

1 **Soil carbon sequestration by three perennial legume pastures is greater in deeper**
2 **soil layers than in the surface soil**

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21 **ABSTRACT**

22 Soil organic carbon (SOC) plays a vital role as both a sink for and source of
23 atmospheric carbon. Revegetation of degraded arable land in China is expected to
24 increase soil carbon sequestration, but the role of perennial legumes on soil carbon
25 stocks in semiarid areas has not been quantified. In this study, we assessed the effect
26 of alfalfa (*Medicago sativa* L.) and two locally-adapted forage legumes, bush clover
27 (*Lespedeza davurica* S.) and milk vetch (*Astragalus adsurgens* Pall.) on the SOC
28 concentration and SOC stock accumulated annually over a 2-m soil profile. The
29 results showed that the concentration of SOC in the bare soil decreased slightly over
30 the 7 years, while 7 years of legume growth substantially increased the concentration
31 of SOC over the 0-2.0 m soil depth. Over the 7-year growth period the SOC stocks
32 increased by 24.1 Mg C ha⁻¹, 19.9 Mg C ha⁻¹ and 14.6 Mg C ha⁻¹ under the alfalfa,
33 bush clover and milk vetch stands, respectively, and decreased by 4.2 Mg C ha⁻¹ in the
34 bare soil. The sequestration of SOC in the 1-2 m depth of the soil accounted for 79, 68
35 and 74 % of the SOC sequestered in the 2 m deep soil profile under alfalfa, bush
36 clover and milk vetch, respectively. Conversion of arable land to perennial legume
37 pasture resulted in a significant increase in SOC, particularly at soil depths below 1 m.

38 **Keywords:** Soil organic carbon, SOC stocks, SOC sequestration, perennial legumes
39 pasture.

40 **1. Introduction**

41 Concerns about global warming and increasing atmospheric greenhouse gas
42 concentrations (CO₂, CH₄, and N₂O) have led to questions on the role of soils as a
43 source or sink for carbon. Excluding carbonated rocks, soils constitute the largest
44 surface carbon pool, approximately 1500 Gt, equivalent to almost twice that in the
45 terrestrial biomass and three times that in the atmosphere (IPCC, 2000). Globally,
46 soil cultivation has resulted in the loss of more than 40 Pg C, at a rate of about 1.6 Pg
47 C year⁻¹, to the atmosphere during the 1990s (Smith, 2008). Chinese agricultural soils
48 have also lost 30-50% or more of the soil carbon pool (Lal, 2004a).

49 Soil organic carbon (SOC) is a significant component of the global carbon stocks
50 (Chen et al., 2008). Globally, 24% of the SOC stock has been lost through the
51 conversion of forest to cropland (He et al., 2011) and 59% through the conversion of
52 pasture to cropland (Zhu, 1994). Fortunately, the loss of SOC can be slowed down by
53 implementing crop management practices such as conservation tillage (Lal, 2004b;
54 Puget and Lal, 2005), converting degraded arable land to perennial grassland (Gentile
55 et al., 2005), using diverse rotations, and introducing legume and grass mixtures into
56 the rotation (Lal, 2002, 2004b, c).

57 In the USA, the revegetation of highly-erodible cropland or other environmentally-
58 sensitive areas to resource-conserving vegetation for a period of 10 to 15 years
59 increased the SOC content in the upper 3 m of soil at average rate of 1.1 Mg C ha⁻¹
60 year⁻¹ (Osborn, 1993). This conservation reserve program (CRP) also significantly
61 increased the soil C pool (Staben et al., 1997) and provided multiple benefits both
62 environmentally and economically (Munson et al., 2012; Wu and Lin, 2010). Like the

63 CRP program in USA, a program of soil and water conservation, namely “Grain for
64 Green” was implemented on the Loess Plateau of China in 1999 to alleviate land
65 degradation. The program of eco-environmental revegetation focused on the recovery
66 of damaged ecosystems (Wang et al., 2010) by the use of perennial vegetation to
67 control soil erosion, increase the stocks of SOC and prevent the occurrence of dry
68 layers in the loess soils (Fu et al., 2010). Alfalfa (*Medicago sativa* L.) has been widely
69 grown on the Loess Plateau to increase livestock production and improve water-use
70 efficiency and soil fertility through high forage production, and for its ability to
71 decrease soil erosion and fix atmospheric N (Guan et al., 2013). Additionally,
72 locally-adapted legume species such as bush clover (*Lespedeza davurica* S.) and milk
73 vetch (*Astragalus adsurgens* Pall.) have been widely grown as cover crops or
74 windbreaks to protect the soil from water and wind erosion in arid and semiarid
75 regions of northern China (Wang, 2003; Xu et al., 2006). The “Grain for Green”
76 program has reduced wind and water erosion of marginal arable land and is expected
77 to significantly contribute to soil C sequestration. Recent studies on the Loess Plateau
78 have investigated and estimated the changes in SOC stocks in the top 1 m of soil as a
79 result of revegetation of regional watersheds (Fu et al., 2010; Wang et al., 2011; Yan
80 et al., 2007; Zhang et al., 2013). However, deep-rooted perennial legumes may
81 penetrate deeper in the soil profile than 1 m, likely underestimating the SOC-
82 sequestration potential of these forage legumes in northwest China (Smith et al.,
83 2015).

84 The objective of this research was to assess the effect of alfalfa and two locally-
85 adapted forage legumes, bush clover and milk vetch, on the SOC concentration and
86 SOC stock accumulated annually for 7 years over a 2-m soil profile. The SOC content
87 in the 2-m soil profile was measured at the end of each growing season to quantify the
88 SOC concentration and stock under the locally-adapted forage legumes and alfalfa,
89 and to provide specific information for estimating the SOC sequestration potential of
90 an important agricultural area. The hypothesis tested was that long-term growth of
91 deep-rooted perennial legumes will increase soil organic C, provide a feed source for
92 animals and provide a sink for atmospheric carbon.

93 **2. Materials and methods**

94 **2.1 Experimental site description**

95 The experiment was conducted at the Changwu Agro-ecological Experiment Station
96 on the Loess Plateau (35°12'N, 107°40'E), Shaanxi province, China, from 2004 to
97 2010. The level site is located at 1220 m above sea level. The climate is semiarid,
98 with an annual mean temperature of 9.1 °C and a mean annual precipitation of 579
99 mm (1979-2003) with rainfall concentrated in the period from July to September.
100 Precipitation and temperature data were recorded at the Changwu Meteorological
101 Station, 20 m from the experimental site. The groundwater table is 50 – 80 m below
102 the soil surface, making it unavailable for plant growth. Prior to the establishment of
103 this experiment, the site was planted to winter wheat for many (at least 20) years. For
104 winter wheat production, the site was ploughed to a depth of 0.3 m twice a year, after

105 harvest in early July and again in September before sowing; only wheat stubble was
106 returned to the soil, but 108 kg N ha⁻¹ and 276 kg P₂O₅ ha⁻¹ of fertilizer was applied
107 each year before sowing. In 2003, after the winter wheat was harvested, the site
108 remained fallow for 280 days to allow moisture accumulation over the winter before
109 the legumes were sown in May 2004.

110 Soil at the experimental site belongs to the Loess series. The texture in the top 5 m is
111 a uniform silty clay loam (haplic greyxems, FAO-UNESCO, 1988), with a mean sand,
112 silt, and clay content of 3.5, 65.6, and 30.9%, respectively. The soil physical
113 characteristics do not significantly change in the upper 5 m. The measured average
114 bulk density of the soil in the upper 2 m is 1.31 g cm⁻³, does not change with depth,
115 and the top 0.3 m contained 1.55% total organic matter, 0.106% nitrogen, and 0.095%
116 available phosphate prior to the commencement of the experiment in 2004.

117 **2.2 Treatments and measurement of aboveground biomass**

118 Twelve experiment plots, each 4 m by 3 m, were established in early May 2004 with
119 one of three forage legume species, milk vetch (*Astragalus adsurgens* Pall.), alfalfa
120 (*Medicago sativa* L.) and bush clover (*Lespedeza davurica* S.), and an unplanted
121 control. Each legume species was grown as an evenly-spaced monoculture (but
122 assumed a more patchy distribution with time) at a seeding density of 25 plants m⁻²,
123 weeds were removed from all plots by hand. The plots were adjacent to each other.
124 During the experimental period from 2004 to 2010, there was no irrigation or other
125 form of supplementary water and the plots were not fertilized. The plants were not

126 inoculated, but relied on the naturally-occurring root nodule bacteria from previous
127 growth of the three species on the experimental station. Lack of nodulation was not
128 observed to be a problem. Treatments were completely randomized in three replicate
129 blocks.

130 Each year from 2005-2010, measurements of aboveground biomass production for
131 each legume were taken at the end May, July and September (in 2004 only one cut
132 was made in September) by cutting the plants at ground level with hand-held shears in
133 a randomly-selected 1 m × 1 m quadrat within each plot, but avoiding border areas. At
134 the same time, the rest of the plot was also cut at the same height and the forage
135 removed. The oven-dry weight was determined after drying at 105 °C for 0.5 h and
136 then further dried at 75 °C for 48 h (Guan et al., 2013).

137 **2.3 Soil sampling and analysis**

138 Soil samples were taken with a cylindrical steel corer (diameter 40 mm and height
139 200 mm) at two random positions in each plot which were combined into one
140 composite sample per plot before analysis. Each plot was sampled from the surface to
141 2 m deep at depths of 0-0.3, 0.3-0.6, 0.6-1.0, 1.0-1.5 and 1.5-2.0 m before sowing on
142 10 May 2004 and at the end of each growing season (29 October) from 2004 to 2010.
143 The soil samples were air-dried, roots and organic debris removed, ground and sieved
144 through a 2 mm sieve, then stored at room temperature before analyzing the SOC.

145 The concentration of SOC (in g kg⁻¹) was measured using the wet dichromate
146 oxidation procedure (Moinuddin and Khanna-Chopra, 2004). Briefly, a 0.5 g soil

147 sample was digested with 5 mL of 1N $K_2Cr_2O_7$ and 5 mL of concentrated H_2SO_4 at
148 $150^\circ C$ for 0.5 h, followed by titration of the digest with standardized $FeSO_4$.

149 **2.4 SOC stock calculation and statistical analyses**

150 Soil organic C stock was calculated as Eq (1):

$$151 \quad C_{\text{stock}} = SOC \times \rho \times H \times 10 \quad (1)$$

152 where SOC is the SOC concentration ($g\ kg^{-1}$) in each soil layer, ρ is the bulk density
153 ($g\ cm^{-3}$) of the soil, and H is the depth of each layer.

154 The data were analyzed by analysis of variance (ANOVA), and means were compared
155 using the LSD at $P < 0.05$ to characterize the differences among treatments. PROC
156 GLM (General Linear Model) in the Statistical Analysis System (SAS Institute, Cary,
157 NC, version 8.02) software was used to assess the temporal changes in SOC stock and
158 the rate and amount of SOC sequestered.

159 **3. Results**

160 **3.1 Meteorological conditions**

161 The average monthly mean temperature from June to August, the primary growth
162 period for the legumes, was about $20^\circ C$. Monthly mean temperatures were about $1^\circ C$
163 warmer than the long-term mean throughout the experimental period (Fig. 1). Over
164 the experimental period, the total annual precipitation varied from 470 mm in 2006 to
165 583 mm in 2010, and was below the long-term mean in all years except 2010 when
166 the rainfall was similar to the long-term mean (Fig. 1). Rainfall from July to

167 September accounted for 55-60% of total annual precipitation, while rainfall in the
168 legume-growing season (from April to October) was about 90% (range from 84% in
169 2009 to 96% in 2005) of the total annual precipitation (Fig. 1).

170 **3.2 Aboveground forage biomass production**

171 The results of the aboveground biomass production over the seven years have been
172 reported by Guan et al. (2013). Briefly, the annual production of milk vetch increased
173 from 2.2 t ha⁻¹ in the first year to 14.3 t ha⁻¹ in 2006 and then decreased, alfalfa
174 increased from 2.3 t ha⁻¹ in the first year to a maximum of 22.2 t ha⁻¹ in 2006 and then
175 decreased, while bush clover increased from 0.2 t ha⁻¹ in the first year to 7.8 t ha⁻¹ in
176 2009 and did not decrease significantly thereafter (Table 1). Total aboveground
177 biomass production over the experimental period was highest in alfalfa at 91 t ha⁻¹
178 (equivalent to 43 Mg C ha⁻¹ assuming a C to dry weight ratio of 0.475, Magnussen
179 and Reed, 2004) compared to 56 t ha⁻¹ (27 Mg C ha⁻¹) in milk vetch and 42 t ha⁻¹ (20
180 Mg C ha⁻¹) in bush clover (Table 1).

181 **3.3 SOC concentration over the soil profile**

182 The legumes significantly ($P < 0.001$) increased the SOC concentration at each soil
183 depth, and this effect varied with legume species and experimental year (Table 2).
184 The initial concentration of SOC in May 2004 decreased with increasing soil depth
185 (Fig. 2). In the upper 0-0.3 m of soil, the initial SOC concentration was 8.0 ± 0.03 g kg⁻¹
186 ¹, while it was only 3.3 ± 0.27 g kg⁻¹ in the 1.5-2.0 m soil layer (Fig. 2). Comparison of
187 the SOC concentration between the initial values on 10 May 2004 and those at the end

188 of the experimental period in October 2010 showed that the concentration of SOC in
189 the bare soil decreased slightly over the 7 years, while 7 years of legume growth
190 substantially increased the concentration of SOC over whole 2 m soil depth. There
191 were large increases in the concentration of SOC at 0.6-1.0 m, 1.0-1.5 m and 1.5-2.0
192 m soil depth and a small, but significant, increase in the upper 0.3 m of the soil in
193 bush clover, but not in milk vetch and alfalfa. No significant changes were observed
194 after 7 years at the 0.3-0.6 m depth (Fig. 2).

195 **3.4 SOC stock over the experimental period**

196 SOC stock was calculated by converting SOC concentration to the amount of SOC
197 per soil layer per unit area. The SOC stock in 2004 varied from $21.4 \pm 0.85 \text{ Mg C ha}^{-1}$
198 in the 0.3-0.6 m soil layer to $32 \pm 0.14 \text{ Mg C ha}^{-1}$ and $32 \pm 0.68 \text{ Mg C ha}^{-1}$ at 0-0.3 and
199 1.0-1.5 m depth respectively (Fig.3). In the bare soil, the SOC stock decreased at all
200 depths across the experimental period, but only decreased significantly at -0.36 Mg C
201 $\text{ha}^{-1} \text{ year}^{-1}$ ($P < 0.05$) in the 1.5-2.0 m layer (Fig. 3d), presumably from the decay and
202 turnover of the wheat roots accumulated over the many years of wheat production
203 prior to the planting of the legumes. In the legume plots, the SOC stock increased
204 linearly with time (2004-2010) in the 0-0.3 m, 0.6-1.0 m, 1.0-1.5 m and 1.5-2.0 m soil
205 layers, but not in the 0.3-0.6 m soil layer (Fig. 3). The change in SOC stock over the 7
206 years was greatest at soil depths below 1.0 m in all three species and was greatest in
207 the alfalfa plots with rates of $1.35 \text{ Mg C ha}^{-1} \text{ year}^{-1}$ at a depth of 1.0-1.5 m ($P < 0.001$)
208 (Fig. 3d), and $1.39 \text{ Mg C ha}^{-1} \text{ year}^{-1}$ at a depth of 1.5-2.0 m ($P < 0.001$) (Fig. 3e). The

209 highest accumulation of SOC stock occurred at a depth of 1.0-1.5 m in bush clover
210 where it averaged $1.58 \text{ Mg C ha}^{-1} \text{ year}^{-1}$ ($P < 0.001$) (Fig. 3d).

211 Over the full 0-2.0 m depth, the SOC stock at the beginning of the experiment in 2004
212 was 137 Mg C ha^{-1} , decreased to 133 Mg C ha^{-1} in the bare soil plots, while it
213 increased to 151, 157 and 161 Mg C ha^{-1} under the milk vetch, bush clover and alfalfa
214 stands, respectively, by the end of the experiment in 2010 (Fig. 4). The SOC stock
215 increased more under the stand of alfalfa than milk vetch, but there was no significant
216 difference between alfalfa and bush clover (Fig. 4). When calculated over the full 2-m
217 soil layer, over the 7-year growth period, the SOC stocks increased by $24.1 \text{ Mg C ha}^{-1}$,
218 $19.9 \text{ Mg C ha}^{-1}$ and $14.6 \text{ Mg C ha}^{-1}$ under the alfalfa, bush clover and milk vetch
219 stands, respectively, and decreased by 4.2 Mg C ha^{-1} under bare soil (Fig. 5). In the
220 1.0-2.0-m soil layer the stocks of SOC increased by $19.1 \text{ Mg C ha}^{-1}$, $13.6 \text{ Mg C ha}^{-1}$
221 and $10.8 \text{ Mg C ha}^{-1}$, under the alfalfa, bush clover and milk vetch stands, respectively,
222 that is, by 79, 68 and 74% of the increases over the whole soil profile (Fig. 5).

223 **4. Discussion**

224 While the unplanted plots lost SOC over the 7 years of the study, particularly over the
225 first 5 years, all three forage legumes more-than countered this loss, so that over the 7
226 years the SOC increased on average by $2.1 \text{ Mg C ha}^{-1} \text{ y}^{-1}$, $2.8 \text{ Mg C ha}^{-1} \text{ y}^{-1}$ and 3.4
227 $\text{Mg C ha}^{-1} \text{ y}^{-1}$, to sequester 15 Mg C ha^{-1} , 20 Mg C ha^{-1} and 24 Mg C ha^{-1} under the
228 milk vetch, bush clover and alfalfa stands, respectively (Fig. 5). The legumes

229 increased the SOC primarily at depth (Fig. 2) with between 68 and 79% of the SOC
230 being sequestered below 1 m.

231 An increase in SOC stock is usually associated with the production, turnover,
232 sloughing off of epidermal cells, and exudation of soluble carbon compounds by the
233 roots, particularly the turnover and exudation by fine roots (Luo et al., 1995). A high
234 rate of turnover of fine roots and a high rate of exudation of carbon by the roots
235 influences the stability of plant C in soil and the accumulation of SOC (Shahzad et al.,
236 2015). Although the root biomass was not measured in this study, root biomass is
237 usually associated with aboveground biomass. If the root:shoot biomass ratio is
238 known, the root biomass and root carbon accumulated in the soil can be estimated
239 from the aboveground biomass and compared with the changes in SOC over the same
240 period. Using an average root:shoot ratio of 0.77:1 in the three species (Chen and Nie,
241 1978; Cheng et al., 2004; Fan et al., 2015; Sun et al., 2001), the 91 t ha⁻¹ of
242 aboveground biomass (43 Mg C ha⁻¹) produced over the 7-year life of the alfalfa
243 pasture would result in the production of 70 t ha⁻¹ of root biomass or 33 Mg C ha⁻¹ of
244 root carbon, significantly higher than the measured increase in SOC stock in the upper
245 2 m of the soil of 24 Mg C ha⁻¹. Similarly, using an aboveground biomass of 56 t ha⁻¹,
246 the estimated production of root biomass by milk vetch would be 43 t ha⁻¹ and the
247 estimated root carbon would be 20 Mg C ha⁻¹, also higher than the increase of 15 Mg
248 C ha⁻¹ in SOC stock measured under milk vetch. However, using the same root:shoot
249 ratio of 0.77:1, the aboveground biomass of bush clover of 42 t ha⁻¹ would produce an
250 estimated root biomass of 32 t ha⁻¹ and a production of root carbon of 15 Mg C ha⁻¹,

251 compared with the measured SOC of 20 Mg C ha⁻¹. Thus the actual accumulation of
252 SOC was about 70% of that estimated from the aboveground biomass in alfalfa and
253 milk vetch, but was 33% higher than that estimated in bush clover. The difference
254 between the estimated root carbon accumulated and the measured SOC stock under
255 alfalfa and milk vetch over the 7-year period was presumably the result of losses by
256 respiration by the roots and associated soil microbial populations. The observation
257 that the measured values of SOC were higher than the root carbon production
258 estimated from aboveground biomass may indicate that the root:shoot ratio was
259 greater than 0.77:1 in bush clover (a root:shoot ratio of 1:1 would make the estimated
260 and measured values the same), or the bush clover had a greater proportion and
261 turnover of fine roots than milk vetch and alfalfa, resulting in a greater accumulation
262 of SOC (Shahzad et al., 2015). This is consistent with Sun et al. (2001) who reported
263 that the fine roots (root diameter < 0.5 mm) of bush clover accounted for 42% of total
264 root biomass in 0-0.3 m soil layer, while the fine roots of milk vetch were only 25%
265 of total root biomass (Chen and Nie, 1978).

266 The accumulation of SOC by bush clover was particularly high in the upper 0.3 m of
267 the soil (Fig. 2, Fig. 3). This accumulation of SOC in the upper soil layer may be
268 attributable to the high accumulation of legume residues and litter (Zhou et al., 2006),
269 or due to the proliferation and turnover of roots in this surface layer. Nevertheless, the
270 sequestration of SOC in the upper 0.3 m of the soil in this study was significantly
271 lower than Zhang et al. (2009) who reported that the SOC stocks in upper 0.3 m of the
272 soil increased by 16 Mg C ha⁻¹ in 10 years from the conversion of a wet reed meadow

273 to an irrigated alfalfa pasture in the Hexi Corridor of north-west China. This suggests
274 that well-managed legume pastures in areas with higher precipitation and with
275 appropriate fertilizer use could sequester significantly more SOC than in the present
276 unirrigated and unfertilized legumes growing in a semiarid environment.

277 An unexpected result from this study was the greater increase in SOC at soil depths
278 from 1 – 2 m than above 1 m, accounting for 79, 68 and 74% of SOC sequestered
279 through the whole 2 m of soil under alfalfa, bush clover and milk vetch, respectively.
280 This suggests the presence of roots deep in the soil profile. Indeed, alfalfa and milk
281 vetch have taproots that can penetrate to 6.8 and 7.6 m, respectively, in loess soils in 6
282 years (Cheng et al., 2005; Cheng et al., 2004), and the water extraction patterns
283 measured on the plots used in this study clearly showed the presence of roots
284 throughout the upper 2 m of the soil profile (Guan et al., 2013). In bush clover in
285 which the proportion of SOC below 1 m was smaller than in the other two species, the
286 taproot predominates in the 0-0.3 m soil layer with coarse roots (root diameter > 2
287 mm) accounting for 48% of the total root biomass, and fine roots predominating
288 below 0.3 m (Cheng et al., 2005; Cheng et al., 2004; Sun et al., 2001). Although
289 alfalfa extracted more water below 1 m than the other two species, the water
290 extraction patterns do not suggest a greater root presence below 1 m in milk vetch
291 than bush clover (Guan et al., 2013). We suggest that the greater increase in SOC at
292 depth may be associated with a greater proliferation and turnover of fine roots at
293 depth, or alternatively may reflect the movement down the profile of soluble C

294 compounds from the roots as a result of the movement of water after heavy rainfall
295 events.

296 The conversion of arable land that had been growing crops for many years to
297 perennial legume pasture resulted in a significant increase in SOC, particularly at soil
298 depths below 1 m. All three legume species increased the SOC in the top 2 m of the
299 soil profile, but the increase was greatest in alfalfa and least in milk vetch. While the
300 production of aboveground biomass was least in bush clover, the SOC sequestration
301 in the soil profile was not significantly different from alfalfa, indicating that carbon
302 sequestration in the soil is not simply associated with aboveground biomass
303 production in a system in which the forage is removed for animal feed, as in the
304 present study. Further study of root biomass production, turnover of fine roots and
305 exudation of carboxylic acids and other carbon compounds by the roots of the legume
306 species would be a valuable step in understanding the differences in carbon
307 sequestration by the three species.

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420 **Table 1. Annual aboveground biomass production of the three legume species,**
 421 **milk vetch, alfalfa and bush clover, from 2004 to 2010. Adopted from Guan et al.**
 422 **(2013) and used with permission.**

Year	Aboveground biomass production (t ha ⁻¹)		
	Milk vetch	Alfalfa	Bush clover
2004	2.2Ac	2.3Ad	0.2Bd
2005	14.1Ba	20.2Aa	5.3Cc
2006	14.3Ba	22.2Aa	7.8Ca
2007	6.8Bb	9.3Ac	6.4Bbc
2008	5.6Bb	13.4Ab	7.3Bab
2009	7.2Bb	12.4Ab	7.8Ba
2010	5.8Bb	10.8Abc	7.4Bab
2004-2010 Mean	8.0B	13.0A	6.0C
2004-2010 Total	56.0B	90.7A	42.1C

423 Data in each column with a different lower-case letter are significantly different ($P <$
 424 0.05) and data in each row with a different capital letter are significantly different (P
 425 < 0.05).

426

427 **Table 2. Results of the ANOVA for soil organic carbon concentration as affected**
 428 **by legume species, soil depth and experimental year. The bare soil plot is**
 429 **considered as a legume species in the analysis. The SAS PROC ANOVA was**
 430 **used in the analysis.**

Factors	df	<i>F</i> value	<i>Pr</i> > <i>F</i>
Species	3	38.52	0.0003
Depth	4	1649.40	<0.0001
Year	7	31.68	<0.0001
Species*Depth	12	5.65	<0.0001
Species*Year	21	5.96	<0.0001
Depth*Year	28	3.20	<0.0001
Species*Depth*Year	84	0.95	0.6053

431

432

433 **Figure Captions**

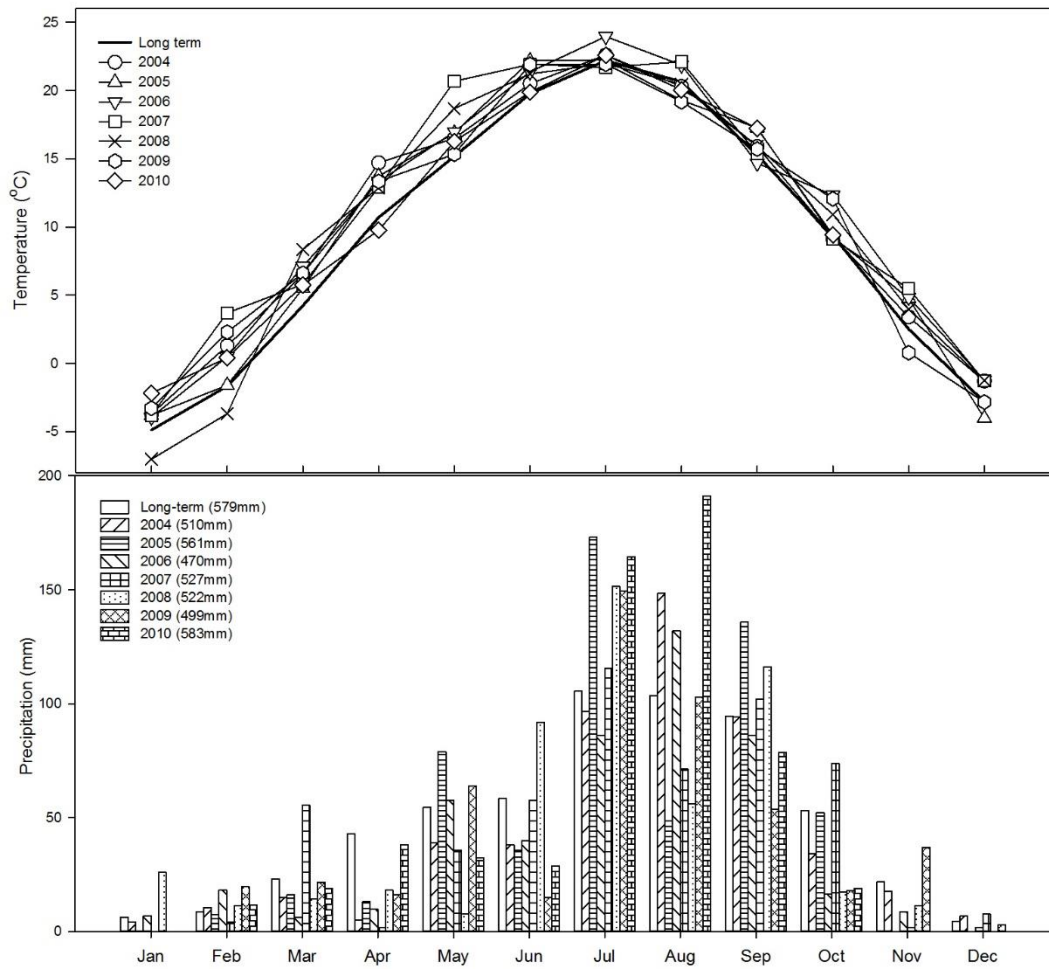
434 **Fig. 1.** Mean monthly temperature and precipitation from 2004-2010 and the long
435 term mean at the experimental site at Changwu Agricultural Research Station,
436 Shaanxi Province, China.

437 **Fig. 2.** Concentration of soil organic carbon (SOC) with depth in May 2004 (IV) and
438 in October 2010 under three forage legumes: milk vetch, alfalfa and bush clover, and
439 bare soil (CK). Bars give + one standard error of the mean ($n = 3$).

440 **Fig. 3.** Change with stand age in soil organic carbon amount (stock) per hectare at soil
441 depths of 0-0.3m (a), 0.3-0.6m (b), 0.6-1.0m (c), 1.0-1.5m (d) and 1.5-2.0m (e) under
442 milk vetch, alfalfa, bush clover and bare soil (CK). Note the soil layers vary in depth.
443 Data are means \pm one standard error of the mean ($n = 3$) when larger than the symbol.
444 Linear regressions fitted when significant and fitted regressions given.

445 **Fig. 4.** The soil organic carbon amount (stock) under milk vetch, alfalfa, bush clover
446 and under bare soil (CK) over the upper 2 m of the soil profile. The lower case letters
447 indicate significant differences ($P < 0.05$) between forage types and bare soil (CK)
448 within a year. IV denotes initial value, the soil organic carbon stock in May 2004.
449 Bars give + one standard error of the mean ($n = 3$).

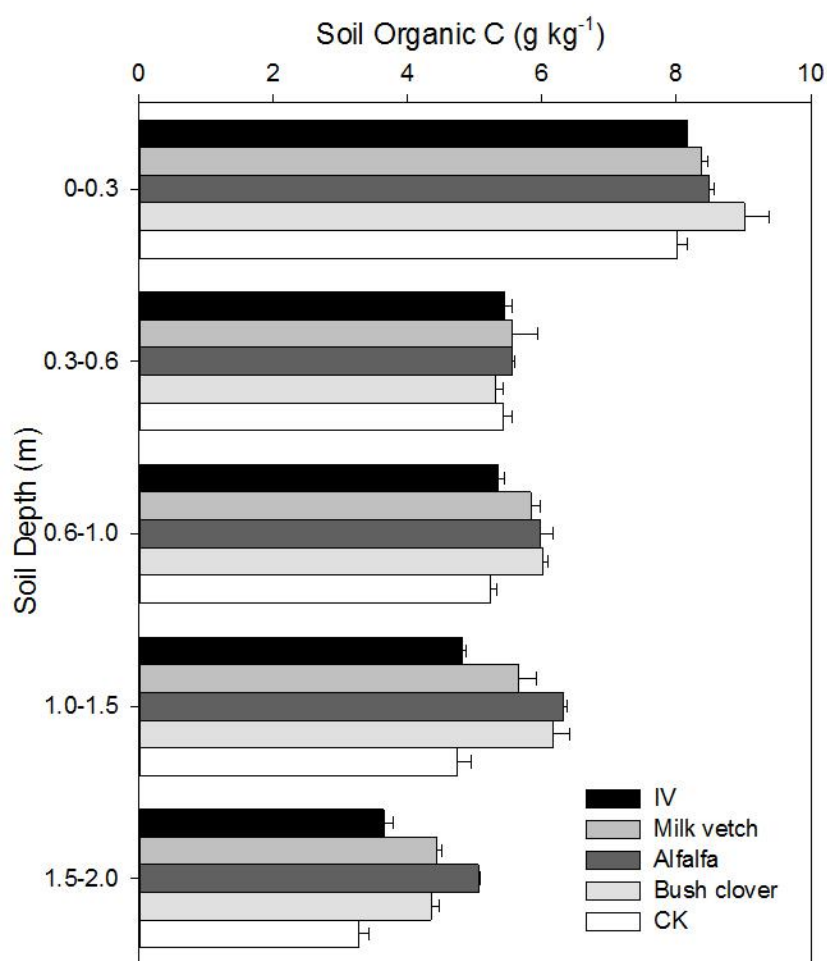
450 **Fig. 5.** Change in soil organic carbon amount (stock) in different soil layers under
451 milk vetch, alfalfa, bush clover and bare soil (CK) from May 2004 to October 2010.
452 Different letters indicate significant differences ($P < 0.05$) between total carbon stocks.



453

454 Figure 1

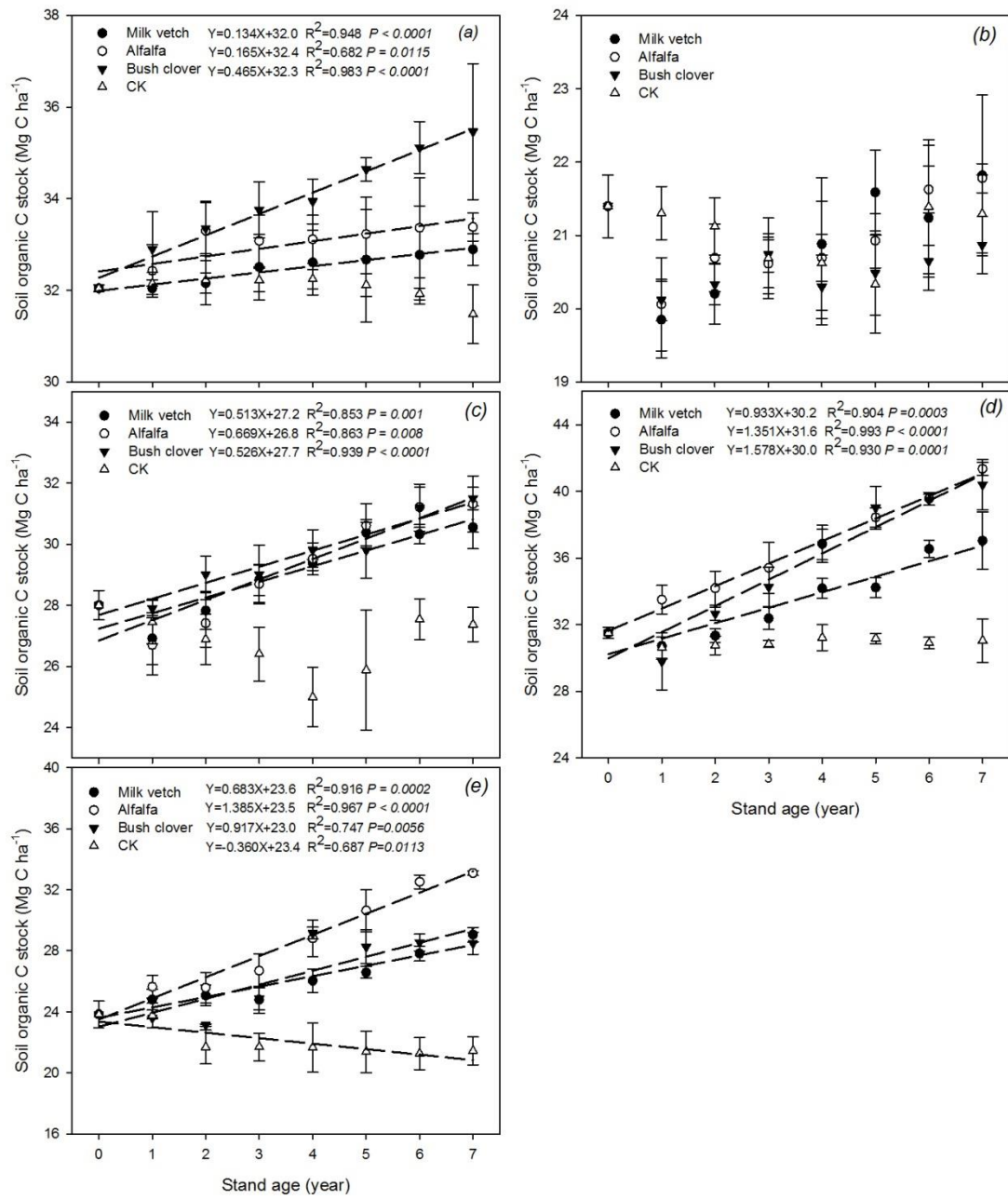
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457 Figure 2

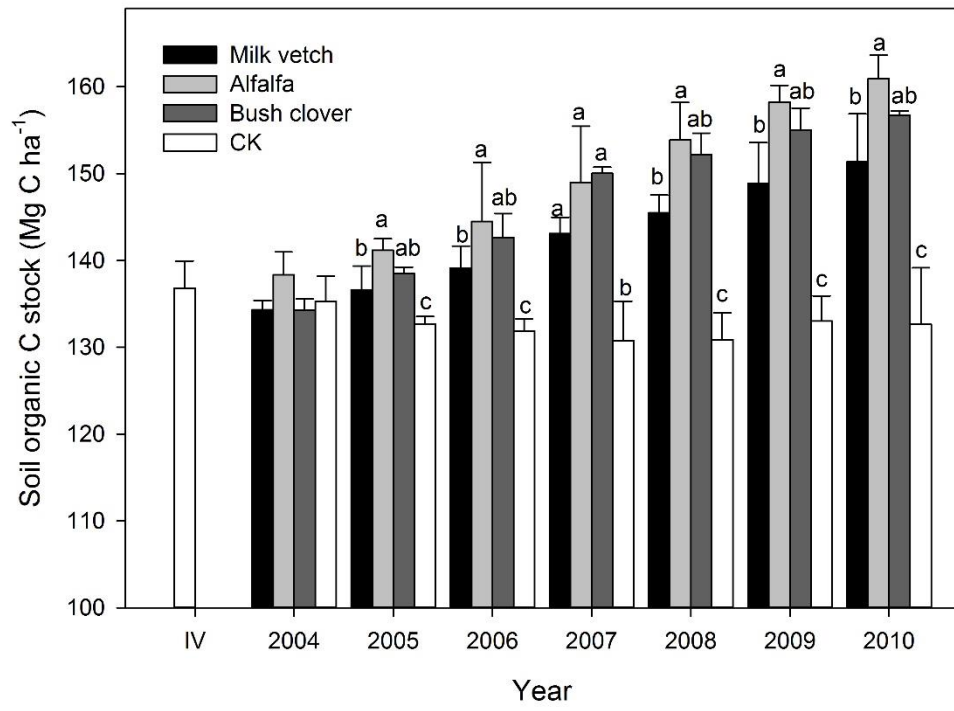
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460 Figure 3

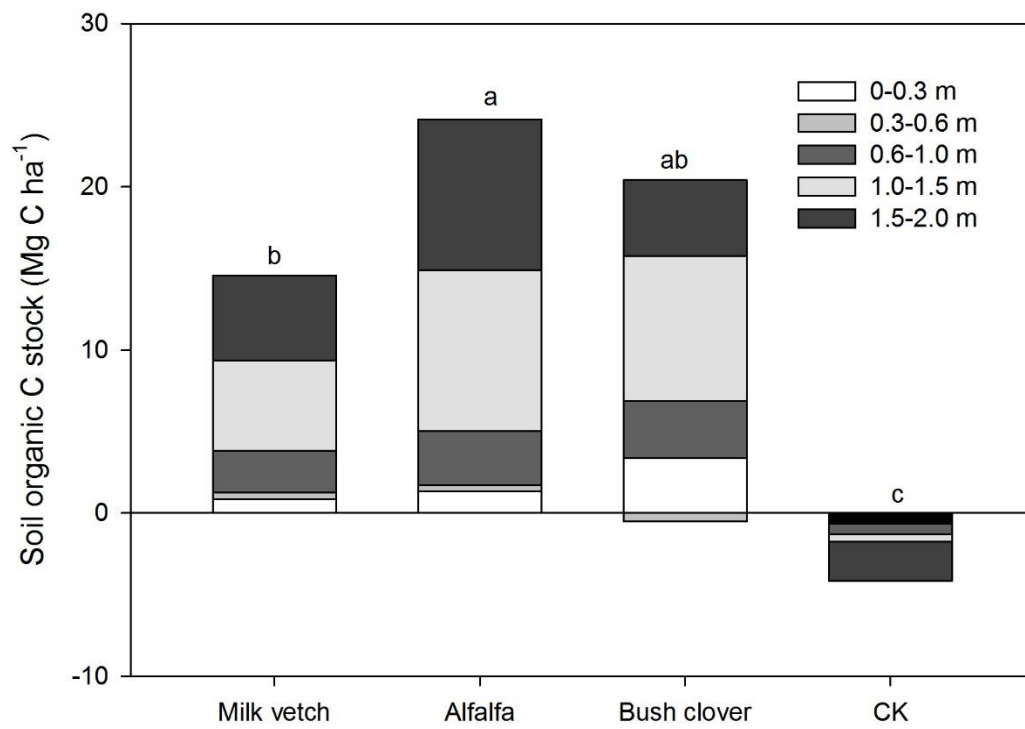
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463 Figure 4

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466 Figure 5