



12 **Abstract:**

13 Vegetation restoration was effective way of protecting soil erosion and water
14 conservation on the Loess Plateau. Carbon fractions and enzyme activities were
15 sensitive parameters for assessment of soil remediation through revegetation. Forest,
16 forest steppe and grassland soils were collected at 0-5 cm and 5-20 cm soil layers in
17 Yanhe watershed, Shaanxi Province. Urease, sucrase, alkaline phosphatase, soil
18 organic carbon (SOC), microbial biomass carbon (MBC), easily oxidized organic
19 carbon (EOC) and dissolved organic carbon (DOC) were measured. The results
20 showed that carbon fraction contents and enzyme activities in the same soil layer
21 followed the order that forest was higher than others. Carbon fraction contents and
22 enzyme activities appeared that the 0-5 cm was higher than 5-20 cm soil layer. In
23 addition, correlation analysis showed that urease activity was related to SOC, MBC,
24 EOC and DOC at 0-5 cm layer; it was correlated with SOC, MBC and EOC at 5-20
25 cm layer. Sucrase activity had significant positive relationship with SOC, MBC and
26 EOC. Alkaline phosphatase activity was related to EOC and DOC at 0-5 cm layer; it
27 was correlated with MBC and EOC at 5-20 cm layer. The CCA reflected the
28 relationship between sucrase activity and SOC. The contributions from the various
29 forms of carbon fractions and enzyme activities as evaluated by the canonical
30 coefficient of CV were on the order of SOC > DOC > MBC > EOC; sucrase > urease >
31 alkaline phosphatase. Vegetation type was an important factor influencing the
32 variation of soil enzyme activities and carbon fractions on the Loess Plateau.

33 **Key Words:** vegetation types; soil organic carbon; soil microbial biomass carbon; soil



34 easily oxidized organic carbon; soil dissolved organic carbon; soil enzyme activities

35 **1. Introduction**

36 Land degradation and soil erosion are serious problems in the Loess Plateau of China
37 (Fu et al., 2005; Zheng et al., 2005). Zheng et al., (2005) reported that the nutrient loss
38 was strongly related to erosion patterns and erosion intensity. Since 1999, the Grain
39 for Green Project had been implemented in the Loess Plateau. It induces improvement
40 in vegetation conditions may benefit soil erosion alleviation and carbon sequestration
41 in the Loess Plateau (Wang et al., 2011; Zhou et al., 2012). Studies of revegetation
42 after farmland abandonment in the Loess Plateau of China indicated that soil physical
43 properties are closely related to the vegetation recovery stages (Li and Shao, 2006;
44 Zuo et al., 2009). Some researchers stress that the vegetation restoration in Loess
45 Plateau is very important for soils health, a long-term experiment show that
46 integrative measures restore forests and stop soil erosion on the severely eroded bare
47 land (Zhang et al., 2004); Chen and Cai (2006) found that reduction of reclamation
48 rate and the increase of tree and grass vegetation could control soil erosion in the
49 sandy and coarse sandy areas; and when human activities destroyed secondary forests,
50 soil erosion increased (Zheng, 2006). Recently, some studies have concentrated on the
51 vegetation restoration, for instance, Jiao et al., (2011) found that revegetation had
52 positive effects on the soil physical properties. In the protected vegetation areas,
53 relative humidity of air increased and wind velocity is greatly reduced. Additionally,
54 bulk density of the surface layer (0-20 cm) significantly decrease while soil porosity,
55 water-holding capacity, aggregate stability, and saturated hydraulic conductivity



56 significantly increase. SOC stocks are increased by 19% in the surface soil layer at
57 0-20 cm soil depth from 1998 to 2006, because of the vegetation restoration in the
58 Loess Plateau (Wang et al., 2011).

59 The soil carbon fractions include soil organic carbon (SOC), microbial biomass
60 carbon (MBC), easily oxidized organic carbon (EOC) and dissolved organic carbon
61 (DOC). Soil organic carbon and enzyme activities are indicators of soil fertility
62 (Gregorich et al., 1994; Lagomarsino et al., 2011). SOC storage is estimated about
63 two and three times the size of carbon pools in the atmosphere and vegetation,
64 respectively (Jobbágy and Jackson, 2000; Lal, 2004). SOC stocks in 0-30 cm soil
65 layer are highly variable among the vegetation communities (Yimer et al., 2006). SOC
66 plays a key role in the global C cycle (Noble et al., 2000) and as indicator of soil
67 quality (Gregorich et al., 1994); it is also an important component of agricultural soils
68 (Fang et al., 2012). Labile organic carbon (MBC, EOC and DOC) plays an important
69 character in short-term turnover of soil nutrients and provides energy for microbes
70 (Piao et al., 2000); it has a higher activity for microbes (Shen et al., 1999). Soil MBC
71 is used as an indicator of changes in soil organic matter (Jenkinson, 1988; Saffigna et
72 al., 1989), it generally represents 2–3% of soil organic C (Anderson and Domsch,
73 1989). EOC is an indicator of soil labile organic carbon (Biederbeck et al., 1994).
74 DOC is sensitive to soil quality and fertility transformations, hence it can better reflect
75 the soil physical and chemical properties (Lu et al., 2006).

76 Enzyme activities can express soil quality by providing useful linkages between
77 the microbial community structures and the environmental factors (Zhang et al., 2015).



78 Ecoenzymatic stoichiometry, microbial respiration, and organic matter decomposition
79 are responsive to resource availability and the environmental drivers of microbial
80 metabolism (Hill et al., 2014). Large numbers of these enzymes are expressed and
81 released into the environment by microorganisms in response to environmental
82 signals (Sinsabaugh et al., 2009). The soil enzyme activities can be crucial in
83 detecting differences among forest, monoculture and intercropping (de Medeiros et al.,
84 2015). Microbial enzyme allocation is sensitive to differences in nutrient limitation
85 (Moorhead et al., 2012). However, there is a lack of information on the relationship
86 between soil carbon and enzyme activities for soils with different vegetation types.
87 We advanced the following three hypotheses.

88 H1: both carbon fraction contents and enzyme activities in the same soil layer are
89 higher for forest than for forest steppe and grassland.

90 H2: carbon fraction contents and enzyme activities under all vegetation soils are
91 higher in the surface layer than in the underlying layer.

92 H3: different carbon fractions have different effects on enzyme activities in soil.

93 To this end we investigated four carbon fractions and three enzyme activities under
94 various type vegetations considered in our experiments.

95 **2. Materials and methods**

96 2.1 Study sites

97 The field site (107°41'~110°31'E, 35°21'~37°31'N) is located in Yanhe watershed,
98 northern Shaanxi Province, China (Table 1). It belongs to the hilly-gully part of the
99 Loess Plateau and has a total area of 37029 km². Its average elevation is about 1000 m.



100 It has a continental arid to semi-arid climate, with an annual average frost-free period
101 of 170 d, an annual average temperature of 9.2 °C, an annual average sunshine
102 duration of 2500 h, and an annual average precipitation of about 500 mm (CCSLC,
103 2000).

104 2.2 Soil collection and processing

105 Soils were collected in August, 2013, on three typical vegetation types (grassland,
106 forest steppe and forest). For each vegetation type, four representative plant
107 communities were chosen (Table 1), and, as replicates, three sampling areas were
108 defined in the field for each representative plant community. In each representative
109 plant community, three sampling plots were delineated. The sizes of the sampling
110 plots were: 20×20 m for forest, 5×5 m for forest steppe and 1×1 m for grassland.
111 Within each plot, based on an S-shaped sampling pattern, the incompletely-degraded
112 litter was removed and 9 sub-samples were simultaneously and randomly collected
113 then mixed them in the same bag which as a representative soil sample, separately at
114 0-5 cm and 5-20 cm depth. The representative soil sample was split into two parts,
115 one was stored intact at -20 °C in order to determine carbon fractions and enzyme, and
116 the other was air-dried for measuring soils' physics and chemical properties.

117 2.3 Methods

118 2.3.1 Carbon assay

119 SOC was determined by wet digestion with a mixture of potassium dichromate and
120 concentrated sulfuric acid (ISSSC, 1981). The soil organic matter is various in
121 different type soil, 0.1 g of air-dried soil was weighed in boiling tube, then 5 ml



122 K_2CrO_4 and 5 ml concentrated sulfuric acid were added and shaken well. The sample
123 was put into the 185-190 °C paraffin oil bath. Soil sample was taken out after boiling
124 for 5 min. After cooling, the substance in the tube was transferred into an Erlenmeyer
125 flask, and 2-3 drops of phenanthroline indicator were added before being titrated with
126 $FeSO_4$ solution. The color of the solution changed from orange to blue and in the end
127 it turned brick red with $FeSO_4$ titration solution adding.

128 The MBC was measured using the chloroform fumigation–extraction method
129 (Ross, 1990). The soil sample was taken from -20 °C freezer and thawed, 100 g soil
130 which adjusted to 60% of field capacity was added to a 500 ml jar, incubated for 7
131 days at 25 °C. The soil sample was exposed to chloroform vapor in a vacuum
132 desiccator at 25 °C for 24 h. After chloroform fumigation, the total carbon content was
133 determined in the 0.5 M K_2SO_4 extract. Determination of carbon content used the
134 TOC-1020A organic carbon analyzer (Phoenix 8000, USA).

135 Soil EOC was measured using a slightly modified version of the light group
136 organic compound separation method of (Janzen et al., 1992). A sample containing 15
137 mg of carbon was put into a 100 ml centrifuge tube. 25 ml of 333 mMol/L potassium
138 permanganate was added and shaken for an hour, and then centrifuged at 4000 rpm
139 for 5 min. The supernatant was diluted with deionized water at 1:250, and then the
140 absorbances of the blank and soil sample were determined by spectrophotometry at
141 565 nm (TOC-1020A organic carbon analyzer, Phoenix 8000, USA). By comparing
142 the absorbances of the blank and soil sample, the change of the potassium
143 permanganate concentration was calculated, and then the amount of oxidated carbon.



144 Soil DOC was measured by K_2SO_4 leaching-TOC method (Murphy et al., 2000).
145 10 g of air-dried 2-mm-sieved sample was weighed, and distilled water was added at a
146 ratio of 2:1. After shaking for 30 min at 25 °C constant temperature, filtering was
147 carried out on a membrane filter and was determined using the TOC-1020A organic
148 carbon analyzer (Phoenix 8000, USA)

149 2.3.2 Enzyme assay

150 Urease activity was determined by indophenol blue colorimetry (Guan et al., 1991). 5
151 g of air-dried 2-mm-sieved soil was added into 50 ml Erlenmeyer and 1 ml toluene
152 was added. Then it was left for 15 min before adding 10 ml of 10% urea solution and
153 20 ml of sodium citrate buffer, and shook well. The sample was subsequently
154 incubated at 37 °C for 24 h and then diluted to 50 ml with 37 °C distilled water. The
155 suspension was filtered and 1 ml of the extract was added to a 50 ml flask with 4 ml
156 sodium phenol solution and 3 ml sodium hypochlorite solution. After shaking well
157 and then let it rest for 20 minutes, the released ammonium was extracted with
158 potassium chlorite solution. The ammonium was quantified colorimetrically with a
159 spectrophotometer (2800 UV/VIS) at 578 nm.

160 Sucrase activity was determined by 3, 5 - dinitrosalicylic acid colorimetry (Guan
161 et al., 1991). 5 g of soil was weighed in an Erlenmeyer and 5 drops of toluene was
162 added before being gently shaken. Let it rest for 10 minutes, then 15 mL of 8%
163 glucose solution and 5 mL of phosphate buffer at pH 5.5 were added. The sample was
164 subsequently incubated at 37 °C for 24 h. The suspension was filtered and 1 ml of the
165 extract was added to a 50 ml flask. 3 mL of 3, 5 - dinitrosalicylic acid was added



166 before a 5 min heating in a boiling water bath. It was then diluted to 50 ml. The
167 sample colorimetric measurement was determined in spectrophotometer (2800
168 UV/VIS) at 508 nm.

169 Alkaline phosphatase was determined using disodium phenyl phosphate method
170 (Guan et al., 1991). 2 g sample was weighed in 50 ml tube, then 2.5 ml of toluene and
171 20 ml of 0.5% disodium phenyl phosphate were added. The sample was subsequently
172 incubated at 37 °C for 24 h. 100 ml of 0.3% aluminum sulfate solution was added
173 before filtering. 3 ml of filtrate was added into a 50 ml flask. The sample colorimetric
174 measurement was determined in spectrophotometer (2800 UV/VIS) at 660 nm.

175 2.4 Data analysis

176 Data were processed by Excel 2010; statistical analyses were carried out with SPSS
177 19.0 and plotting by Origin Pro. 8.0. One-way ANOVA conducted by the Scheffe test
178 ($p < 0.05$) was used to compare the differences among vegetation types.

179 A canonical correlation coefficients analysis (CCA) was carried to assess the
180 relationship between two datasets: soil carbon fractions and enzyme activities. The
181 CCA is designed to identify linear combinations of variables in one dataset that
182 account for the greatest variation in a linear combination of variables for the other
183 dataset (Lattin et al., 2003). In this study, the CCA was performed using the soil
184 carbon fractions and enzyme activities, three pairs of canonical variates (CVs) were
185 extracted. The U_1 and V_1 refer to the first group equation between soil carbon
186 fractions canonical variate (X-CV) and the enzyme activities canonical variate (Y-CV).
187 The indices of the X-CV were: SOC (X1), MBC (X2), EOC (X3), and DOC (X4).



188 The indices of the Y-CV were: urease activity (Y1), sucrase activity (Y2), alkaline
189 phosphatase activity (Y3).

190 **3. Results**

191 3.1 Physical and chemical properties depending on the vegetation type

192 Vegetation types had great effects on the soil basic physical and chemical properties.
193 The bulk density was significantly different between forest and grassland (Table 2).
194 Forest steppe vegetation's bulk density was significant difference between the 0-5 cm
195 and 5-20 cm soil layers. The pH was no significant difference between the two layers
196 of a given vegetation type and it was significantly different between forest and
197 grassland in both soil layers ($p < 0.05$). The total N concentration of forest was
198 significantly higher than the total N of both forest steppe and grassland, in both soil
199 layers. The total P concentration of grassland vegetation was significantly lower than
200 for both forest and forest steppe, in both soil layers. There was no significant
201 difference in the same vegetation's total P and total N concentration between the two
202 soil layers. No significant difference in soil organic matter was seen between forest
203 steppe and grassland, while soil organic matter of forest was significantly higher.
204 Forest steppe and grassland vegetation's soil organic matter was significantly different
205 between the 0-5 cm and 5-20 cm soil layers.

206 3.2 Soil carbon fractions depending on the vegetation type

207 The SOC, MBC and EOC contents of forest soils were significantly different from
208 both forest steppe and grassland soils, and there was no significant difference between
209 forest steppe and grassland vegetation (Fig. 1). The DOC concentration of forest



210 vegetation was significant difference to grassland vegetation at 0-5 cm soil layer, and
211 grassland vegetation also was significant difference to both forest steppe and forest
212 vegetation at 5-20 cm soil layer. Forest steppe and grassland vegetation's SOC and
213 EOC concentrations were significant difference between the upper and lower soil
214 layers, except for forest vegetation. There was no significant difference among forest,
215 forest steppe and grassland vegetation's MBC concentration between the two layers.
216 Forest and forest steppe vegetation's DOC concentrations were significantly different
217 between the upper and lower soil layer, except for grassland vegetation. SOC contents
218 at 0-5 cm soil layer was more 6.73, 3.13 and 1.29 $\text{g}\cdot\text{kg}^{-1}$ than 5-20 cm soil layer under
219 forest, forest steppe and grassland; MBC contents at upper soil layer was more 227.44,
220 102.94 and 62.05 $\text{mg}\cdot\text{kg}^{-1}$ than lower soil layer under forest, forest steppe and
221 grassland; EOC contents at upper soil layer was higher 2.24, 0.31 and 0.21 $\text{g}\cdot\text{kg}^{-1}$ than
222 lower soil layer under forest, forest steppe and grassland; DOC contents at upper soil
223 layer was higher 166.06, 122.07 and 44.97 $\text{mg}\cdot\text{kg}^{-1}$ than lower soil layer under forest,
224 forest steppe and grassland respectively.

225 3.3 Soil enzyme activities depending on the vegetation type

226 For the 0-5 cm soil layer, the urease activity of forest vegetation was significantly
227 different from forest steppe and grassland vegetations (Fig. 2A), while the sucrase
228 activity and the alkaline phosphatase activity of forest vegetation were significantly
229 different from forest steppe and grassland vegetations (Fig. 2B & 2C). For the 5-20
230 cm soil layer, the urease was significantly different between forest and grassland
231 vegetations (Fig. 2A), while the sucrase activity was non-significantly different



232 among forest, forest steppe and grassland vegetations (Fig. 2B). The alkaline
233 phosphatase activity was significant difference between forest and grassland
234 vegetation (Fig. 2C). Forest and forest steppe vegetation's urease and alkaline
235 phosphatase activities were significantly different between the upper and lower soil
236 layers, except for grassland vegetation (Fig. 2A & 2C). However, forest, forest steppe
237 and grassland vegetation's sucrase activities were not significantly different between
238 the upper and lower soil layers (Fig. 2B). The urease activity of all vegetation type
239 soils appeared that the upper more than lower soil layer, and forest, forest steppe and
240 grassland increased 35.94, 58.44 and 46.55 percentage which increased 0.46, 0.45 and
241 0.27 mg kg^{-1} respectively. The sucrase activity of all vegetation type soils appeared
242 that the upper more than lower soil layer, and forest, forest steppe and grassland
243 increased 53.74, 31.66 and 29.19 percentage which increased 8.9, 3.59 and 2.84
244 mg kg^{-1} respectively. At 0-5 cm soil layer, sucrase activity of forest vegetation was
245 1.71 and 2.03 times compared with forest steppe and grassland vegetation,
246 analogously at 5-20 cm soil layer, sucrase activity of forest vegetation was 1.46 and
247 1.70 times by comparing with forest steppe and grassland vegetation. Soil alkaline
248 phosphatase activity of all vegetation type soils appeared that the upper more than
249 lower soil layer, and forest, forest steppe and grassland increased 27.58, 33.84 and
250 28.26 percentage which increased 0.91, 0.89 and 0.39 mg kg^{-1} respectively. At 0-5 cm
251 soil layer, alkaline phosphatase activity of forest vegetation was 1.20 and 2.38 times
252 compared with forest steppe and grassland vegetation, analogously at 5-20 cm soil
253 layer, alkaline phosphatase activity of forest vegetation was 1.25 and 2.39 times by



254 comparing with forest steppe and grassland vegetation.

255 3.4 Correlations between soil carbon fraction contents and enzyme activities

256 3.4.1 Correlations between soil carbon fraction contents and enzyme activities of

257 different vegetation types at 0-5 cm soil layer

258 At 0-5 cm soil layer under all vegetations, soil urease activity was significant

259 correlated extremely with SOC, MBC, EOC and DOC which correlation coefficient

260 were 0.823, 0.787, 0.775 and 0.886. Soil sucrase activity was positively significant

261 correlated with SOC, MBC and EOC which correlation coefficient was 0.907, 0.877

262 and 0.818, there was non-significant correlation with DOC. Soil alkaline phosphatase

263 activity was positively significant correlated with DOC which correlation coefficient

264 was 0.727, and it also was significant correlated with EOC which correlation

265 coefficient was 0.588, and there was non-significant correlation with SOC and MBC

266 (Table 3).

267 3.4.2 Correlations between soil carbon fraction content and enzyme activity of

268 different vegetation types at 5-20 cm soil layer

269 Soil urease activity was positively significant correlated with SOC which correlation

270 coefficient was 0.762, meanwhile, it was significant related to MBC and EOC which

271 correlation coefficient was 0.633 and 0.621, however, there was non-significant

272 correlation with DOC. Soil sucrase activity was positively significant correlated with

273 SOC and MBC which correlation coefficient was 0.759 and 0.840, simultaneously, it

274 was also significant related to EOC which correlation coefficient was 0.593, there was

275 non-significant correlation with DOC. Soil alkaline phosphatase activity was



276 significant related to MBC and EOC which correlation coefficient was 0.656 and
277 0.600 (Table 4).

278 The CCA was performed using the soil carbon fractions and enzyme activities,
279 and three pairs of canonical variates (CVs) were extracted. The canonical correlation
280 between the first soil carbon fractions canonical variate ($X-CV_1$) and the first enzyme
281 activities canonical variate ($Y-CV_1$) was significant ($R=0.964$; $P<0.001$). This first
282 canonical variate mainly reflected the relationship between the sucrase activity and
283 SOC. Around 70% of the variance in the $Y-CV_1$ was explained by the $X-CV_1$ (Table 3).
284 The contributions from the various forms of carbon fractions as evaluated by the
285 canonical coefficient of CV were on the order of $SOC > DOC > MBC > EOC$. The
286 enzyme activities were on the order of sucrase $>$ urease $>$ alkaline phosphatase.

287 **4. Discussion**

288 4.1 Soil carbon fraction of different type vegetations

289 Various factors influence on SOC such as the climate, soil, vegetation, and human
290 disturbance (Solomon et al., 2007). There are differences among soil carbon fractions
291 in various type vegetations, due to the diverse restoration years, stages and types of
292 vegetations (Novara et al., 2015). Meanwhile, decomposition also the main reason, it
293 is a fundamental ecosystem process and a key ecological process that controlled
294 nutrient availabilities to plants in terrestrial ecosystems (Moorhead et al., 1996).
295 Johnsen et al. (2013) found that the amount of C entering the soil through greater
296 forest litter and belowground biomass production. Decomposition in response to
297 variations in litter quality and key parameter values (Moorhead et al., 2012), and the



298 first step in detritus decomposition results from the activity of enzymes produced by
299 soil microbes (Wallenstein et al., 2009). The soil physical and chemical properties
300 regulate decomposition rates (Xu et al., 2016). The soils of the study are sampled
301 from different vegetation types, therefore, their decomposition rates are various and
302 the carbon pools also are different (Xu et al., 2016). Soil organic matter represents the
303 largest terrestrial pool for carbon storage (ParrasAlcántara et al., 2015). About
304 three-fourth of organic carbon contained in terrestrial ecosystems are found in soil
305 organic matter and plant litters (Schlesinger, 1997; Lal, 2008). Organic matter
306 decomposition is responsive to resource availability and the environmental drivers of
307 microbial metabolism (Hill et al., 2014). The decomposition of plant litter may be the
308 biosphere's most complex ecological process (Sakamoto and Oba, 1994).
309 Microorganism plays an important role in forest soil carbon and nutrient
310 transportation (Lipson et al., 2002). Microbial communities are mainly influenced by
311 local environmental properties (Fierer and Jackson, 2006). Any changes in the
312 microbial biomass may affect the cycling of carbon (Saffigna et al., 1989; Clein and
313 Schimel, 1995; Stone et al., 2014), for example, Holt (1997) found that MBC was
314 lower in the soils of the area that had been subjected to poor grazing management, it
315 was significantly higher in vegetated soils than in the unvegetated control (Sanullah
316 et al., 2011). Soil depth has a highly significant effect on the microbial communities
317 (Li et al., 2014; Stone et al., 2014). Soil C, MBC are highly correlated with each other
318 across all soil and forest types and depth increments (Stone et al., 2014), it is
319 consistent with our conclusion. Result is that different carbon fraction contents at



320 upper soil layer are higher than at the lower.

321 4.2 Soil enzyme activity of different type vegetations

322 The study results show that enzyme activity at 0-5 cm soil layer is higher than at 5-20
323 cm soil layer, mainly due to the large stocks of litter leaves, plants and animal residues
324 quantity or species at upper layer soil. The soil quality would also affect enzyme
325 activities (Bandick and Dick, 1999; Zhang et al. 2015), adequate nutrients through
326 degrading detritus and well soil air permeability make soil microorganism thriving
327 and enzyme activity higher (Bastida et al., 2013). With the soil layer deepen, soil air
328 permeability go to worse nutrients made microbial metabolism slowly, these wrong
329 soil conditions would affect enzyme activities. The soil microbial activity at upper soil
330 layer is stronger than at the lower, with the microbial activity increasing, the enzyme
331 activity become higher. Surface soil has adequate excreta from plants, animals and
332 microorganisms, the physiological activity of upper layer soil is stronger and make
333 soil released more enzymes. Thereby, enzyme activity declines exponentially with
334 depth (Stone et al., 2014). Different land used treatments has an influence on soil
335 enzyme activity (de Medeiros et al., 2015), and enzyme activity associate with plant
336 litters (Sinsabaugh, 2010). Forest had more plant litters and soil microorganism than
337 the others, soil enzyme activity is higher under forest vegetation. The paper assume
338 that various carbon fractions have different effects on soil enzyme activities.

339 4.3 Relationship between soil carbon fraction and enzyme activity

340 Majority kinds of soil carbon fractions are sources of microorganism, and have
341 different effectiveness. Stone et al. (2014) observed strong and interrelated gradients



342 in soil C, microbial biomass and enzyme activities with depth. Enzymes activities per
343 unit of total organic carbon and MBC are more important in explaining differences
344 between soils than absolute enzyme activities in sandy entisol (de Medeiros et al.,
345 2015). Studies of MBC and enzyme activities provide information on the biochemical
346 processes occurring in the soil and soil biological parameters may have a potential as
347 early and sensitive indicators of soil ecological stress and restoration (Dick et al.,
348 1992; Demisie et al. 2014). Activities of the enzymes are calculated by dividing
349 enzyme activities by the MBC (Waldrop et al., 2000; Waring et al., 2014). Enzymatic
350 activity is related to carbon dynamics in soil and can indicate the incorporation of
351 labile carbon in soil (Kang et al., 1998; Sardans et al., 2008; Lagomarsino et al., 2011).
352 Enzymes participated in the transformation process of soil nutrients, in the soil
353 environment, enzyme activity plays vital role on soil microbial activity and soil
354 quality (Dick, 1994; Mastro et al., 2006). Shao et al. (2015) found that there was no
355 significantly correlated between SOC and urease, while MBC and DOC were
356 significantly positively correlated with urease. Therefore, Enzyme activity and carbon
357 fraction has influence each other on conversion and circulation (Plaza et al., 2004;
358 Mandal et al., 2007).

359 **5. Conclusions**

360 Through analysis and research relationship between soil carbon fractions and enzyme
361 activities of different type vegetation soils in northern Shaanxi Province Loess Plateau,
362 it revealed the influence of different carbon fractions on soil enzyme activities, the
363 conclusions were as followings. First, the concentrations of SOC, MBC, EOC and



364 DOC under different type vegetation soils showed that forest was higher than forest
365 steppe and grassland at the same soil layer. And these carbon fractions concentration
366 of all vegetation type soils appeared that the upper was higher than the lower. Second,
367 the patterns of the enzyme activities were similar to soil carbon fractions. Third,
368 correlation analysis showed that the SOC, MBC, EOC and DOC influenced on the
369 urease activities; SOC, MBC and EOC affected sucrase activities; MBC, EOC and
370 DOC influenced on alkaline phosphatase activities. The CCA reflected the
371 relationship between the sucrase activity and SOC. The contributions from the various
372 forms of carbon fractions as evaluated by the canonical coefficient of CV were on the
373 order of SOC > DOC > MBC > EOC. The enzyme activities were on the order of
374 sucrase > urease > alkaline phosphatase. In conclusion, vegetation type was an
375 important factor influence the variation of soil enzyme activities and carbon fractions
376 on the Loess Plateau.

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564 Table 1



565 The basic information of different vegetation types sampling points

Site	Vegetation type	Dominant vegetation
		<i>Platycladus orientalis(L.) Franco</i>
Fuxian	Forest	<i>Quercus liaotungensis</i> <i>Pinus tabulaeformis</i> <i>Robinia pseudoacacia L.</i> <i>Sophora viciifolia</i>
Ansai	Forest steppe	<i>Robinia pseudoacacia L.</i> <i>Compositae</i> <i>Hippophae rhamnoides Linn</i> <i>Compositae</i>
Lian Daowan	Grassland	<i>Artemisia giraldii Pamp</i> <i>Xeric phragmitesaustralis</i> <i>Thymus mongolicus Romn & Compositae</i>

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568 Table 2
 569 Basic physical and chemical properties of the soils for the four vegetation types

Vegetation type	soil layers	Bulk density (g.cm ⁻³)	Soil pH	Total N (g.kg ⁻¹)	Total P (g.kg ⁻¹)	Soil organic matter (g.kg ⁻¹)
Forest	0-5 cm	0.95±0.17Ba	7.93±0.12Ba	1.81±0.54Aa	0.55±0.04Aa	34.82±12.94Aa
	5-20 cm	1.11±0.07Ba	8.02±0.12Ba	1.20±0.24Aa	0.54±0.02Aa	23.23±5.68Aa
Forest steppe	0-5 cm	1.11±0.05ABb	8.11±0.08ABa	0.89±0.32Ba	0.55±0.02Aa	11.88±2.00Ba
	5-20 cm	1.21±0.03ABa	8.21±0.06ABa	0.49±0.09Ba	0.53±0.03Aa	6.48±0.46Bb
Grassland	0-5 cm	1.26±0.03Aa	8.25±0.04Aa	0.57±0.08Ba	0.46±0.02Ba	8.44±1.31Ba
	5-20 cm	1.25±0.05Aa	8.29±0.04Aa	0.43±0.07Ba	0.45±0.01Ba	6.22±0.88Bb

570 Note: Different capital letters mean significant difference at p<0.05 for the same soil
 571 layer and different vegetation type.

572 Different small letters mean significant difference at p<0.05 for the same vegetation
 573 type and different soil layers (n=72).

574 Table 3
 575 The correlation coefficients between soil labile organic carbon content and enzyme
 576 activities in 0-5 cm
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Correlation coefficients	SOC	MBC	EOC	DOC	Urease	Sucrase	Alkaline phosphatase
SOC	1	.928**	.843**	.579*	.823**	.907**	0.512
MBC		1	.799**	0.537	.787**	.877**	0.57
EOC			1	.702*	.775**	.818**	.588*
DOC				1	.886**	0.568	.727**
Urease					1	.738**	.717**
Sucrase						1	0.478
Alkaline phosphatase							1

578 Note: **. Significant relation at 0.01 levels = *.Significant relation at 0.05 levels

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581 Table 4
 582 The correlation coefficients between soil labile organic carbon content and enzyme
 583 activities in 5-20 cm

Correlation coefficients	SOC	MBC	EOC	DOC	Urease	Sucrase	Alkaline phosphatase
SOC	1	.902**	.855**	0.18	.762**	.759**	0.57
MBC		1	.762**	0.106	.633*	.840**	.656*
EOC			1	0.391	.621*	.593*	.600*
DOC				1	0.414	0.222	0.229
Urease					1	0.503	.680*
Sucrase						1	0.397
Alkaline phosphatase							1

584 Note: **. Significant relation at 0.01 levels = * .Significant relation at 0.05 levels

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586 Table5

587 The canonical correlation coefficients (CCA) between soil carbon fractions and enzyme
 588 activities

No.	Canonical correlation coefficient	Proportion that can be explained (%)						
		Chi-SQ	DF	Sig.	X-CV		Y-CV	
Correlation	significance test				Within-cluster	Between-cluster	Within-cluster	Between-cluster
1	0.964	59.543	12	0.000	0.743	0.690	0.728	0.677
2	0.537	9.273	6	0.159	0.052	0.015	0.076	0.022
3	0.370	2.800	2	0.247	0.122	0.017	0.196	0.027

$U_1 = -0.558X_1 - 0.309X_2 - 0.108X_3 - 0.365X_4$
 $V_1 = -0.500Y_1 - 0.549Y_2 - 0.046Y_3$

589 Note: The CCA was performed using the soil carbon fractions and enzyme activities,
 590 three pairs of canonical variates (CVs) were extracted. The U_1 and V_1 refer to the first
 591 group equation between soil carbon fractions canonical variate (X-CV) and the
 592 enzyme activities canonical variate (Y-CV) which has the highest significant
 593 coefficients of <0.05. The rest two equations of U_2 , V_2 - U_3 , V_3 did not show since its
 594 canonical correlation coefficients were higher than 0.05. The indices of the X-CV were:
 595 SOC (X1), MBC (X2), EOC (X3), and DOC (X4). The indices of the Y-CV were:
 596 urease activity (Y1), sucrase activity (Y2), alkaline phosphatase activity (Y3).

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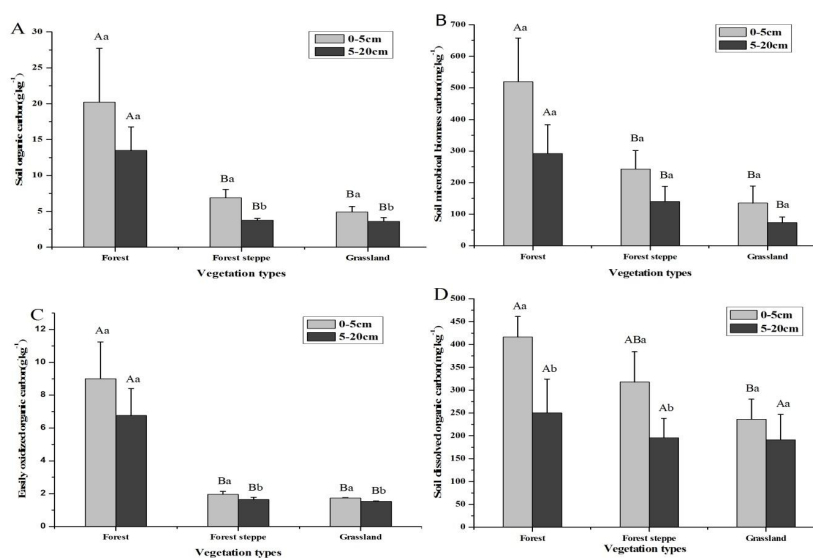
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608 Figure



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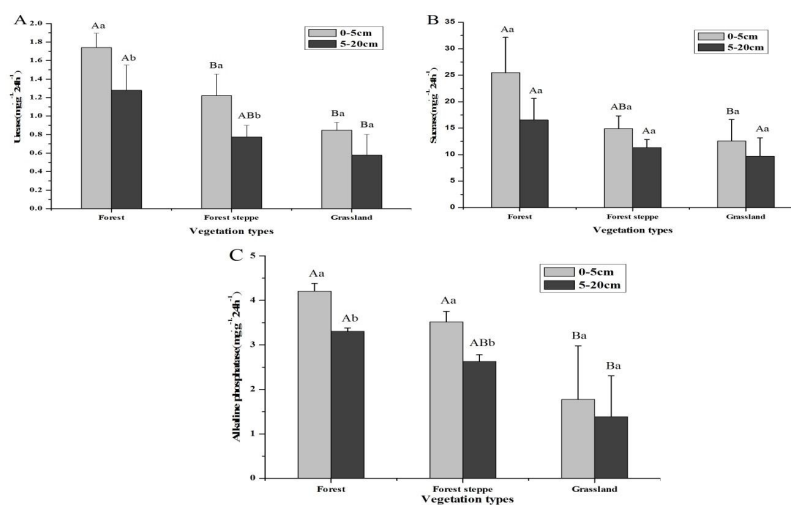
610 Fig.1. SOC (A), MBC (B), EOC (C) and DOC (D) under different vegetation types

611 Note: Different capital letters mean significant differences at $p < 0.05$ for the same soil

612 layer and different vegetation type.

613 Different small letters mean significant differences at $p < 0.05$ for the same vegetation

614 type and different soil layers (n=72).



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616 Fig.2. Soil urease (A), sucrase (B) and alkaline phosphatase (C) activity under
617 different vegetation types

618 Note: Different capital letters mean significant differences at $p < 0.05$ for the same soil
619 layer and different vegetation type.

620 Different small letters mean significant differences at $p < 0.05$ for the same vegetation
621 type and different soil layers (n=72).

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