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1 Sugarcane yield response to legume intercropped: a meta-analysis.

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12 **Abstract**

13 Intercropping with legumes has attracted much attention worldwide, regarded as a sustainable
14 alternative to chemical N-fertilizer. Many studies have investigated the effects of introducing legumes
15 into sugarcane systems. However, how agro-environmental conditions influence the effect of legumes
16 on sugarcane yield is still unclear. Thus, we performed a meta-analysis to i) quantify the effect of legume
17 intercropping on sugarcane yield, ii) assess how agro-environmental and management conditions
18 influence the response of sugarcane yield to intercropping, and iii) assess the influence of legumes in
19 interaction with weeds on sugarcane yield. We combined data from a systematic literature review and
20 experimental data from a network of trials conducted in the tropical island of La Réunion. 315
21 observations (one-year x site x intercropping treatment) were analyzed with mixed effects models to
22 assess the effect of legume intercropping on sugarcane yield and factors influencing the response of
23 sugarcane yield. Intercropping with legumes resulted in lower sugarcane yields compared to the
24 monoculture treatment in 63% of cases. Our study highlighted how resource availability (e.g., annual
25 temperature, global radiation, soil organic carbon content) influenced sugarcane yield response to
26 legumes. Late sowing and early destruction date of the legume mitigated the negative effect of legumes

27 on sugarcane yield. Our study also showed that the negative impact on sugarcane yield increased over
28 years of intercropping, raising questions about legumes' actual contribution to soil fertility. Further
29 studies are needed to optimize legume sowing and destruction dates to find a trade-off between
30 expected services (N enrichment and weed control) and disservices (yield loss, costs, and labor time) in
31 sugarcane agrosystems according to the climate and soil context.

32 **Keywords:** Meta-analysis; Fabaceae; Cover crops; Crop association; *Saccharum officinarum*

33 **Highlights:**

- 34 ▪ Legume intercropping reduced sugarcane yield by an average of 3.34% worldwide.
- 35 ▪ Resource availability influenced sugarcane yield response to legumes.
- 36 ▪ Legumes sowing and destruction dates drove the competitiveness between species.
- 37 ▪ Legumes were twice less damaging than weeds for sugarcane.
- 38 ▪ The negative effect on yield increased with the number of years of intercropping

39 **1. Introduction**

40 Under the extensive use of chemical inputs in the 20th century, the intensification of agriculture has led
41 to the emergence of environmental issues such as soil degradation, water pollution, eutrophication,
42 greenhouse gas emissions, biodiversity loss, or weeds' resistance to herbicides (Bengtsson et al., 2005;
43 Heap, 2014; Howarth, 2008; Jie et al., 2002). The environmental cost of intensification raises the
44 question of the sustainability of this agricultural model (Sutton et al., 2013). Therefore, sustainable
45 cropping systems must be designed to ensure food security while improving resource use efficiency and
46 environmental durability (Bommarco et al., 2013).

47 Sugarcane is a worldwide crop cultivated for sugar, biofuel, and material for electric energy production
48 (Anthunes et al., 2019). The demand for ethanol and electricity will likely increase in the energy sector
49 as it could replace fossil fuels and reduce greenhouse gas emissions (Goldemberg et al., 2014; Leal et al.,
50 2013). Sugarcane is a semi-perennial crop that relies heavily on nitrogen (N) fertilization (FAO 2006).
51 However, the amount of N fertilizer taken up by the sugarcane is particularly low ranging from 10% to
52 40% of the total amount applied (Otto et al., 2016). As a result, concerns about the low N-use efficiency
53 and environmental impacts of N losses in these systems have been raised (Thorburn et al., 2017; Wang
54 et al., 2020). Therefore, there is a need to develop sustainable agricultural management to maintain
55 economically viable production levels of sugarcane with less reliance on chemical N-fertilizer input.

56 Legumes are a potential alternative to synthetic fertilizers thanks to their ability to fix atmospheric N.
57 Many studies have considered the positive effect of legumes as a rotation break crop in sugarcane
58 systems to minimize the requirements for N fertilization by the following plantations (Garside & Bell,
59 1999; Hemwong et al., 2009; Shoko et al., 2007). They showed that legumes could limit mineral N inputs
60 without decreasing sugarcane yield at planting. However, sugarcane is commonly planted every 7 to 15
61 years, limiting legumes' positive effects as a rotation break crop.

62 In recent years, many studies have been carried out to investigate the effect of cover crop intercropping,
63 such as legumes, in sugarcane systems. Besides enriching soils with N, legumes may compete with
64 weeds and help reduce herbicide use (Verret et al., 2017). However, excessive interspecific competition
65 can lead to a significant yield loss. As observed in other intercropping systems (e.g., in maize: Sileshi et

66 al., 2008; in maize, sorghum and pearl millet: Namatsheve et al.,2020), the impact of legumes on the
67 main crop sugarcane yield varies significantly from one study to another (Ambrosano et al., 2013; de
68 Resende et al., 2003; Nadeem et al., 2020; Tian et al., 2020). The impact of legume intercropping on
69 sugarcane yield could vary depending on climate (Waddington et al., 2007), soil (Xu et al., 2020),
70 management practices (e.g., crop species, fertilization, and irrigation regime, crop density, the time and
71 duration of cohabitation, Pelzer et al., 2014; Yu et al., 2016, 2015). The interactions in species mixtures
72 are complex with key issues regarding competition and the timing of N restitution to the main crop
73 (Justes et al., 2014). Apart from sugarcane and cover crops, a third component, the weed community,
74 increases the complexity of these multi-species interactions.

75 The diversity of the results observed in sugarcane yield response to legume grown as a cover crop in the
76 inter-row highlights the need for a comprehensive and quantitative analysis. To our knowledge, the
77 impact of climate, soil, and management practices variability on sugarcane yield response to legume
78 intercropping has never been analyzed in a quantitative study. The objectives of this study were i) to
79 quantify the effect of legume intercropping on sugarcane yield, ii) to assess how agro-environmental
80 and management conditions influence the response of sugarcane yield to intercropping, and iii) to
81 assess the influence of legumes in interaction with weeds on sugarcane yield. We performed a meta-
82 analysis by combining data from a literature review and experimental data from a network of trials
83 conducted in the tropical island of La Réunion.

84

85 **2. Materials and methods**

86 **2.1. Complementarity of the two data sets**

87 A meta-analysis was performed to assess the sugarcane yield response to legume intercropping
88 compared to sugarcane grown as a monocrop. The database included two complementary data sets. The
89 first data set was based on a systematic literature review (section 2.2, Table 1). This data set had the
90 advantage of covering a wider range of agro-pedo-climatic conditions but the disadvantage of having
91 less information on crop and legume management. The second data set included data from a network of

92 trials performed in the Island of La Réunion (section 2.3, Table 1). This data set had the advantage of
 93 providing more information than the first one (e.g., actual temperature and rainfall, and
 94 destruction/disappearance day of the cover crop, ground cover by legume and weeds).

95

96 **Table 1. Variables used in the meta-analysis from the two data sets. The number of observation (obs) or**
 97 **mean (minimum and maximum) values in the datasets are presented.** The characteristics of the climate
 98 types are shown in Table A1. DASP/H: Day after sugarcane planting or harvesting. NA: Not available for
 99 the data set.

Explanatory variables	Levels	Dataset 1: Literature	Dataset 2: Reunion
Climate	Desert	30 obs	
	Semi-arid	25 obs	
	Oceanic	48 obs	4 obs
	Subtropical	94 obs	
	Tropical	42 obs	72 obs
Altitude		479 m (10-1671)	103 m (69-545)
Mean annual temperature		23.3 °C (19-28.7)	24 °C (20.3-26)
Mean annual rainfall		1254 mm y ⁻¹ (200-2674)	1353 mm y ⁻¹ (718-2008)
Soil texture	Fine	78 obs	76 obs
	Medium	104 obs	
	Coarse	57 obs	
Soil organic carbon		10.6 % (3.5-33)	33.5 % (17-33.5)
Applied N fertilizer rate		174 kg N/ha (0-525)	168 kg N/ha (132-287)
Irrigated trial	Irrigated	177 obs	60 obs
	Rainfed	122 obs	16 obs
Legume species		21 species	5 species
Growth habit	Creeping	10 obs	10 obs
	Semi-erected	25 obs	6 obs
	Erected	105 obs	
	Semi-twining	82 obs	60 obs
	Twining	17 obs	
Legume sowing date		14.5 DASP/H (0-90)	51 DASP/H (0-119)
Legume destruction/ disappearance date		NA	282 DASP/H (125-365)
Width of the inter-row (m)		1.2 m (0.45-3.6)	1.5 m
Crop cycle	Plantation	203 obs	12 obs
	Ratoon	36 obs	64 obs
Number of years of IC		1,9 year (1-10)	2,8 years (1-6)
Type of trial	On-station	218 obs	63 obs
	On-farm	21 obs	13 obs
Global mean ground cover		NA	34 % (6-81)
Weeds mean ground cover		NA	10 % (1-45)
Legume mean ground cover		NA	35 % (1-65)

100

101 **2.2. Literature review**

102 A literature search was performed using the online database Scopus (Elsevier) in June 2021. The search
103 equation was (an asterisk being a replacement for any starting/ending of the respective term) : (legum*
104 OR faba* OR pulse* OR "cover crop" OR "green manure*" OR Arachis OR Cajanus OR Canavalia OR cicer
105 OR crotalaria OR glycine OR lens OR phaseolus OR Pisum OR vicia OR vigna) AND (*cane OR "Saccharum
106 officinarum") AND (intercrop* OR "crop association" OR "associated crops" OR "companion crop*" OR
107 "mixed crop*" OR mixtures OR "multiple crops*" OR companion crop*) in the title, abstract or
108 keywords. The database searches were increased with searches in library resources for relevant papers
109 from citations in retrieved papers.

110 Studies had to meet the following selection criteria to be included in the meta-analysis: (i) studies report
111 sugarcane yield from a field experiment in which a monoculture treatment (CT) was compared with at
112 least one legume-intercropped treatment (IC), (ii) the mean and number of replicates were directly
113 reported as numerical or graphical data, (iii) full-text articles were available. Data reported as an
114 average across years, locations, or factors were excluded. We did not include treatments with more than
115 one legume species (e.g., legume mixture or two species planted one after the other in the same crop
116 cycle). Studies were excluded if crop management practices other than intercrop practices differed
117 between CT and IC, except for weed management. If data from the same experiment were reported in
118 several papers, the data were included from the article that reported the data in the most significant
119 degree of detail. The literature search and the selection procedure were presented in a Prisma diagram
120 (Appendix A, Fig. A1).

121 A total of 31 papers fit our selection criteria for the meta-analysis (Table 2). The resulting database
122 included 239 observations (one-year x site x IC treatment) from the literature review.

123 **Table 2. Selected studies for the meta-analysis, from both data set: Reunion and Literature.** Details on references are available in Appendix A.

Reference	Country	Duration (years)	Legume cover crop species	Soil texture	Mean annual precipitation (mm)	Mean annual temperature (°C)
Trial 1	La Réunion (-20.903, 55.532)	2	<i>Vigna unguiculata</i>	Fine	1344	26.3
Trial 2	La Réunion (-20.903, 55.532)	6	<i>Canavalia ensiformis</i> , <i>Crotalaria juncea</i> , <i>Crotalaria spectabilis</i> , <i>Desmodium intortum</i> , <i>V. unguiculata</i>	Fine	1344	26.3
Trial 3	La Réunion (-20.903, 55.532)	3	<i>C. ensiformis</i>	Fine	1344	26.3
Trial 4	La Réunion (-20.903, 55.532)	5	<i>C. ensiformis</i> , <i>D. intortum</i> , <i>V. unguiculata</i>	Fine	1344	26.3
Trial 5	La Réunion (-20.903, 55.532)	1	<i>V. unguiculata</i>	Fine	1344	26.3
Trial 6	La Réunion (-21.047, 55.682)	3	<i>C. ensiformis</i> , <i>V. unguiculata</i>	Fine	1603	22.6
Trial 7	La Réunion (-21.067, 55.280)	1	<i>C. ensiformis</i> , <i>D. intortum</i>	Fine	816	20.1
de Souza Filho and de Andrade 1985	Brazil	3	<i>Phaseolus vulgaris</i>	Fine	1797	22
Parsons and Khubone 1999	South-Africa	1	<i>Arachis hypogaea</i> , <i>P. vulgaris</i>	Fine	1123	21.9
Gana and Busari 2001	Nigeria	2	<i>Glycine max</i> , <i>Sesbania rostrata</i> , <i>V. unguiculata</i> ,	Coarse	1075	28.4
Nazir et al., 2002	Pakistan	1	<i>Lens culinaris</i> , <i>Pisum sativum</i>	Coarse	346	24.2
de Resende et al., 2003	Brazil	2	<i>C. ensiformis</i> , <i>C. juncea</i> , <i>C. spectabilis</i> , <i>Mucuna pruriens</i>	Coarse	1354	23.5
Parsons 2003	South-Africa	1	<i>G. max</i> , <i>P. vulgaris</i> , <i>V. unguiculata</i>	Fine	1123	21.9
Saini et al., 2003	India	3	<i>P. sativum</i>	Coarse	769	23.9
Berry et al., 2009	South-Africa	1	<i>A. hypogaea</i> , <i>M. pruriens</i> , <i>Phaseolus lunatus</i>	Coarse	861	20
Manimaran et al., 2009	India	1	<i>Vigna mungo</i>	Coarse	1048	28.7
Rasool et al., 2011	Pakistan	1	<i>Cicer arietinum</i> , <i>L. culinaris</i>	Coarse	941	21.3
Prellwitz and Coelho 2011	Brazil	1	<i>C. juncea</i>	Fine	1073	24.4

Ambrosano et al., 2013	Brazil	1	<i>Cajanus cajan</i> , <i>C. ensiformis</i> , <i>Crotolaria brevifolia</i> , <i>C. juncea</i> , <i>Crotolaria mucronata</i> , <i>M. pruriens</i>	Fine	1255	20.8
Rehman et al., 2014	Pakistan	2	<i>C. arietinum</i> , <i>G. max</i>	Medium	346	24.2
Teshome et al., 2015	Ethiopia	1	<i>G. max</i>	Fine	200	28.7
Kaur et al., 2015	India	2	<i>P. sativum</i>	Coarse	1363	22.5
Córdova-Gamas et al., 2016	Mexico	2	<i>C. cajan</i> , <i>C. ensiformis</i>	Fine	1646	26.3
Khippal et al., 2016a	India	2	<i>Cyamopsis tetragonoloba</i> , <i>V. mungo</i> , <i>Vigna radiata</i> , <i>V. unguiculata</i>	Medium	772	24.1
Khippal et al., 2016b	India	2	<i>C. arietinum</i> , <i>L. culinaris</i> , <i>P. sativum</i>	Medium	766	24.4
Nadiger al., 2017	India	1	<i>G. max</i> , <i>P. sativum</i>	Medium	602	25.2
Pereira et al., 2017	Brazil	1	<i>C. cajan</i>	Fine	1341	20
He et al., 2018	China	2	<i>V. radiata</i>	Medium	1363	22.5
She et al., 2019	China	2	<i>A. hypogaea</i>	Medium	1727	22.2
Yasin et al., 2018	Pakistan	1	<i>L. culinaris</i>	Medium	346	24.2
Zarekar et al., 2018	India	1	<i>A. hypogaea</i> , <i>V. radiata</i>	Medium	2674	26
Salazar Cajas 2019	Australia	2, 1	<i>G. max</i>	Medium	1457	19.8
Solanki et al., 2019	China	1	<i>A. hypogaea</i> , <i>G. max</i>	Medium	1363	22.5
Geetha et al., 2019	India	1	<i>C. juncea</i> , <i>G. max</i> , <i>L. culinaris</i> , <i>V. unguiculata</i>	Coarse	618	26.3
Jhansi 2020	India	1	<i>A. hypogaea</i> , <i>C. juncea</i> , <i>C. tetragonoloba</i> , <i>G. max</i> , <i>V. mungo</i> , <i>V. radiata</i> , <i>V. unguiculata</i>	Medium	1108	28.2
Tian et al., 2020 ; Luo et al., 2016	China	10	<i>G. max</i>	Medium	1727	22.2
Nadeem et al., 2020	Pakistan	2	<i>L. culinaris</i>	Coarse	346	24.2

125 **2.3. Network trials in La Réunion**

126 *2.3.1. Study site*

127 A network of eight trials was carried out to assess the ability of legume cover crops intercropped with
128 sugarcane to control weeds in the inter-row and their impact on sugarcane yield. These trials were
129 managed by the sugarcane research center in Reunion Island (eRcane institute) on three sites in
130 Reunion Island from 2012 to 2019 (Table 2) as part of the CanécoH and AgriécoH projects. Most of the
131 trials were conducted in the north of the island on an experimental research station (Sainte-Marie, 69m
132 a.s.l). Two trials were also conducted on-farm in the east (Saint-Benoit, 150m a.s.l) and in the west of
133 the island (Saint-Paul, 545m asl).

134 The resulting database contained 76 observations (one-year x site x treatment) from the Reunion
135 network trials.

136 *2.3.2. Experimental design and crop management*

137 All trials compared one or several sugarcane-legume intercropping treatments (IC) to a control
138 treatment (CT) where sugarcane grown as a monoculture with chemical weeding. Depending on trials,
139 different legume species, legume sowing dates, and destruction dates have been tested. Treatments
140 were replicated two to four times with plots ranging from 60 to 780 m² depending on the trials. Details
141 on trial's experimental design and crop management are available in Table A2 to A5.

142 In all trials, sugarcane was planted with a 1.5 m inter-row. Legume was sown in the inter-row
143 mechanically or manually. Five legume species were tested (*Canavalia ensiformis*, *Crotalaria juncea*,
144 *Crotalaria spectabilis*, *Desmodium intortum*, *Vigna unguiculata*). The sowing date of the legumes
145 (expressed in day after planting - DASP or harvest - DASH) varied between trials and treatments. At the
146 end of their development, legumes were destroyed or left in the field, gradually disappearing thanks to
147 the sugarcane canopy closure.

148 Recommended non-limiting fertilizer rates were applied to sugarcane for all trials (Table A4). All trials
149 were irrigated, except for those in Saint-Benoît with enough annual rainfall. A pre-emergence herbicide
150 was applied after sugarcane planting or harvesting and before legume sowing. Afterward, weeding

151 interventions were performed when weeds reached 30% soil ground cover (chemically for CT and
152 manually for IC).

153 2.3.3. *Field measurements*

154 Ground cover by legume—was measured using a visual notation method used in previous studies
155 (Christina et al., 2021a) described in Table A6. Global ground cover by legume species and weeds,
156 considered as a whole, was measured monthly in the inter-row. Respective ground covers by legume
157 species and weeds were calculated by multiplying the global ground cover by the respective proportion
158 of each pool. The measure was extrapolated between two measurement dates to estimate a mean
159 ground cover over the sugarcane crop cycle. Sugarcane stalk fresh mass (hereafter sugarcane yield) was
160 measured at harvest by sampling all sugarcane stalks on the two rows in the middle of the plot (i.e., a
161 minimum of 30 m² areas).

162 2.4. Database

163 We compiled and harmonized the data from the literature and the Reunion network trials in a database.
164 We collected the means sugarcane yield of the control treatment (CT) and the intercropped treatment
165 (IC), their standard deviation, and replicate numbers. Standard deviations were calculated from
166 standard errors, coefficients of variation, 95% confidence interval, or least significant differences in
167 cases where they were not reported in the papers.

168 To explain the variability in sugarcane yield responses due to legume intercropping, we considered
169 several explanatory variables related to climate, soil, and management practices. The explanatory
170 variables used in the meta-analysis are shown in Table 1. Information for these variables was extracted
171 from the text, tables, or figures using a web-based image analysis, Webplotdigitizer v4.5 (Rohatgi, 2018).
172 When soil organic matter was reported, we converted it to soil organic carbon by dividing the value by
173 2 (Pribyl, 2010). If not reported, average annual rainfall and annual temperature data were retrieved
174 using the New_LocClim 1.10 database (Grieser, Gommès, and Bernardi 2006). The legume species were
175 classified according to their usual growth habit (creeping, erected, semi-erected, semi-twining, and
176 twining) as identified by the Useful Tropical Plants (<https://tropical.theferns.info/>) and the PROTA4U
177 (<https://www.prota4u.org/>) databases (Table A7).

178 Some variables had a few missing data (< 15%). We used single imputation method to include them in
179 the analysis (Scheffer 2002). The country's median was imputed when the applied N fertilizer rate for
180 sugarcane was missing (31/315 observations). The mean value according to soil texture was imputed
181 when the initial soil carbon content of the topsoil layer (%) was missing (46/315).

182 Some of the variables initially considered were omitted for the analysis of the literature data because
183 there were too many missing values (actual rainfall and temperature during the experiment, applied
184 NPK fertilizer for legumes, applied PK fertilizer for sugarcane, use of herbicide with IC, legume'
185 destruction/disappearance day of legume).

186 **2.5. Data analysis**

187 *2.5.1. Linear mixed model*

188 Three mixed-effect models were built to estimate the effect of legumes and explanatory variables on
189 sugarcane yield for each subset of the database:

- 190 - Model 1: Full data set (Literature + Reunion)
- 191 - Model 2: Literature data set
- 192 - Model 3: Reunion data set

193 As the observations from La Réunion were over-represented in the database, comparing models 1 and
194 2 allowed us to verify that these observations did not induce a bias in our analysis. The third model was
195 built to consider the variables not or not widely available in the literature. The approach used to build
196 these models is described in the following sections.

197 *2.5.2. Effect size*

198 The natural logarithm of the ratio of IC-to-CT sugarcane yields was used as the effect size in the meta-
199 analysis. The natural logarithm linearized the metric and provided a more normal sampling distribution
200 in small samples (Hedges, Gurevitch, and Curtis 1999). The results were back transformed and
201 presented as the percent change in sugarcane yield of the IC treatment compared to the CT treatment in
202 the figures and results.

203 *2.5.3. Weight of the observations*

204 In meta-analysis, observations were commonly weighted by the inverse of their variance (i.e.,
205 observations with low variance were considered more reliable and received a higher weight in the meta-
206 analysis). Unfortunately, this information was missing in many selected studies (86 obs from 8 papers).
207 In addition, some of the selected studies had a very low coefficient of variation (< 5%) compared to those
208 usually reported for sugarcane yields (Fig. A2). We doubted that the values reported in these articles
209 represent the existing variability in these trials. As a comparison, the average coefficient of variation of
210 the Reunion network trials was 11.91 ± 1.05 %. Therefore, we decided to weight observations using the
211 number of replicates to ensure robustness and statistical significance by keeping a sufficient sample
212 size.

213 *2.5.4. Preliminary analysis*

214 A first exploration showed that the full data set and the literature had a non-Gaussian residuals
215 distribution. To correct it, observations with a Cook distance greater than $4/n$ were considered suspect
216 and eliminated from the data set (with n =number of observations) (Viechtbauer and Cheung 2010). The
217 new full data set contained 281 observations (212 from the literature data set and 69 from the Reunion
218 data set). A new literature data set was extracted from it. Alone, the Reunion data set showed a Gaussian
219 residuals distribution and did not require a selection based on Cook distance. Finally, the full, literature
220 and Reunion data sets contained respectively 281, 212, and 76 observations. The residual diagnostics
221 before and after outlier removal were presented in Fig. A3. The observations from He et al. (2018) had
222 a unique behavior compared to other studies regarding nitrogen fertilization effect on the response
223 variable (Fig. A4). An indicator variable “He2018” was created to take it into account, which takes the
224 value 1 for this study and 0 for the others.

225 *2.5.5. Identification of the best random structure*

226 The best random structure was identified by comparing models with different random structures and
227 the same fixed structure with all variables included (model beyond the optimal). Models were fitted
228 using the restricted maximum likelihood (REML) estimation, and comparisons were made using
229 Akaike’s information criterion (AIC) (Zuur et al., 2009). The “trial” (ensemble of individual IC

230 observations from the same trial) and “trial_control” factors (ensemble of individual IC observations
231 with the same control treatment) were tested as random effects to account for the hierarchical
232 dependence between multiple observations within a study (Lajeunesse 2011). A first-order
233 autocorrelation structure AR(1) was also tested to account for repeated observations not being
234 independent over the years within one trial. The random structure minimizing the AIC, including only
235 the random effect “trial_control” was used in following analyses.

236 *2.5.6. Identification of the best-fixed structure*

237 We extended the models to determine the influence of explanatory variables (Table 1) on the yield
238 response ratio. The fixed effect selection was performed using a stepwise forward procedure based on
239 Akaike's information corrected criteria (AICc). During the selection, models were fitted using the
240 maximum likelihood (ML) estimation (Zuur et al., 2009). The best-fitted models were presented in Table
241 3.

242 Finally, the model parameters of the resulting model were fitted using REML estimation. The
243 assumptions of homogeneity and normality were checked graphically using histograms of residuals,
244 quantile-quantile plots of Pearson's residuals versus standardized gaussian sample, and plots of
245 residuals against fitted values, respectively (Fig. A5 to A7).

246 *2.5.7. Publication bias*

247 Publication bias was assessed by drawing funnel plots, in which standard errors were plotted against
248 the effect size log-ratio (Fig. A8, Duval and Tweedie 2000). Publication bias was tested on data with a
249 reliable coefficient of variation (>5%). Funnel plot asymmetry was tested with a rank correlation test
250 (Begg and Mazumdar 1994).

251 *2.5.8. Analyses*

252 All analyses were performed in R 4.1.1 (R Core Team, 2021) except the test of the autocorrelation
253 structure performed in SAS 9.3 (SAS Institute, Cary NC) using the PROC MIXED procedure. In R, linear
254 regressions with mixed-effects models were performed with the function lme() from the package nlme
255 (Pinheiro et al., 2021). For categorical variables, pairwise comparisons were performed with the

256 function lsmeans() from the package emmeans (Lenth et al., 2018). The rank correlation tests were
257 performed with the function ranktest() from the package metafor (Viechtbauer 2010).

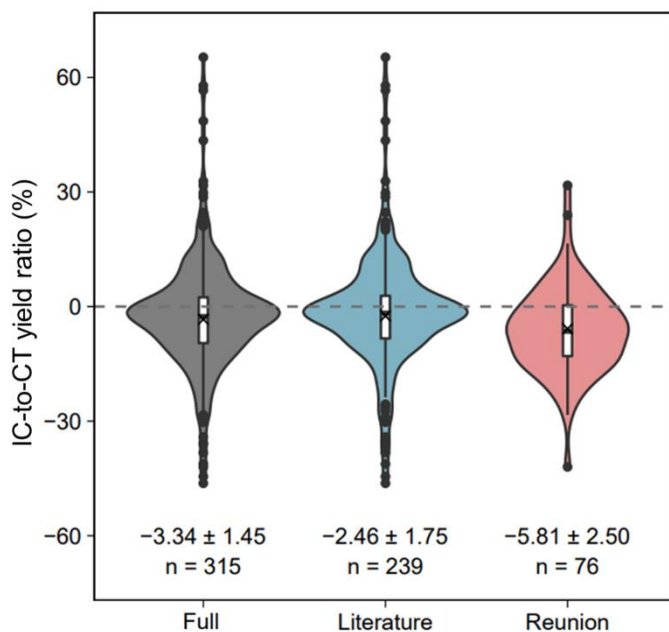
258 The data used in this study are available from the Dataverse repository in Viaud et al. (2022).

259

260 3. Results

261 3.1. Sugarcane yield response to intercrop treatment

262 The effect of legumes intercrops on sugarcane yields varied widely, ranging from a 65% increase to a
263 47% decrease compared to the monoculture treatment (Fig. 1). In the full data set, the mean IC-to-CT
264 yield ratio was -3.3 % (confidence interval (CI): [-1.9, -4.8]). In 22% of observations, legumes had a
265 strong depressive effect on sugarcane yield (IC-to-CT yield ratio < -10%). The mean IC-to-CT yield ratio
266 was lower in the Reunion data set (-5.8%, CI: [-3.3, -8.3]) than in the literature data set (-2.5%, CI: [-0.7,
267 -4.2]). Several outliers were found in the full data set with Cook's distances. When outliers were
268 removed, the mean IC-to-CT yield ratios were -2.2, -1.0, and -5.8 %, respectively, for the full, literature,
269 and Reunion data set (Appendix B, Fig. B1).



270

271 **Fig. 1. The IC-to-CT yield ratio depending on the data sets.** The violin plots represent the data
272 distribution. The horizontal lines within boxes indicate medians and the crosses indicated the mean.

273 The upper and lower edges of boxes indicate 75th and 25th percentiles, whiskers below and above the
 274 boxes indicate the 10th and 90th percentiles. Outliers are plotted as individual points. The mean value,
 275 confidence intervals (\pm) and the number of observations (n) are indicated below the violin plots.

276 3.2. Explanatory variables selected for the three models

277 All three models explained a significant proportion of the IC-to-CT yield ratio variability with conditional
 278 coefficient of determination values (R^2) of 0.74, 0.77 and 0.77 in model 1, 2 and 3, respectively (Table
 279 B1 to B3). Selected explanatory variables differed between models (Table 3).

280 **Table 3. Analysis of variance (ANOVA) of the random-effects models built to assess the influence of**
 281 **explanatory variables on the IC-to-CT yield ratio for the three data sets.** P = p-value, NA = not available for
 282 the data set, boxes left blank = variables not selected based on the AICc criteria.

Variables	Full Model 1	Literature Model 2	Reunion Model 3
Climate	P = 0,0002	P = 0,0057	
Mean annual temperature	P = 0,0650	P = 0,0001	
Climate x Temperature	P = 0,0001		
Mean annual rainfall			
Altitude	P = 0,0633		
Soil texture		P = 0,0762	NA
Soil organic carbon		P = 0,1383	P = 0,0074
Applied N fertilizer rate			
Irrigated trial			
Legume species			
Growth habit	P = 0,0798		
Sowing date of CC	P = 0,0728	P = 0,1093	
Destruction date of CC	NA	NA	P = 0,0071
Width of the interrow	P = 0,0799		NA
Crop cycle		P = 0,0831	
Number of years of IC	P = 0,0823	P = 0,1364	
Type of trial			
Global mean ground cover	NA	NA	
Weeds mean ground cover	NA	NA	P = 0,0098
Legume mean ground cover	NA	NA	P = 0,0206

283

284

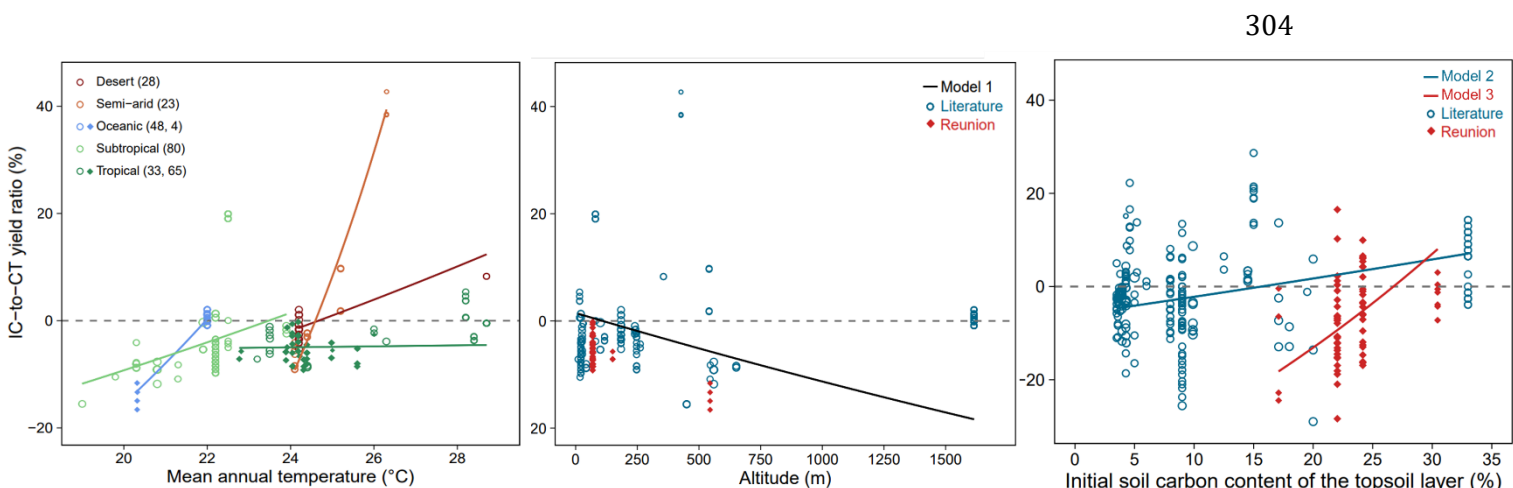
285 3.3. Climate and soil effects

286 The interaction of temperature and climate significantly influenced the IC-to-CT yield ratio in the full
 287 data set (Model 1, $F = 6.59$, $P = 0.0001$, Table 3). The IC-to-CT yield ratio increased with the temperature

288 in all climates except for tropical climate (Fig. 2a). The yield ratio response in desert (5 studies, 28 obs),
 289 semi-arid (4 studies, 23 obs), and oceanic climates (2 studies, 52 obs) should be interpreted carefully
 290 due to limited observations. In the subtropical climate, the IC-to-CT yield ratio increased up to 24°C.
 291 Above 24°C, in the tropical climate, the yield ratio did not change with temperatures. While the
 292 interaction between temperature and climate was not selected in the literature data set (Model 2, Table
 293 3), the temperature (Model 2, $F = 18.12$, $P = 0.0001$) and the climate (Model 2, $F = 3.95$, $P = 0.0057$)
 294 significantly influenced the IC-to-CT yield ratio. The effects of these variables were similar to the results
 295 obtained in the full data set (Fig. B2).

296 The IC-to-CT yield ratio significantly decreased with the altitude in the full data set (Model 1, $F = 3.5$, P
 297 $= 0.0633$, Table 3, Fig. 2b). On the contrary, the average rainfall was not a significant covariate. However,
 298 the IC-to-CT yield ratio increased with the mean annual rainfall in the rainfed trials, while it tended to
 299 decrease in the irrigated trials (Fig. B3).

300 The IC-to-CT yield ratio was also significantly influenced by the initial amount of C in the topsoil layer
 301 in both the literature (Model 2, $F = 2.24$, $P = 0.1383$, Table 3) and the Reunion data sets (Model 3, $F =$
 302 8.88 , $P = 0.0074$, Table 3). In both cases, the IC-to-CT yield ratio increased with the soil organic carbon
 303 (Fig. 2c). This increase was higher in the Reunion data set than in the Literature data set.



305
 306 **Fig. 2. The IC-to-CT yield ratio depending on (a) the mean annual temperature (°C) in interaction**
 307 **with the climate, (b) the trial altitude (m) and (c) the initial soil carbon content of the topsoil**
 308 **layer (%).** Lines in (a) represent the fitted random effects model regression in the model 1. Black, blue

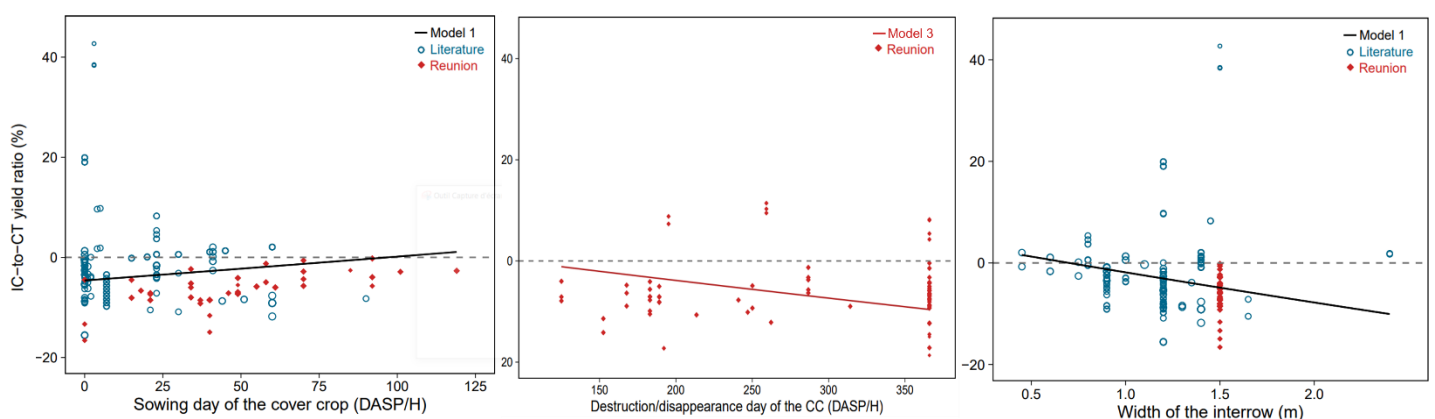
309 and red lines in (b) and (c) represent the fitted random-effects model regression in models 1, 2 et 3,
 310 respectively. The open circles and filled points represent the literature and Reunion predicted values,
 311 respectively.

312

313 3.4. Management effects

314 Depending on data sets the IC-to-CT ratio was significantly influenced by the legume sowing date, the
 315 destruction or disappearance date of the legume and the width of the sugarcane inter-row (Table 3).
 316 The sowing date of the legume was a significant covariate in the full data set (Model 1, $F = 3.26$, $P =$
 317 0.0728) and literature data set (Model 2, $F = 2.6$, $P = 0.1093$) but was not selected in the Reunion data
 318 set. In both cases, our results showed a strong negative effect of legumes on sugarcane yield for early
 319 sowing (Fig. 3a and Fig. B4). The destruction/disappearance date of the legume was a significant
 320 covariate in the Reunion data set (Model 3, $F = 7.87$, $P = 0.0071$) but was not available in the literature.
 321 The IC-to-CT yield ratio decreased with the destruction date (Fig. 3b). Finally, the IC-to-CT yield ratio
 322 decreased with an increase in the inter-row width in the full data set (Model 1, $F = 3.11$, $P = 0.0799$, Fig.
 323 3c).

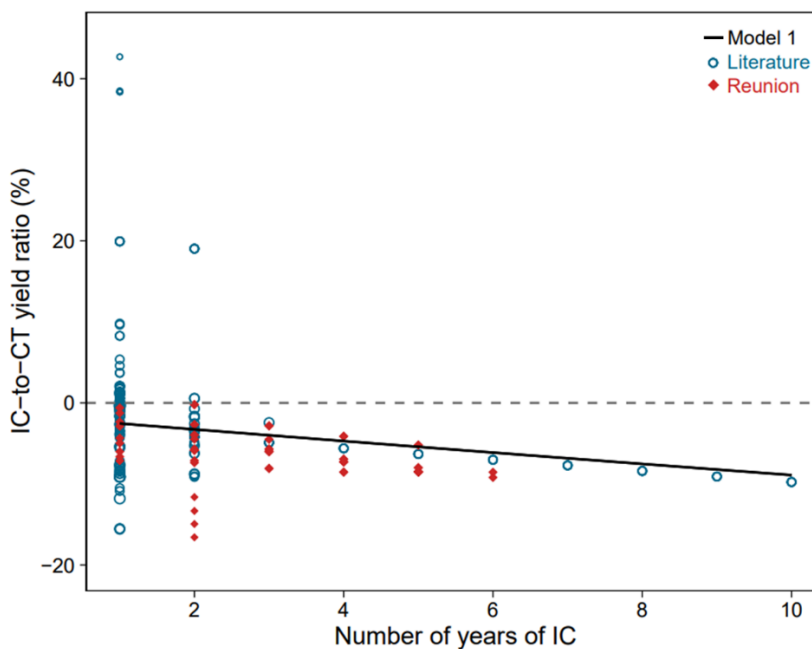
324 The legume species growth habit and the type of sugarcane cycle (plantation vs. ratoon crop) were
 325 selected in both the full data set (Model 1, $F = 2.13$, $P = 0.0798$) and the literature data set (Model 2, $F =$
 326 3.08 , $P = 0.0831$). However, the pairwise comparison conducted on these variables showed no
 327 significant difference between the different growth habits (Fig. B5), and between plantation and ratoon
 328 crop cycles (Fig. B6).



329 **Fig. 3. The IC-to-CT yield ratio depending on (a) the legume sowing date (in days after planting**
 330 **or harvest DASP/H), (b) the legume destruction or disappearance date (DASP/H) and (c) the**
 331 **width of the inter-row (m).** The black and red lines represent the fitted random-effects model
 332 regression of the model 1 and 3, respectively. The blue open circles and red filled points represent the
 333 literature and Reunion predicted values, respectively.

334 3.5. Effect of intercropping with legume over the years

335 The number of years of intercropping was a significant covariate in the full data set (Model 1, $F = 3.18$,
 336 $P = 0.0823$) and the literature data set (Model 2, $F = 2.27$, $P = 0.1364$) but it was not selected in the
 337 Reunion data set. In the full data set, the IC-to-CT ratio decreased by 0.7% per year of intercropping (Fig.
 338 4).



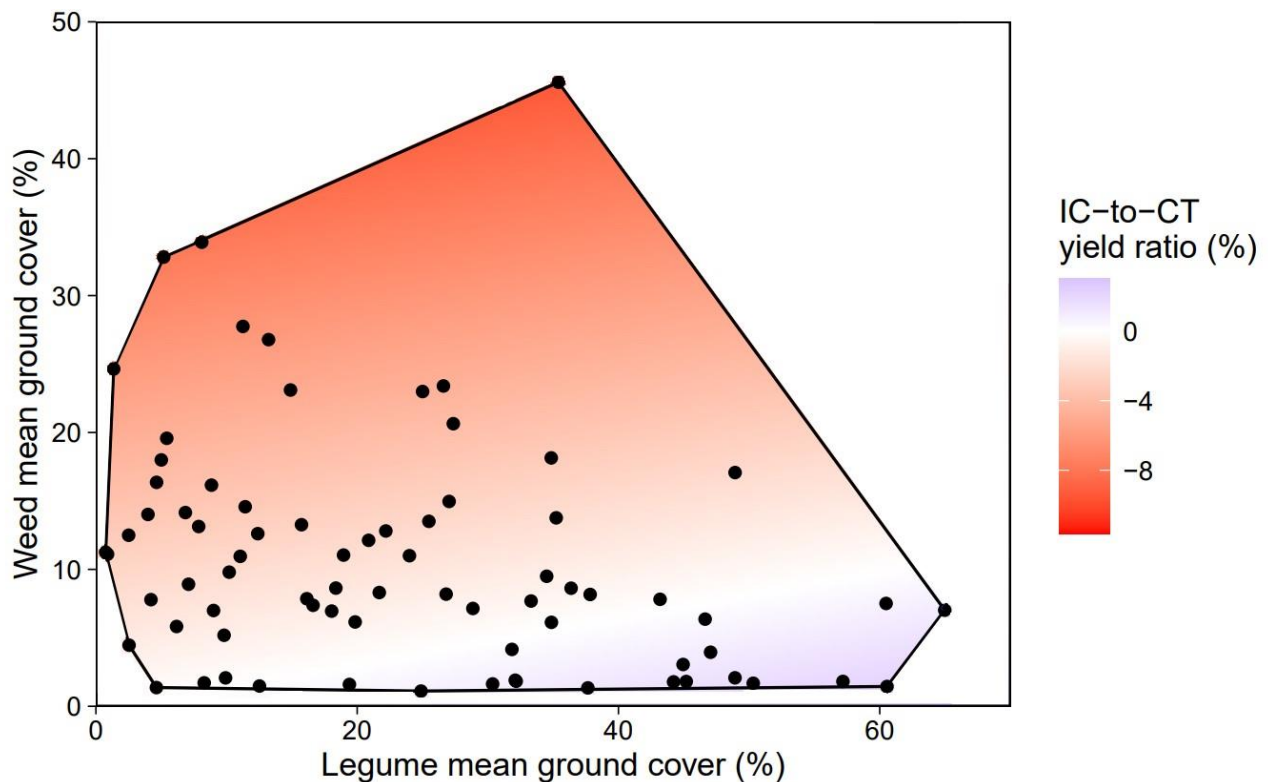
339

340 **Fig. 4. The IC-to-CT yield ratio depending on the number of years since the implementation of the**
 341 **IC treatment (years).** The black line represents the fitted random-effects model regression in the
 342 model 1. The blue open circles and red filled points represent the literature and Reunion predicted
 343 values, respectively.

344

345 **3.6. Legumes and weed effects on cane yield**

346 We used the Reunion data set to quantify the impact of legume and weed development on sugarcane
 347 yield (Table 3). The IC-to-CT yield ratio decreased with the global mean ground cover (Fig. B7). The
 348 analysis showed that weeds had an effect two times stronger than legumes on sugarcane yield when
 349 respective ground covers were used in the model. The IC-to-CT yield ratio decreased with weeds' mean
 350 ground cover (slope = -0.33, Model 3, $F = 7.19$, $P = 0.0098$), while it increased with legume mean ground
 351 cover (slope = 0.15, Model 3, $F = 5.71$, $P = 0.0206$, Fig. 5). Nonetheless, when using the global ground
 352 cover by both weeds and legume, the IC-to-CT yield ratio decreased with an increase in the global ground
 353 cover of the inter-row.



354

355 **Fig. 5. The IC-to-CT yield ratio (% , red to purple gradient) depending on the ground cover by**
 356 **legume (x-axis) and weeds (y-axis).** The IC-to-CT was predicted based on Model 3 (Reunion data set).
 357 The black line delimited the area of observed ground cover values in the Reunion data set. The red filled
 358 points represent the Reunion predicted values.

359

360

361

362 **4. Discussion**

363 **4.1. Variable effect of legume cover crops on sugarcane yield**

364 Our meta-analysis showed that legumes intercrops had on average a slightly negative effect on
365 sugarcane yield compared to the monoculture treatment. However, the effect varied widely among the
366 studies, negatively affecting sugarcane yield in 63% of cases. Such variability was not unexpected
367 considering the wide range of climate, soil, and management practices considered in this meta-analysis.
368 These results are similar to those obtained by Verret et al. (2017) in a worldwide meta-analysis on
369 intercropping systems with annual cash crops (mainly maize and straw cereals) and legumes. They
370 found a negative effect of legumes on cash crop yield in 56% of cases compared to the monoculture
371 weeded treatment. Similar to our results, the IC-to-CT yield ratio in Verret et al. (2017) varied from -49
372 to 317%.

373 The negative effect on sugarcane yield was larger in data issued from Reunion studies than in the
374 literature. There is no apparent reason related to the experimental conditions that could explain such a
375 difference. A publication bias may explain this difference and lead to underestimating the effect of
376 legumes on sugarcane yield in the literature. Publication bias may be due to unpublished studies with
377 negative results and the preferential publication of significant results compared to non-significant
378 results (Makowski et al., 2019). The symmetrical funnel plot suggested no publication bias in our study
379 (Fig. A8). However, this result must be considered cautiously because we could draw the funnel plot
380 only on a restricted part of the data (99/315), excluding data for which the variance was unknown and
381 data with a coefficient of variation <5%.

382 **4.2. Influence of resource availability in sugarcane-legume competition**

383 Identifying which factors influence sugarcane yield response was the second objective of the meta-
384 analysis. Our results showed that sugarcane yield varied with resource availability related to climatic
385 conditions, soil types, management conditions, and legume establishment.

386 The current study highlighted how legume competition was stronger at lower temperatures. Such
387 observation is explained by the fact that sugarcane is a tropical crop sensible to low temperatures
388 (Mongelard and Mimura, 1972). In addition, the ratio decreased with altitude, which is also associated
389 with a decrease in global radiation (Chopart, Mézino, and Nativel 2003). Temperature and global
390 radiation are key factors explaining variability in sugarcane yield on a regional scale (Christina et al.,
391 2021b; Marin and Carvalho 2012). Sugarcane competitiveness is stronger when environmental
392 conditions (e.g., temperature and global radiation) ensure favorable sugarcane development.

393 Our study also highlighted how light access by the legume was an important factor in the competitive
394 interactions between sugarcane and legume. An increase in the distance between sugarcane rows
395 induced a greater and longer light accessibility to the legume. Similar results have been shown with
396 other crops such as cotton, soybeans, corn, and rice, where low row spacing reduced weed density and
397 its impacts on yield (Bradley 2006; Chauhan and Johnson 2011; Tursun et al., 2016). However, row
398 spacing can be difficult to change as it responds to local technical constraints and can only be achieved
399 at planting time.

400 In this study, mean annual rainfall or irrigation did not significantly affect sugarcane-legume
401 competition. However, the sugarcane yield ratio tended to increase with rainfall in the non-irrigated
402 trials (Fig. B3). Several studies have shown that an increase in water availability reduces inter-species
403 competition (Daellenbach et al., 2005; Roy et al., 2015; Waddington et al., 2007). Our study also showed
404 that the ratio increased with soil fertility, even if no significant effect of the amount of nitrogen fertilizer
405 was found. These results are consistent with other meta-analyses on intercropping systems (Pelzer et
406 al., 2014; Xu et al., 2020), showing that the effect of nitrogen fertilizer is not significant but appeared to
407 be highly variable, stressing the need for deeper analysis.

408 **4.3. Trade-off in sugarcane-legume intercropping**

409 Our study highlighted legume's sowing and destruction date as key factors driving the competitiveness
410 of sugarcane and legume. An early sowing date of the cover crop resulted in a decrease in sugarcane
411 yield in the present study, supporting the assumption that sugarcane is highly sensitive to competition
412 during early growth. It can be assumed that the legume can over-compete sugarcane for light, water,

413 and nutrients when established early. Thus, delaying the sowing of the cover crop relative to the
414 sugarcane planting/harvest day should give the sugarcane a competitive advantage. These results are
415 in agreement with a meta-analysis on cereal-legume intercropping systems (Yu et al., 2016). Yu et al.,
416 (2016) also showed that this effect was more significant when the first sowed species was fertilized.
417 Although no interaction between sowing date and nitrogen fertilization was found in this study, this
418 topic appeared as an interesting avenue to explore in future research.

419 The cover crop's late destruction or disappearance date also negatively impacted sugarcane yield,
420 suggesting that legumes exert a competitive pressure even at the end of sugarcane's growth. Conversely,
421 early destruction of the cover crop may have facilitated sugarcane growth through an additional supply
422 of N from the mineralization of legume residues. To our knowledge, the effect of destruction date has
423 not been addressed in intercropping systems. However, the destruction date is a major issue in relay-
424 intercropping system, influencing cover crops' beneficial or detrimental effect on the following cash
425 crop (Keene et al., 2017; Mirsky et al., 2013). Alonso-Ayuso et al. (2014) showed that the destruction
426 date must be adapted according to the dynamic of mineralization (linked to the residues quality and
427 climate and soil conditions) to ensure synchronization between N release from cover crops and the cash
428 crop N requirement. These principles can be transposed to sugarcane-legume intercropping systems.
429 The destruction date must be considered in future work to maximize the potential benefits of legumes
430 as green manures. Destroying the legume during the sugarcane cycle involves additional costs and labor
431 time and can be technically complex when the sugarcane is well developed. A solution could be to use
432 legumes varieties with short growth cycles.

433 The expected benefits of intercropping with legumes are a gradual enrichment of soil N due to biological
434 nitrogen fixation (Cong et al., 2015), interspecific facilitation through N transfers during senescence and
435 decomposition of the different components of legumes (aerial biomass, roots and nodules) and from
436 root exudates to the non-legume species (Thilakarathna et al., 2016), and increased resource availability
437 (Latati et al., 2014). However, the amount of N fixed by the legume will depend on agro-environmental
438 conditions and the crop's growth. A late sowing date should benefit the sugarcane competitiveness and
439 productivity but at the expense of the legume's growth. One of the studies selected for this meta-analysis

440 observed that biomass and N uptake by legumes was three-fold higher for early sowing compared to
441 late sowing (Prellwitz and Coelho 2011). In addition, an early sowing dates can facilitate legume
442 establishment and enhance weed control (Mansuy et al., 2016). The destruction date must also allow
443 sufficient N accumulation to maximize expected legume benefits.

444 The timing of sowing and destruction of cover crops plays a crucial role in the performance of these
445 intercropping systems. Further work is therefore needed to optimize legume sowing and destruction
446 dates to find a trade-off between expected services (N enrichment and weed control) and disservices
447 (yield loss, costs, and labor time).

448 **4.4. Sustainability of sugarcane-legume intercropping**

449 We hypothesized that sugarcane yield might increase over time related to an increase in soil N fertility
450 thanks to legume-N fixation. However, our study showed that the IC-to-CT yield ratio decreased over
451 time. These results are in opposition to those reported by Li et al. (2021) in long-term maize-wheat and
452 maize-legume experiments. The authors attributed the increasing yield benefits of intercropping over
453 time to an increase in soil fertility in intercropping treatments. Nevertheless, very few studies have
454 investigated the effect of legumes on soil properties in sugarcane-legume intercropping systems. Higher
455 available NPK and microbial activity have been observed in short-term sugarcane experiments (Lian et
456 al., 2019; Solanki et al., 2019; Yadav and Yaduvanshi 2001). However, an increase of soil organic carbon
457 content has not been observed in a seven-year sugarcane intercropping experiment (e.g., in China, Wang
458 et al., 2020), while in other crops, many studies have shown that multi-species systems can enhance the
459 soil organic C and N content (Cong et al., 2015; Dybzinski et al., 2008). A few studies selected for this
460 meta-analysis measured the amount of N supplied via biological N-fixation, ranging from 13 to 60 kg N
461 ha⁻¹ (de Resende et al., 2003; Tian et al., 2020). Therefore, the fate of C and N of legumes in the soil-
462 plant system needs to be investigated in the short- and long-term.

463 To explain the decrease in IC-to-CT yield ratio over the years, a first hypothesis is that the negative
464 impact of legumes one year could be passed on onwards. Indeed, a negative effect on sugarcane growth
465 can reduce the belowground reserves at harvest and thus slow down the growth of the following cycle.
466 This hypothesis must be nuanced because many trials (mainly in Asia) in the literature replant

467 sugarcane yearly. However, we observed the same trend when restricting the data set to only the trials
468 that did not replant each year (Fig. B8). Additionally, weed control by cover crops is limited in
469 intercropping compared to chemical treatment (Verret et al., 2017). Therefore, a second hypothesis
470 could be an increase in weed pressure over time compared to conventional systems (observed in one
471 long-term trial in Reunion Island, Trial 2). Our analysis showed that the IC-to-CT yield ratio decreased
472 with the global mean ground cover (legumes and weeds). However, we also showed that weeds were
473 more damaging than legumes to sugarcane yield. This result can be attributed to lower competition for
474 N of the legume, due to their ability to fix N from the atmosphere. In addition, multi-species communities
475 like weeds have a higher potential competitiveness because species have a range of resource acquisition
476 strategies. Consequently, the sugarcane–legume–weeds interaction must be further investigated to
477 optimize the long-term sustainability of sugarcane-legume intercropping.

478 **5. Conclusion**

479 Despite an average 3.34% decrease in yield in the sugarcane-legume intercropping system compared to
480 monocropping, this meta-analysis supports the assumption that legumes are less damaging than weeds
481 for sugarcane, consolidating sugarcane-legume intercropping as a promising practice to reduce
482 herbicide use. The effect of legumes intercrops on sugarcane yield was significantly influenced by
483 environmental conditions (e.g., annual temperature, soil organic carbon content) and management
484 practices (e.g., sowing and destruction dates), allowing to suggest some management options to limit
485 the negative impact of legumes on sugarcane yield. However, there were currently insufficient elements
486 to reason the management practices regarding different environmental conditions. There is a need for
487 additional studies across various environments and situations to unlock the potential of sugarcane-
488 legume intercropping and to encourage the spread of this technique among farmers. Finally, very few
489 studies have quantified the amount of N provided by legume fixation. Further research should
490 investigate the long-term benefits on soil fertility of legumes in sugarcane systems.

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498 **Author contributions**

499 P. Viaud: Conceptualization, Methodology, Investigation, Formal analysis, Writing - Original Draft. B.
500 Heuclin & P. Letourmy: Methodology, Formal analysis, Visualization. M. Christina, A. Versini & K. Naudin:
501 Conceptualization, Methodology, Validation. A. Mansuy & J. Chetty: Conceptualization, Investigation. All
502 authors: Writing - Review & Editing.

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