., *Rosa* sp., *Eryngium campestre* L.,
180 *Geranium selvaticum* L., *Centaurea paniculata* L., and *Plantago media* L. This type of
181 forest ecosystem is included in the European Union endangered habitats, a list of natural
182 habitats requiring specific conservation measures (Resolution 4/1996 by the Convention
183 on the Conservation of European Wildlife and Natural Habitats), and in the Protected
184 Areas list of the government of Castilla La Mancha (2/2001, Official Diary of Castilla
185 La Mancha N° 8). The forests have been managed since 1895 using the shelterwood
186 method with a shelter-phase of 20 years and a rotation period of 100 years. This
187 shelterwood management practice has been continually practiced to date, which means

188 that several classes of tree age have been generated in approximately 120 years across
189 the management units. The regeneration method in both mixed and pure even-aged
190 Spanish black pine stands consists of a uniform opening of the canopy without soil
191 preparation. The main effort of this forest management plan aims at increasing forest
192 standing stock and transforming age-heterogeneous stands into even-aged stands.

193

194 2.2. Experimental design

195

196 In May 2019, nine plots, each of 50 ha, were selected according to the forest area
197 management plan (Lucas-Borja et al., 2016) of the studied forest:

198 i) three plots mainly covered by *Pinus nigra* Arn. ssp *salzmannii* and *Quercus ilex*
199 (hereinafter indicated as PN+QI);

200 ii) three plots of *Pinus nigra* Arn. ssp *salzmannii* and *Juniperus thurifera* (PN+JT);

201 iii) three plots of *Pinus nigra* Arn. ssp *salzmannii*, *Quercus ilex* and *Juniperus thurifera*
202 (PN+QI+JT).

203 Three smaller reference plots, each one of about one hectare, were also considered,
204 mainly covered by unmanaged forest stands of 100-120 years old *Pinus nigra* Arn. ssp
205 *salzmannii* (PNUM). The stand structural characteristic across the study plots were as
206 follows: (i) PN+QI plots had a basal area and tree height of 21–28 m²/ha and 20–25 m,
207 respectively; (ii)- PN+JT stands had basal area of 20–26 m²/ha and tree height of 21–24
208 m ; (iii) basal area was 22–27 m²/ha and tree height was 20–23 m for PN+QI+JT stands;
209 (iv) PNUM plots showed a basal area and tree height of 80–82 m²/ha and 35–40 m,
210 respectively. All forest stands have been growing under the same climatic conditions
211 during the last century and are comparable in terms of aspect and slope (flat area or
212 slope of 2-5% in all plots). The stands were near-natural, without a distinct

213 anthropogenic impact on their structure in the last three decades, and all tree ages were
214 over 100 years. All plots presented the same understory herbal and shrubs species
215 composition. A more detailed description of vegetation and soil at the different sites can
216 be found in Lucas-Borja et al. (2012; 2016). In each of the management units, three
217 plots covering an area of 9 m² (3 m x 3 m squares) were identified, to measure SHC and
218 SWR, and the physico-chemical soil properties (total of 36 plots). The distance between
219 plots was greater than 300 m, to consider the plots as independent.

220

221 *2.3. Measurement of soil properties and covers*

222

223 Soil properties were measured on three soil samples at each plot. Soil sampling
224 points were always more than three meters far from the nearest tree base. After
225 manually removing the litter, the samples were collected in the surface layer (at a depth
226 of 5 cm). This depth was chosen because (i) this is the initial interface for water-soil
227 interactions and (ii) the effects of tree species (through litter dynamics) should be
228 stronger compared to the deeper layers. Each sample consisted of six sub-samples, each
229 of 200 g, which were mixed to obtain a composite sample. After removing plant
230 residues for soil sampling, the samples were passed through a 2-mm sieve and then kept
231 at 3 °C prior to the analyses, which were carried out one week after soil sampling. Soil
232 texture (sand, silt, and clay contents in percentage, abbreviated as SaC, SiC, and ClC,
233 respectively) was analyzed using the method of Guitian and Carballas (1976). Bulk
234 density (BD) was calculated on triple samples per plot as the weight of soil in each
235 volume of a core extracted by a small cylinder at a depth of 20-30 cm. The electrical
236 conductivity (EC) and pH were measured using a Crison conductivimeter and pH meter,
237 respectively, in a 1/5 (w/v) aqueous extract. Soil organic matter (OM) was determined

238 by oxidation with K_2CrO_7 in an acid medium and titration of the excess dichromate with
239 $(NH_4)_2Fe(SO_4)_2$ (Yeomans & Bremner, 1989). Total nitrogen (TN) was measured using
240 Kjeldhal's method as modified by Bremner & Mulvaney (1982). Calcium carbonate
241 content of limestone ($CaCO_3$) was measured according to Della Porta, Kenter,
242 Bahamonde, Immenhauser, & Villa (2003).

243 In each plot, three longitudinal transects were identified to measure the percentage
244 of areal covers of vegetation (VC), dead matter (DW), rock (RC) and bare soil (BS)
245 (hereinafter "soil covers") using touch lengths in each transect, which was 3-m long.
246 Depth, area and weight were excluded from the field measurements of soil covers,
247 because litter characteristics are affected by seasonal variability as a response to other
248 external factors (such as the wind and runoff); this variability may alter the significance
249 of measurements. Moreover, the bare soil area may be considered as an indirect
250 indication of litter influence on the hydrological variables, since litter is absent where
251 the soil is bare.

252

253 2.4. *Measurement of SHC and SWR*

254

255 SHC and SWR were measured in three points, randomly selected for each plot,
256 using a Mini-Disk Infiltrometer (MDI, Decagon Devices, 2013) and the Water Drop
257 Penetration Test (WDPT) method (Letey, 1969; Woudt, 1959). The selected points were
258 always more than three meters of distance from the nearest tree base. These
259 measurement points were chosen at different distance from trees, in order to smooth the
260 influence of this parameter on SHC and SWR, which has been evidenced in some
261 studies (e.g., Madsen et al., 2008).

262 The MDI infiltrometer is commonly used for field measurements, given its small
263 size and easy handling (Robichaud, Lewis, & Ashmun, 2008). The MDI test is an
264 effective indirect technique to quickly assess the existence and degree of water
265 repellency of soil, since sorptivity and early infiltration rates are good indicators of
266 repellency (Robichaud et al., 2008; Lozano Baez et al., 2020; Williams et al., 2020).
267 However, we preferred to rely on direct measurements of SWR, using the WDPT
268 method, which is a simple but reliable procedure to measure SWR (Dekker et al., 2009;
269 Doerr, 1998).

270 In more detail, the SHC measurements were carried out according to the MDI
271 technical manual, and Robichaud et al., 2008a; 2008b). In summary, the litter layer on
272 the small area of soil surface was removed by a small shovel. This area was leveled to
273 prepare a horizontal surface for placing the infiltrometer. Then, the volume of water
274 infiltrated in the device was measured every 30 seconds for no less than 10 min. SHC
275 was estimated applying the equations proposed by Zhang (1997). Firstly, the measured
276 cumulative infiltration values (I , [m]) were regressed against the measured time
277 intervals (t , [s]) by equation (1):

278

$$279 \quad I = C_1 t + C_2 t^{1/2} \quad (1)$$

280

281 where:

282 - C_1 = coefficient related to soil hydraulic conductivity [m s^{-1}]

283 - C_2 = coefficient related to absorption capacity [$\text{m s}^{-1/2}$].

284 Then, the SHC (k , [mm h^{-1}]) was calculated by the following equation:

285

$$286 \quad k = C_1/A \quad (2)$$

287

288 A is a value corresponding to the Van Genuchten parameters (n and α) for a given soil
289 type to the suction rate (h_0 , equal in this study to -2 cm) and disk radius (2.25 cm) of the
290 infiltrometer. According to n , α and h_0 values for the experimental soils, the values of A
291 were equal to 6.64 (clay loam soil of PNUM and PN + QI plots) and 6.27 (loam soil of
292 PN + JT and PN + QI + JT plots) (Decagon Devices, 2013).

293 As regards the SWR determination in situ, WDPT measures the time that a drop
294 takes to completely infiltrate into soil. In this study, 15 drops of distilled water were
295 released, using a pipette, on the soil surface of a 1-m transect, to homogenize the
296 variable soil conditions; the time necessary for drops to infiltrate completely into the
297 soil was measured by a stopwatch. Before measurement, the litter cover was removed,
298 and the soil surface was cleaned using a brush.

299 SWR was classified according to the values of WDPT as Bisdom, Dekker, & Schoute
300 (1993):

- 301 i) non water-repellent or wettable soil (class 0, $WDPT < 5$ s);
- 302 ii) slightly water-repellent soil (class 1, $5 < WDPT < 60$ s);
- 303 iii) strongly water-repellent soil (class 2, $60 < WDPT < 600$ s);
- 304 iv) severely water-repellent soil (class 3, $600 < WDPT < 3600$ s); and
- 305 v) extremely water-repellent soil ($WDPT > 3600$ s) with three sub-classes (class 4, $1 <$
306 $WDPT < 3$ h, class 5 ($3 < WDPT < 6$ h) and class 6 ($WDPT > 6$ h).

307 The mean values of both SWR and SHC were calculated at the plot scale and used
308 for the statistical analyses. Finally, since SWR strictly depends on its water content
309 (SWC, e.g., Dekker and Ritsema, 1994; Lichner et al., 2013; Vogelmann et al., 2013;
310 Alagna et al., 2017), this property was measured simultaneously to SWR and SHC by a
311 device (Vegetronix VG400. accuracy of 2%, measurement range 0-50%), placed on the

312 soil surface and connected to a data logger (UX120 4-channel Analog Logger, Onset
313 HOBO, Massachusetts, USA).

314

315 *2.4. Statistical analyses*

316

317 The reciprocal relationships among the soil properties and covers (RC, VC, BS,
318 DW, SaC, SiC, ClC, BD, pH, EC, OM, TN, C:N and CaCO₃) as well as their influence
319 of SHC and SWR were evaluated adopting a combination of statistical techniques. First,
320 the statistical differences in SHC or SWR measurements were determined by the
321 multivariate permutational analysis of variance (PERMANOVA) (Anderson, 2005),
322 using the tree diversity level (PN+QI, PN+JT, PN+QI+JT and PNUM) as factors.
323 PERMANOVA tests the simultaneous response of one variable to one or more factors
324 in an experimental design based on any resemblance measure, using permutation
325 method. Before PERMANOVA, the soil properties and covers were $\log(x+1)$
326 transformed, whereas SHC and SWR data were square root transformed; the
327 resemblance matrix was built using the Euclidean and Bray Curtis distance for
328 environmental and SHC or SWR data, respectively. The sums of squares type were type
329 III (partial) and the four-level (tree diversity) factor was a fixed effect. The permutation
330 method used was the unrestricted permutation of raw data and the number of
331 permutations was 999. Then, an analysis of similarities (ANOSIM), described by Clarke
332 (1993), was carried out for the soil properties and covers, and the multivariate
333 resemblances were analyzed, according to the tree diversity level. Secondly, the Non-
334 metric Multi-Dimensional Scaling (NMDS) and the Kruskal stress formula (minimum
335 stress: 0.01) were applied to soil properties and covers, to evaluate the similarity level in
336 the individual cases of the dataset. Thirdly, a comparative (Mantel-type) test on

337 similarity matrices was developed, to assess the statistical relationships among soil
338 properties and covers on one side, and SHC or SWR on the other side. Fourthly, a
339 DISTLM function (distance-based linear modelling) was developed, to determine the
340 relative importance of any soil properties and covers on SHC and SWR variables. For
341 the DISTLM function, we developed “marginal” tests of the relationship between the
342 response variable (SWR or SHC) and an individual variable (soil property or cover), to
343 identify the independent variables that explain the variations in the soil samples.
344 Following the marginal tests, “sequential” tests of individual variables were performed
345 to assess whether adding an individual variable contributes significantly to the
346 explained variation of the response variable. Finally, distance-based redundancy
347 analysis (dbRDA) was applied to SHC and SWR, to build a regression model against
348 two new response variables (“axis” 1 and “axis” 2), built on the soil properties and
349 covers. The AICc (Akaike, 1974) criterion was adopted to select the best model and the
350 step-wise procedure was followed to build the model.

351 For the statistical analyses the software PRIMER V7[®] with PERMANOVA add-on
352 (Anderson, 2005) and Statgraphics Centurion XVI[®] (StatPoint Technologies, Inc.,
353 Warrenton, VA, USA) were used. A significance level of 0.05 was used, unless
354 otherwise indicated.

355

356 **3. Results**

357

358 Vegetation cover was the most developed in PNUM forest stands ($70.0 \pm 12.3\%$)
359 and lowest in PN + QI plots ($26.7 \pm 12.3\%$); PN + QI + JT and PN + JT plots showed a
360 vegetation cover of $30 \pm 15\%$ and $41.1 \pm 19.0\%$, respectively. Soil texture, although
361 being prevalently clay loam for PNUM and PN + QI plots and loamy for PN + JT and

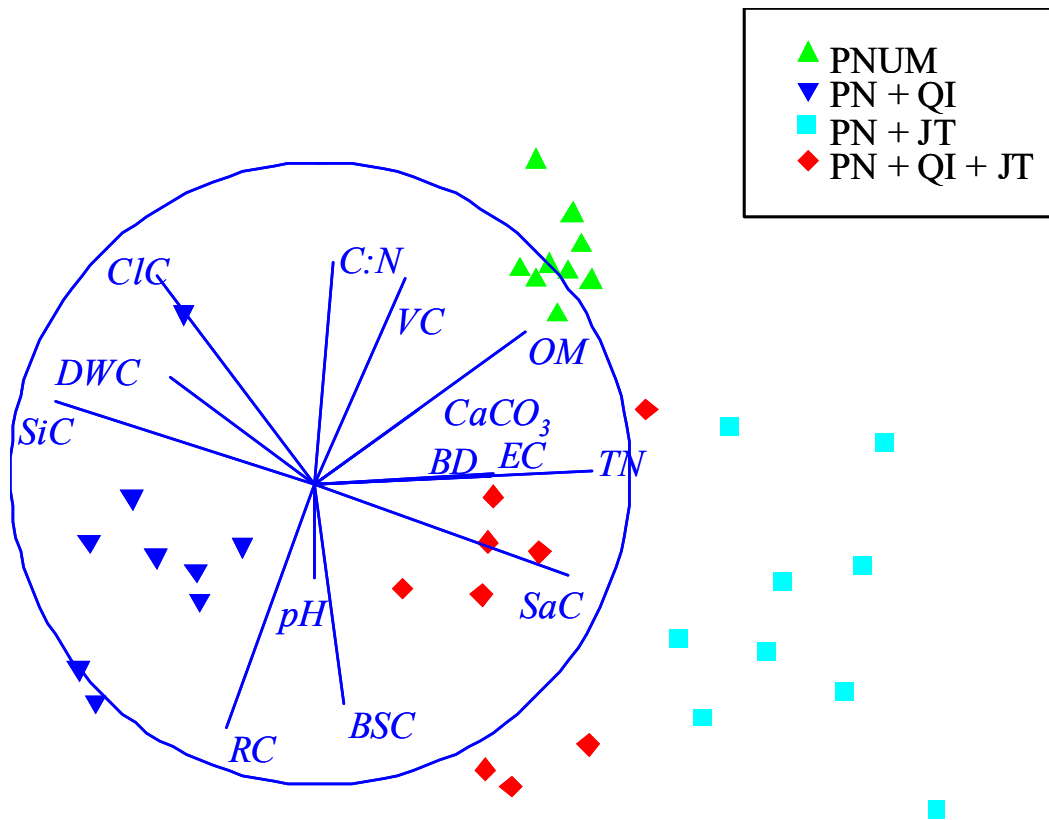
362 PN + QI + JT plots, showed significant differences, with higher sand content in PN + JT
363 and PN + QI + JT forests stands ($49.4 \pm 1.90\%$ and $45.0\% \pm 2.02\%$, respectively), while
364 the clay content was higher in PNUM and PN+QI soils ($31.0 \pm 1.63\%$ and $29.59 \pm$
365 2.02%). The latter forest soils showed the highest OM content ($24.5 \pm 0.58\%$) and the
366 PN + QI the lowest ($9.27 \pm 0.75\%$). The lowest TN content was detected PN + QI soils
367 ($0.38 \pm 0.04\%$) and the highest in PN + JT ($0.72 \pm 0.11\%$). Based on the values of OM
368 (converted to total carbon) and TN, the values of C:N were in the range 15.77 ± 1.34
369 (PN + JT stands) to 24.54 ± 0.75 (PNUM plots). The PN + QI + JT soils were more
370 compacted compared to the other forest stands, as shown by the lower BD (1.36 ± 0.01
371 g/cm^3 against values between $1.51 \pm 0.01 \text{ g/cm}^3$ $1.53 \pm 0.04 \text{ g/cm}^3$ and 1.70 ± 0.04
372 g/cm^3 for PNUM, PN + QI, and PN + JT, respectively). The soils were slightly acid
373 (PNUM, $\text{pH} = 6.31 \pm 0.54$) or close to the neutrality (pH between 7.10 ± 1.33 and 7.69
374 ± 0.43 , for the other stands). EC was in the range 0.17 ± 0.05 (PN + QI + JT) to $0.24 \pm$
375 0.02 (PN + JT) mmhos/cm. Finally, while in the forest stands the CaCO_3 content was
376 quite high ($4.67 \pm 0.59\%$, PNUM, and $4.84 \pm 0.47\%$, PN + QI + JT), this percentage
377 was 1.47 ± 0.35 in PN + QI forest plots (Table 1).

378 **Table 1** - Main physico-chemical properties and covers (mean \pm standard deviation) of soil sampled in plots under four tree diversity levels by
 379 pairwise test using Permutational multivariate analysis of variance (PERMANOVA) (Los Palancares y Agregados, Castilla-La Mancha, Spain).
 380

Soil properties/covers		FOREST STAND			
		PNUM	PN + QI	PN + JT	PN + QI + JT
Covers	<i>RC (%)</i>	0.00 \pm 0.00 a	14.44 \pm 8.82 ab	10.00 \pm 11.18 b	17.78 \pm 17.16 b
	<i>VC (%)</i>	70.00 \pm 12.25 c	26.67 \pm 12.25 a	41.11 \pm 19.00 b	30.00 \pm 15.00 ab
	<i>BSC (%)</i>	0.00 \pm 0.00 a	15.56 \pm 11.30 a	36.67 \pm 28.72 b	14.44 \pm 11.30 c
	<i>DWC (%)</i>	27.78 \pm 13.94 a	47.78 \pm 23.33 b	15.56 \pm 25.55 a	17.78 \pm 17.16 a
Texture	<i>SaC (%)</i>	36.93 \pm 1.59 b	27.27 \pm 1.71 a	49.44 \pm 1.90 d	44.98 \pm 2.02 c
	<i>SiC (%)</i>	31.38 \pm 1.70 b	42.59 \pm 2.52 c	26.49 \pm 2.01 a	29.32 \pm 3.37 b
	<i>CiC (%)</i>	31.04 \pm 1.63 c	29.59 \pm 2.02 c	22.20 \pm 1.97 a	26.41 \pm 1.89 b
Physical properties	<i>BD (g/cm³)</i>	1.53 \pm 0.04 b	1.51 \pm 0.01 b	1.70 \pm 0.04 c	1.36 \pm 0.01 a
	<i>pH (-)</i>	6.31 \pm 0.54 a	7.63 \pm 0.88 b	7.69 \pm 0.43 b	7.10 \pm 1.13 b
	<i>EC (mmhos/cm)</i>	0.21 \pm 0.03 ab	0.18 \pm 0.06 a	0.24 \pm 0.02 b	0.17 \pm 0.05 a
Chemical properties	<i>OM (%)</i>	24.49 \pm 0.58 d	9.27 \pm 0.78 a	20.36 \pm 0.90 c	14.82 \pm 1.18 b
	<i>TN (%)</i>	0.54 \pm 0.09 b	0.38 \pm 0.04 a	0.72 \pm 0.11 c	0.52 \pm 0.06 b
	<i>C:N (-)</i>	24.54 \pm 0.75 b	16.40 \pm 0.78 a	15.77 \pm 1.34 a	15.82 \pm 1.46 a
	<i>CaCO₃ (%)</i>	4.67 \pm 0.59c	1.47 \pm 0.35a	3.60 \pm 0.54 b	4.87 \pm 0.47 c

381 Note: different letters indicate statistically significant differences according to PERMANOVA tests (at $p < 0.05$). See the related list for the abbreviations.

382 In relation to the physico-chemical properties and cover of soils in the forest plots,
 383 ANOSIM (providing a global R of 0.94 and a significance level of 0.001) showed
 384 significant differences among all tree diversity levels. In accordance to the ANOSIM
 385 results, the NMDS clearly grouped the four forest stands in as many clusters (Figure 2),
 386 depending on the soil physico-chemical and cover variables. The importance of each
 387 property or cover of soil in clustering all tree level is shown by the loadings of each
 388 variable on the two axes, NMDS1 and NMDS2 (Table 2).
 389



390
 391 **Figure 2** - Biplot of physico-chemical properties and covers of soil samples collected in
 392 plots under four tree diversity levels using nonmetric multidimensional scaling (NMDS)
 393 routine (Los Palancares y Agregados, Castilla-La Mancha, Spain). The stress value for
 394 the NMDS is 0.09. See the related list for the abbreviations.
 395

396 In more detail, while the soil texture fractions (mainly SaC and SiC) as well as BD
 397 significantly weigh on the axis one (loadings over 0.80), most of the soil cover
 398 characteristics as well as OM and CIC have a large effect on axis two (loadings over
 399 0.65). This means that clusters of soils under the four forest stands are differentiated
 400 according to a combination of these variables and, in particular, the soil texture mainly
 401 on axis one, and the soil covers and many important chemical properties on axis two.

402

403 **Table 2** - Spearman correlations of physico-chemical properties and covers of soil
 404 samples collected in plots under four tree diversity levels using multidimensional
 405 scaling (MDS) routine (Los Palancares y Agregados, Castilla-La Mancha, Spain).

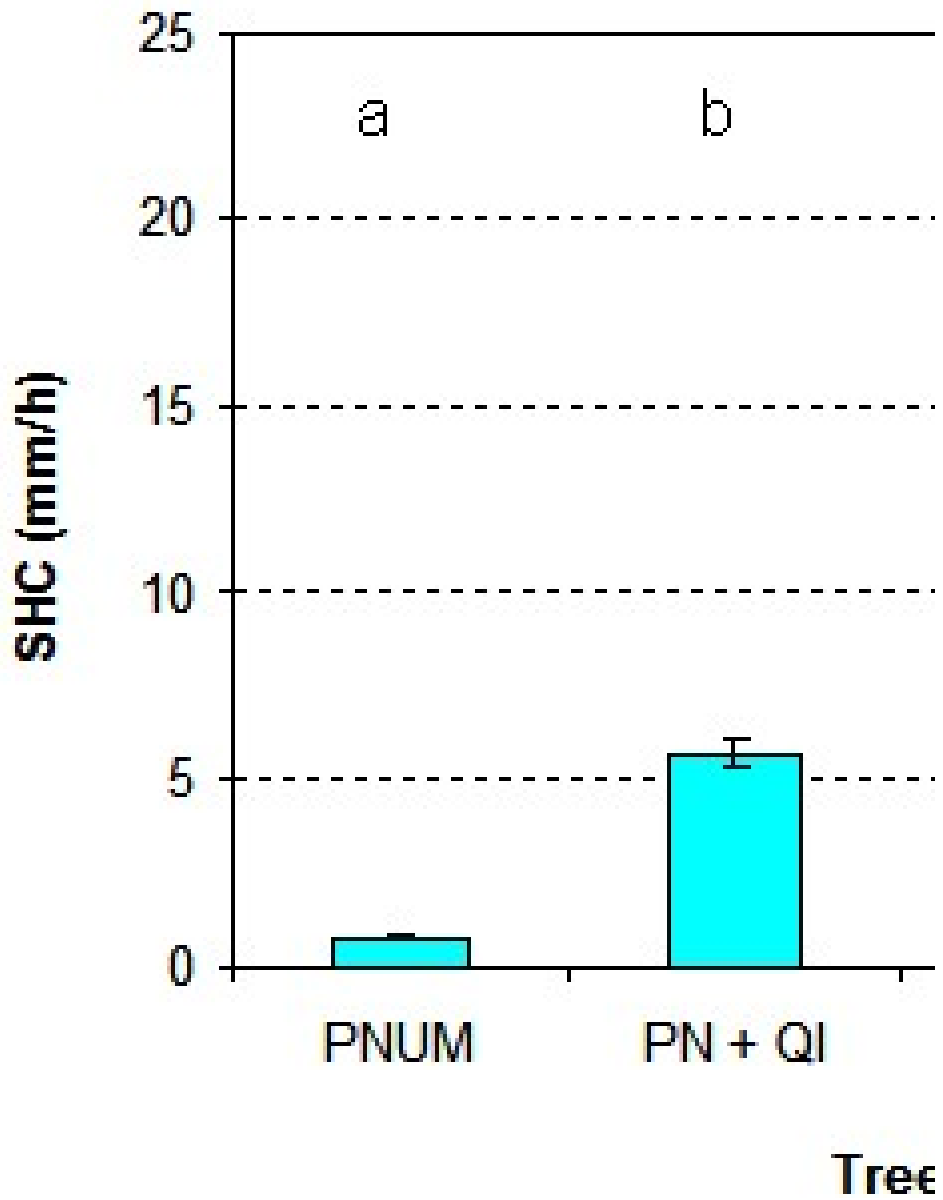
Soil property/cover	NMDS axes	
	axis 1	axis 2
RC	-0.285	-0.779
VC	0.292	0.666
BSC	0.097	-0.701
DWC	-0.459	0.347
SaC	0.818	-0.286
SiC	-0.829	0.272
CIC	-0.507	0.674
pH	0.005	-0.300
EC	0.567	0.026
OM	0.065	0.713
TN	0.334	0.239
C:N	0.576	0.041
CaCO ₃	-0.459	0.347
BD	0.900	0.050

406

Note: See the related list for the abbreviations.

407

408 The values of SHC were in the range 0.79 ± 0.04 mm/h (PNUM plots) to $11.4 \pm$
 409 10.2 mm/h (PN + JT) (Figure 3a), whereas WDPT was between 2.3 ± 0.5 s (PN + QI
 410 plots) and 106 ± 23.4 s (PN + JT) (Figure 3b). According to the SWR classification
 411 adopted in this study, PN + QI and PN + QI + JT soils were classified as “wetable”,
 412 PNUM soil as “slightly repellent” and PN + JT soils as “repellent”.



414

415 **Figure 3** - SHC (a) and SWR (b) (mean \pm standard deviation) in plots under four tree

416 diversity levels (Los Palancares y Agregados, Castilla-La Mancha, Spain). Different letters

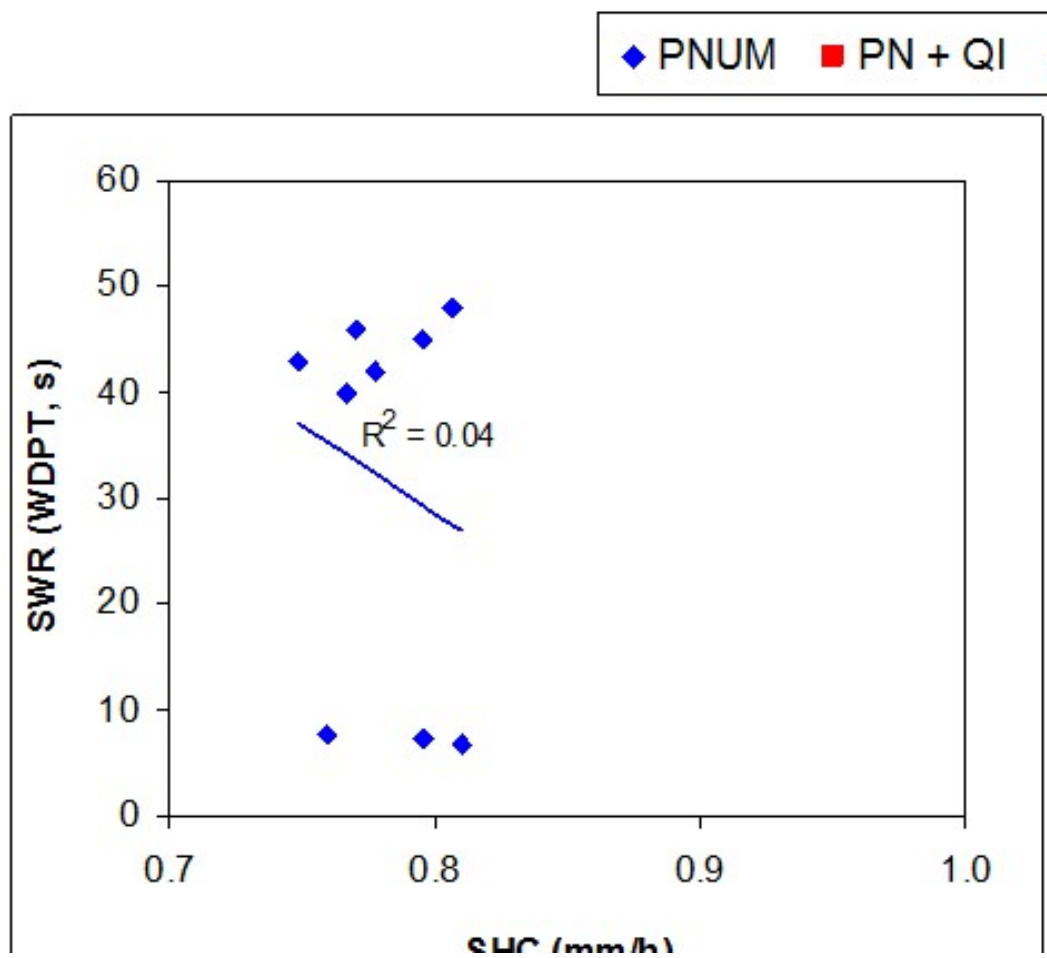
417 indicate significant differences at $p < 0.001$. See the related list for the abbreviations.

418

419

420 SHC and SWR were both significantly different (Pseudo-F = 47.9; P(perm) < 0.001
421 and Pseudo-F = 95.5; P(perm) < 0.001, respectively) among the tree diversity levels
422 according to the PERMANOVA routine. In relation to the pairwise comparisons among
423 tree diversity levels, for SHC, only the differences between PN + QI and PN + JT as
424 well as PN + JT and PN + QI + JT were not significant; for SWR, all couples of tree
425 diversity levels were significantly different, except the difference between PN + QI and
426 PN + QI + JT (non-significant at $p < 0.001$) (Figure 3). Furthermore, no significant
427 correlations were found between these hydrological variables for the studied forest
428 stands ($R^2 < 0.20$), except for PN + QI forest soil ($R^2 = 0.87$) (Figure 4).

429



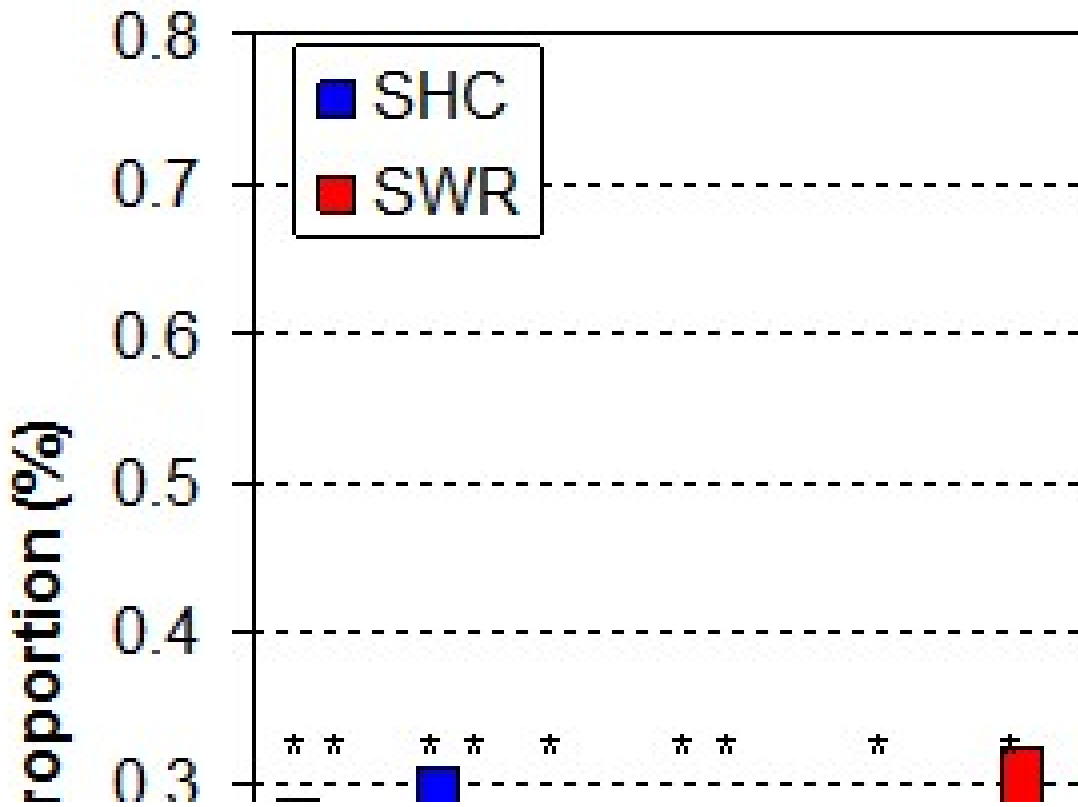
430

431 Figure 4 - Linear regressions between SHC and SWR in forest plots under four tree
432 diversity levels (Los Palancares y Agregados, Castilla-La Mancha, Spain). See the related
433 list for the abbreviations.

434

435 The comparative (Mantel-type) tests indicated that several physico-chemical
436 properties and covers of soil samples in plots under different tree diversity levels were
437 correlated both with SWR (Sample statistic (Rho): 0.21, significance level: 0.1%) and
438 SHC (Rho: 0.34; significance level: 0.1%). By applying the distance linear models
439 (DISTLM), the marginal tests revealed that all soil covers as well as CIC, pH and OM
440 significantly influenced SHC, while almost all the evaluated physico-chemical soil
441 properties and covers (except BSC and pH) affected SWR, when those variables were
442 considered as isolated (Figure 5). The sequential tests indicated that the best distance
443 linear model ($R^2 = 0.81$; AICs = 180.96) for predicting SHC consisted of C:N, CIC, BD,
444 OM and SaC (Table 3), while a much larger set of variables should be used in the best
445 distance linear model ($R^2 = 0.92$, AICs = 216.7) for predicting SWR (Table 4).

446



447

448 Figure 5 - Proportion of the variability of SWR or SHC explained by the selected soil
 449 property/cover under four tree diversity levels after the marginal tests using matched
 450 resemblance matrices (DISTLM) (Los Palancares y Agregados, Castilla-La Mancha,
 451 Spain). Notes: asterisks show the soil properties and covers that significantly influence SHC and SWR.
 452 See the related list for the abbreviations.

453 **Table 3** – Sequential tests of the relationship between the response variables (SWR or
 454 SHC) and soil properties after fitting one or more variables together under four tree
 455 diversity levels using matched resemblance matrices (DISTLM) (Los Palancares y
 456 Agregados, Castilla-La Mancha, Spain).
 457

Soil property/cover	P	Prop.	Cumul.
SHC			
+ C:N	0.001	0.709	0.709
+ ClC	0.061	0.027	0.736
+ BD	0.013	0.041	0.777
+ OM	0.057	0.233	0.800
+ SaC	0.029	0.027	0.827
- C:N	0.396	0.005	0.822
SWR			
+ RC	0.028	0.120	0.120
+ VC	0.075	0.076	0.196
+ BSC	0.221	0.035	0.231
+ DWD	0.013	0.118	0.349
+ SaC	0.035	0.085	0.434
+ SiC	0.185	0.032	0.466
+ ClC	0.002	0.168	0.634
+ pH	0.594	0.004	0.639
+ EC	0.240	0.019	0.658
+ OM	0.002	0.111	0.768
+ TN	0.646	0.003	0.771
+ C:N	0.888	0.001	0.771
+ CaCO ₃	0.090	0.027	0.799
+ BD	0.001	0.122	0.921

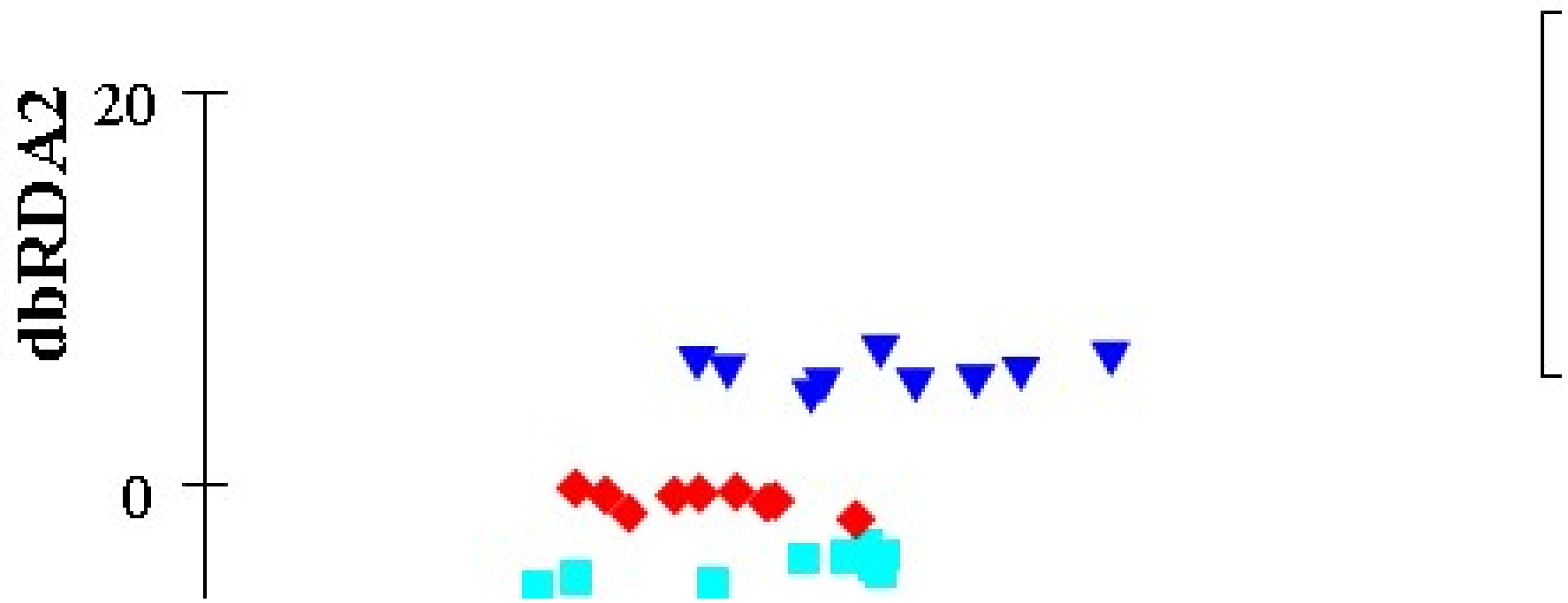
458 Notes: P = P value; Prop. = Proportion of the variability explained by the selected soil property/cover;
 459 Cumul. = Cumulative proportion variability explained by the selected soil property/cover; + indicates that
 460 the variable is added to the model, while - indicates a variable removed from the model. See the related
 461 list for the abbreviations.

462

463 According to the variations (out of the fitted model and out of the total variation)
 464 explained by the axes of dbRDA, axis one (dbRDA1) applied to SHC reflected 97.2%

465 of the fitted model and 78.7% of the total variation of the variables, whereas the axis
466 two (dbRDA2) explained 2.8% of the fitted model and 2.2% of the total variation
467 (Figure 6a). When dbRDA was applied to SWR, the axis one (dbRDA1) explained
468 94.8% of the fitted model and 87.3% of the total variation, whereas the axis two
469 (dbRDA2) explained 5.0% of the fitted model and 4.6% of the total variation (Figure
470 6b). CIC, SaC and OM content were the soil properties that most influenced these two
471 axes in the dbRDA model predicting SHC. Instead, all the evaluated soil properties and
472 covers influence the two axes in the SWR prediction model, although with variable
473 loadings (Figures 6a and 6b).

474 Finally, SWC was very similar among both the plots subjected to SWR
475 measurements, which furthermore showed a very low variability (due to the same
476 morphological conditions of the plots and physico-chemical properties and covers of
477 soils). The SWC were $0.132 \pm 0.04\%$ (PNUM), $0.152 \pm 0.004\%$ (PN + QI), $0.117 \pm$
478 0.004% (PN + JT) and $0.121 \pm 0.005\%$ (PN + QI + JT).





480

481 **Figure 6** - Biplots of dbRDA applied to SHC (a) and SWR (b) of soils under four tree diversity levels (Los Palancares y Agregados, Castilla-La

482 Mancha, Spain). See the See the related list for the abbreviations.

483

484 **4. Discussions**

485

486 It is well known how and to what extent the hydrology of the Mediterranean
487 forests strictly depends on infiltration capacity and surface conditions of soils (Di Prima
488 et al., 2020). These features are clearly driven first by the soil characteristics (e.g.,
489 texture and organic matter) (Bens et al., 2007; DeBano & Rice, 1973; Martínez-Zavala
490 & Jordán-López, 2009; Wahl et al., 2003), but also the interactions of soil
491 characteristics with plant composition can play a significant role. Recent laboratory
492 studies have shown that vegetation can influence soil water flow and thus SHC, by
493 inducing very low levels of water repellency (Lichner et al., 2007). Moreover, water-
494 repellent soils appear to be the rule rather than the exception in forest soil (Stoof et al., ,
495 2011).

496 The Mediterranean forest sites selected for our study are representative of *Pinus*
497 *nigra* Arn. ssp *salzmanni* monospecific and mixed forest stands of Cuenca Mountains.
498 Moreover, these selected forest stands had similar ecological, physiographical and
499 climatic conditions (same precipitation and temperature for all surveyed plots, Figure
500 S1). Thus, the influence of the tree diversity levels on SHC and SWR may be mainly
501 associated to other factors, such as soil and covers properties (e.g. mainly texture, but
502 also OM quantity), some of these factors being influenced by tree species composition.
503 In our study, SHC was the highest in PN + JT and PN + QI + JT forest stands and the
504 lowest in PNUM soils. According to literature, Madsen et al. (2008) demonstrated the
505 importance of *Juniperus* and *Pinus* vegetation in modifying soil unsaturated hydraulic
506 conductivity, whilerelatively high SHC under juniper trees has been reported by Wilcox
507 et al. (2003). Jarvis et al. (2013) and Olorunfemi & Fasinmirin (2017) reported that

508 SHC strongly depends on OM content of soil, which influences also bulk density and
509 aggregate size.

510 In the studied forest stands, the SHC of all forest stands decreases with
511 increasing clay contents ($R^2 = 0.78$, $p < 0.05$, data not shown) and decreasing sand
512 fractions ($R^2 = 0.45$, $p < 0.05$, data not shown). Moreover, the higher SHC in PN + JT
513 and PN + QI + JT stands compared to the PN + QI forest detected may be explained
514 primarily by the different soil texture, but also by the higher OM content of these soils
515 (which increases the stability of aggregates and therefore the soil macro-porosity, Chenu
516 et al., 2000; Devine et al., 2014), which in turn influence SWR. The reasons of the
517 lowest SHC detected in the PNUM soils (having the highest OM content among the
518 studied stands) is instead less clear. Presumably, the higher clay content of PNUM soils
519 compared to the other forest stands could have played a major influence on the low
520 SHC, but also the not negligible SWR could have played a role in decreasing the soil
521 infiltration (Wahl et al., 2003). The highest vegetation cover of PNUM soils among the
522 stands also suggests that the fine roots have a limited effect on infiltration.

523 In general, none of the studied forest stands showed a high level of repellency,
524 but only a slight (PNUM plots, dominated by pines) or a moderate hydrophobicity (PN
525 + JT soils) was evident, while forest soils with holm oaks were wettable. The soils with
526 higher OM (PNUM and PN + JT) are those showing significant SWR. This result is in
527 accordance with those of Lozano et al. (2013), who found higher SWR under *Pinus*
528 trees beside strongest persistence of SWR in *Quercus* forests, attributed these contrasts
529 to the significant differences between soils under different plant species, in particularly
530 for OM. Therefore, the differences in SWR can be related primarily to the soil OM
531 content (Cesarano et al., 2016; Esposito et al., 2014), and the latter is the main driver in
532 formation of SWR. In forest soils, the high SWR may be due to its high carbon content

533 in all the sieve fractions (both the finest and the coarsest fractions can be repellent) as
534 well as to the OM quality (Rodríguez-Alleres et al., 2007). Buczko et al. (2002) found
535 that soil OM and WDPT had positive linear correlations ($r = 0.73$), explained by the
536 higher proportion of mor-type humus and a greater thickness of the humose topsoil at
537 mixed beech stands compared to the pure pine sites. More in general, significant
538 positive relationships between SWR and soil OM were reported in literature (e.g.,
539 González-Romero et al., 2018; Martínez-Zavala & Jordán-López, 2009; Olorunfemi &
540 Fasinmirin, 2017; Plaza-Álvarez et al., 2018), particularly when the other soil physico-
541 chemical properties are similar (Chenu et al., 2000; Kajiura et al. 2012; Mataix-Solera
542 & Doerr, 2004; McKissock et al., 2002).

543 Past studies has showed that (i) soil OM, alone, cannot fully account for SWR
544 variability, and (ii) in some cases, the soil texture may be a factor with significant
545 influence on SWR level. In our study, besides OM content, the differences in soil
546 texture, detected among the studied forest stands, could have affected SWR, but these
547 soil properties seem to be less important compared to OM. For instance, soils samples
548 in PN + QI + JT stands showed a large content of sand but not SWR, while PN + JT
549 soils with a similar sand content were more repellent. It can not be excluded that soils
550 with significant sand and OM content are more susceptible to repellency.

551 If the hydraulic properties of soils are associated to the forest composition, we
552 observe that the monospecific stand showed a significantly lower SHC (see
553 PERMANOVA) together with a slight SWR. Water infiltration capacity of the soil was
554 instead noticeable in the other mixed forest stands (particularly in PN + JT and PN + QI
555 + JT). The mixed stands showed SHC over the mean value averaged among the tree
556 diversity levels, and this helps to reduce the negative impacts of surface runoff and
557 erosion. Moreover, SHC and SWR were not correlated, as confirmed by the low and

558 non-significant linear regressions. Instead, in some studies, a negative correlation
559 between SHC and SWR have been found (Olorunfemi & Fasinmirin, 2017). This lack
560 of correlations between the two hydrological parameters could be due to the different
561 but distinct influence of the studied soil properties and covers on SWR and SHC. For
562 instance, while many soil cover parameters clearly affect SWR, the same influence on
563 SHC was not as evident. Moreover, although the SHC and SWR were simultaneously
564 measured in the same or, at least, very close points, the different spatial variability of
565 these parameters can have played a role on their different dynamics. It is well known
566 how SWR is highly variable in space and time, even over the same soil moisture, and
567 this variability is difficult to explain (e.g., Pierson et al., 2008; Robichaud et al., 2008).
568 The comparative (Mantel-type) tests highlighted that several physico-chemical
569 properties and covers play individually a significant influence on SWR and SHC (in
570 some cases, on both of the studied hydrological parameters, such as three of the four
571 soil cover variables as well as CIC and OM contents of soil, according to the tree
572 diversity level). This result is in close accordance with those of the previous discussion,
573 showing that CIC and OM are key parameters in driving the hydrological characteristics
574 of a forest soils. However, the synergistic effects among these properties or soil covers
575 cannot be neglected, at least for SHC, since these effects reduced, or eliminated, the
576 significance of some individual parameters (e.g., the ratio C:N for explaining SHC).
577 This was shown by the multiregression model built using dbRDA, which was
578 particularly able to explain the variability of SHC or SWR among the different forest
579 stands. The dbRDA-assisted multiregression model explained high percentages (more
580 than 80% for SHC and 90% for SWR) of the total variation of the soil properties and
581 covers. By this model, silt and clay contents as well as OM of soils were the most
582 meaningful variables in predicting SHC for the different forests stands, while a much

583 larger set of variables must be used to differentiate the SWR among the studied forest
584 soils. Also, in the biplots related to dbRDA, clear clusters were evident, discriminating
585 forest soils with lower SHC (PNUM stands) from soils with the highest SHC (PN + QI,
586 PN + JT and PN + QI + JT stands) as well as repellent or slightly repellent soils (PN +
587 JT and PNUM stands) from non-repellent soils (PN + QI and PN + QI + JT stands). In
588 the study of Šimkovic et al. (2009), the results of multiple regression analysis showed
589 that SWR in topsoil material is significantly controlled by water and organic carbon
590 contents, while this study has shown that the differences among the forest stands are
591 determined by a more complex combination of physico-chemical properties and soil
592 covers. Regarding SHC, this analysis further confirms that particle size but also OM
593 content of soil (i.e., the variables with the large influence on axis 1) create a noticeable
594 gradient (and thus a clear difference) between PNUM and the other three forest stands
595 (having lower OM and clay contents compared to PNUM plots). Finally, the physico-
596 chemical properties of soils were found to exert a greater influence on the studied
597 hydrological properties of soils compared to the soil covers.

598

599 **5. Conclusions**

600

601 This study has shown that soil texture exerts a significant control on the hydraulic
602 conductivity of forests with different stand composition, and this influence can be
603 influenced by soil water repellency, where substantial organic matter is present. Soil
604 hydraulic conductivity was higher in managed stands of *Pinus nigra* Arn. ssp *salzmanii*
605 with *Juniperus thurifera* and *Quercus ilex* compared to unmanaged 100-120 years old
606 *P. nigra* stands. These differences in water infiltrability were due primarily to the soil
607 texture, but also to its organic matter content. None of the surface soils of the studied

608 forests showed a high level of water repellency. Only slight hydrophobicity (in
609 unmanaged plots, dominated by pines) or a moderate repellency (in *P. nigra* stands with
610 *J. thurifera*) were evident; yet, forest surface soils in *P. nigra* stands with *Q. ilex* were
611 wettable. Cross-stand differences in soil water repellency were primarily related to the
612 soil OM content and, secondarily, to texture. Finally, a robust and meaningful
613 multiregression model developed in this study and based on db-RDA, indicated that few
614 soil variables, specialised for each different forest stand, may be good predictors of soil
615 hydraulic conductivity; however, the water repellency level of study forest soils was
616 influenced by several variables.

617 Overall, the results of this study advance efforts to identify differences among key
618 parameters driving the hydrological response of Mediterranean forests according to their
619 biodiversity and soil characteristics. More research is certainly needed, in particular
620 about litter quality (only indirectly considered in this study within the physico-chemical
621 and covers of forest soils) and its feedback with water repellency, since litter quality
622 certainly may influence forest hydrology.

623

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625

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632

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