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Contributions from carbon and nitrogen in roots to closing the yield gap between conventional and organic cropping systems

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Citation:

Córdoba, E. M., Chirinda, Ngonidzashe, Li, F., & Olesen, J. E. (2018). Contributions from carbon and nitrogen in roots to closing the yield gap between conventional and organic cropping systems. *Soil Use and Management*, 1–8 p.

Publisher's DOI:

<http://doi.org/10.1111/sum.12427>

Access through CIAT Research Online:

<http://hdl.handle.net/10568/95869>

Terms:

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1 **Contributions from carbon and nitrogen in roots to closing the yield gap between conventional and**
2 **organic cropping systems**

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9 **Abstract**

10 This study investigates the effect of different crop rotation systems on carbon (C) and nitrogen (N) in root biomass as
11 well as on soil organic carbon (SOC). Soils under spring barley and spring barley/pea mixture were sampled both in
12 organic and conventional crop rotations. The amounts of root biomass and SOC in fine (250-53 µm), medium (425-250
13 µm) and coarse (> 425 µm) soil particulate organic matter (POM) were determined. Grain dry matter (DM) and the
14 amount of N in harvested grain were also quantified. Organic systems with varying use of manure and catch crops had
15 lower spring barley grain DM yield compared to those in conventional systems, whereas barley/pea showed no
16 differences. The largest benefits were observed for grain N yields and grain DM yields for spring barley, where grain N
17 yield was positively correlated with root N. The inclusion of catch crops in organic rotations resulted in higher root N
18 and SOC (g C/m²) in fine POM in soils under barley/pea. Our results suggest that manure application and inclusion of
19 catch crops improve crop N supply and reduce the yield gap between conventional and organic rotations. The observed
20 positive correlation between root N and grain N imply that management practices aimed at increasing grain N could also
21 increase root N and thus enhance N supply for subsequent crops.

22 **Keywords** Particulate organic matter, root carbon, root nitrogen, catch crops, manure, low input system

23 **Introduction**

24 The expansion of resource-intensive (conventional) agriculture has increased productivity of major crops and enhanced
25 global food security (Leifeld, 2012). Although intensive farming systems achieve high crop productivity, meeting
26 environmental goals remains a challenge for conventional crop production systems. Organic cropping systems, which
27 aim to achieve food production without depleting natural resources may be a viable solution (IFAD, 2011). However, the
28 productivity of organic cropping systems is reduced by nutrient limitations, and higher productivity can be obtained by
29 implementing management approaches that enhance soil nutrient availability and increase nutrient use efficiency (Berry
30 *et al.*, 2002).

31 In organic cropping systems, nitrogen (N) may be obtained from animal manure or through strategic inclusion of
32 non-leguminous or leguminous catch crops (Thorup-Kristensen *et al.*, 2003). Non-leguminous catch crops may retain and
33 recycle soil N as they recover surplus N from fertilizer applied to the previous crop or from mineralised soil organic N,
34 preventing it from being lost through several N loss pathways. Leguminous catch crops fix atmospheric N and represent
35 an important renewable source of N that enhances soil fertility for the benefit of subsequent crops, reducing dependence
36 on external N sources (Jensen *et al.*, 2012). Soil-incorporated leguminous and non-leguminous crop residues decompose
37 and contribute nutrients for subsequent crops (Li *et al.*, 2015a).

38 Carbon inputs directly influence soil organic matter (SOM), soil microbial processes and crop productivity
39 (Janzen, 2006). Katterer *et al.* (2011) estimated that C derived from crop roots contributes more to soil C and its stability
40 than that from aboveground residues. Previous studies suggest that different crop management measures (i.e., inorganic
41 fertilizer, organic manure, pesticides, tillage intensity) can affect root biomass (Van Noordwijk *et al.*, 1994; Swinnen *et al.*,
42 1995). Yet, there is growing evidence showing inconsistent management effects on root C inputs (Chirinda *et al.*,
43 2012; Lazicki *et al.*, 2016; Hu *et al.*, 2018). The addition of above- and belowground crop residues to soils increase their
44 particulate organic matter (POM) content (Fronning *et al.*, 2008). Previously, POM (50-2000 μm), which refers mostly to
45 organic matter that has not yet been subject to microbial degradation, has been used as an index of labile SOM, which is
46 also more sensitive to management than total SOM (Carter, 2002). Soil POM content may be considered a predictor of N
47 mineralization potential and thus correlate with the amounts of soil-derived N taken up by crops (Willson *et al.*, 2001).

48 The objective of this study was to determine cropping system effects on C and N in roots and shoots as well as the
49 amount of N returned in above- and belowground residues from cereals and grain legume/cereal intercrops in systems

50 differing in approaches to soil fertility management. Additionally, the study explored whether system differences in soil
51 fertility were related to C in soil POM.

52 **Materials and methods**

53 *Field site and soil description*

54 Field measurements were made in a long-term organic and conventional cropping experiment (established in 1997) at
55 Research Centre Foulum in western Denmark (Olesen *et al.*, 2000). The mean annual temperature at the study site is 7.3
56 °C and average rainfall is 704 mm. The soil type at the site is a sandy loam classified as a Mollic Luvisol according to
57 The World Reference Base for Soils (WRB) with the following chemical and physical characteristics at 0-0.25 m depth
58 at the commencement of the crop rotation experiment: pH 6.5 (CaCl₂), 0.05 g P/kg soil, 0.13 g K/kg soil, 78 % sand, 13
59 % silt, 9 % clay, 23 g SOC/kg soil, 1.8 g total N/kg soil and soil bulk density was 1.35 g/cm³ (Djurhuus & Olesen, 2000).

60 *Experimental layout*

61 The experiment had a factorial design with three factors and two replicate blocks. The experimental factors were 1)
62 different cropping systems, i.e. organic with whole-year green manure (O2) or organic (O4) or conventional (C4) without
63 whole-year green manure, 2) with (+CC) and without (-CC) a catch crop, and 3) with animal manure (+M) or inorganic
64 fertilizer (+IF) and a treatment where fertilizers were excluded (-M). Four-year crop rotations were used between 1997 to
65 2009. All systems were managed as organic until 2004. From 2005 the treatment combination of no manure and no catch
66 crop (-CC/-M) in O2 or O4 was converted to a conventional system (C4) that include application of inorganic fertilizers
67 (+IF) and use of agrochemicals. From 2005, the crops in O4 and C4 were the same, although the species used for catch
68 crops differed, with legume-based mixtures in O4 and non-legumes in C4. During 2005-2009, the C4 and O4 rotations
69 included spring barley (*Hordeum vulgare* L.), faba bean (*Vicia faba* L.), potato (*Solanum tuberosum* L.) and winter wheat
70 (*Triticum aestivum* L.) while the O2 cropping sequence was spring barley with undersown ley, grass-clover, potato and
71 winter wheat. In 2009, the faba bean crop in C4 and O4 was replaced with a mixture of spring barley and pea (*Pisum*
72 *sativum* L.) to avoid soil borne diseases. From 2010, O4 and C4 rotations were changed to spring barley, hemp
73 (*Cannabis sativa* L.), spring barley/pea, spring wheat and potato, and rotation O2 had lucerne (*Medicago sativa* L.)
74 instead of hemp and spring barley/pea. For the current study, three crop sequences were used (Table 1) with either spring
75 barley or spring barley/pea mixture in 2012. The following treatments were included in the organic rotations (O2 and
76 O4): -CC/+M, +CC/-M and +CC/+M. The conventional rotation (C4) included treatments with and without catch crops: -

77 CC/IF and +CC/IF. The size of individual plots was 216 m² and each plot was sub-divided (4 sub-plots of 54 m²) (Olesen
78 *et al.*, 2000; Shah *et al.*, 2017).

79 *Crop management*

80 Spring barley was sown at row distances of 12.5 cm, except for barley/pea in organic systems (24 cm) (Table 2). Sowing
81 was at a target density of 250 plants/m² in O2, 400 plants/m² in O4 and 350 plants/m² in C4 for spring barley and 150/35
82 plants/m² in O4 and 200/45 plants/m² in C4 for barley and pea, respectively, in barley/pea plots. Harvesting was done on
83 20 August 2012.

84 Catch crops in organic rotations were a mixture of perennial ryegrass (*Lolium perenne* L.), chicory (*Cichorium*
85 *intybus* L.), red clover (*Trifolium pratense* L.) and white clover (*Trifolium repens* L.) (10 kg/ha) undersown after weed
86 harrowing. The conventional system (C4/+IF/+CC), had a mixture of winter rye (*Secale cereale* L.) and winter rape
87 (*Brassica napus* L.) (52 kg/ha) as catch crops sown after harvest. All catch crops were sown at a row distance of 12.5 cm
88 and were incorporated by ploughing in spring prior to sowing of spring crops (Table 2). Lucerne was undersown (30
89 kg/ha) in the spring barley after the end of weed harrowing in spring in the O2 rotation and left in the field to complete its
90 growth cycle as a main crop in the following year.

91 The spring barley plots receiving manure were supplied with anaerobically digested slurry (mixture of cattle and
92 pig) in O2 and pig slurry in O4 (Table 2). About 60 kg total N/ha of manure was applied in organically grown spring
93 barley. The barley/pea crop grown in O4 and C4 (*a* and *b*) did not receive any amendments. Spring barley in C4 plots
94 received inorganic fertilizer (122 kg N/ha). Soil incorporation of crop residues was done after harvest.

95 *Plant measurements*

96 In July and August of 2012, shoot and root samples were collected at maturity from plots with spring barley and from
97 plots with barley/pea mixture (Table 1). Shoots were sampled from two 0.5 m² areas and pooled to obtain a
98 representative plot value. We took approximately one kilogram from these samples and separated this sample into barley,
99 pea, catch crop and weeds. The dry matter (DM) content of all plant samples was determined after oven drying at 80 °C
100 for at least 24 h.

101 Root biomass was measured through soil samples collected from within and between crop rows in each
102 treatment plot. The samples were collected after crop harvest in the same area as shoot samples, at 0-0.30 m depth using
103 a hand auger. Composite samples consisting of 3 cores of 5 cm diameter were collected from within and midway

104 between crop rows and pooled according to sampling positions (Bolinder *et al.*, 1997). Soon after sampling, soils were
105 frozen at -18 °C to avoid root decomposition. The roots were processed as described by Chirinda *et al.* (2012). Nitrogen
106 content in finely milled shoot and root samples was assessed using the Dumas method (Hansen, 1989). The root C
107 content was estimated by assuming C concentrations are 45 % of DM biomass. To extrapolate shoot and root biomass
108 and estimated C concentrations from point to field scale, we used a similar approach to Bolinder *et al.* (1997).

109 Crops from each treatment were harvested from two sub-plots (~27 m²) using a combine harvester for grain DM
110 determination. In the barley/pea mixture a sub-sample was used to determine the yield of the respective components.
111 Total N in the pea grain was determined on finely milled samples by the Dumas method, whereas N in barley grains was
112 determined using a near infrared spectroscopy analyzer (Infratec TM 1242 Grain Analyzer, Foss A/S).

113 *POM fractionation and laboratory analysis*

114 The soil sampling strategy was similar to that used for estimating root biomass: samples were collected from between
115 and within rows. Samples stored at -18°C were thawed and fractionated based on size and POM was fractionated as SOM
116 > 53 µm (Marriott & Wander, 2006). The procedure described by Willson *et al.* (2001) was used to process the soil
117 samples. We defined the different fractions of POM-C as coarse (>425 µm), medium (425-250 µm) and fine (250-53
118 µm). The amount of C to 30 cm depth was calculated by multiplying by the bulk density. All three fractions were
119 analyzed for total C by combustion-based elemental analysis (ELTRAs CS-580A “Helios”).

120 *Statistical analyses*

121 Data were first checked for normality, and since all data could be considered normally distributed, no data
122 transformations were applied. The experimental setup applied a division of treatments into sub-blocks, where the three-
123 way interaction between treatments was confounded by the sub-blocks. However, since our selection of treatments did
124 not include three-way interactions, these sub-blocks were not included in the statistical models. All data on barley grain
125 DM, grain N, shoot N, root C and N amounts and the C content in different fractions of POM were analysed using a
126 mixed effect model with treatments as fixed effects and blocks as random effects. Analyses were performed using PROC
127 MIXED of SAS (SAS Institute 1996). Since the experimental layout did not include a balanced design between treatment
128 combinations, we applied four statistical models with different subsets of the data. The first analysis included all data for
129 each crop type and allowed results to be compared between treatment combinations based on Least Significant
130 Difference at P<0.05 (Table 3). In addition, we selected three combinations of subsets to test the overall treatment effects

131 (crop rotation, catch crop, manure, Table 4) as follows: 1) Only organic systems with catch crops to test the effects of
132 rotation and manure for spring barley, 2) Both conventional and organic systems with manure and fertilizer to test the
133 effects of rotation and catch crops for spring barley, and 3) Both crop types for conventional and organic systems with
134 manure/fertilizer to test the effects of crop type and catch crop.

135 In addition, correlation analyses were performed between grain DM and grain N yield and root and soil POM variables to
136 determine relationships for treatment means using PROC CORR in SAS.

137 **Results**

138 *Grain DM and grain N yields*

139 When all treatment combinations for spring barley were compared, the conventional rotations had higher grain DM
140 yields irrespective of catch crop inclusion than the organic rotations, except for O2, which included both catch crops and
141 manure. The latter also had higher grain DM yields than the two O4 rotations, which included either catch crops or
142 manure (Table 3). When only organic rotations were compared, the presence of manure significantly increased spring
143 barley grain DM yields (Table 4). Moreover, the presence of catch crops significantly increased spring barley grain N
144 yields and shoot N, and it tended to increase spring barley grain DM and shoot C (Table 4). The corresponding grain DM
145 yield benefits from catch crops were 30 and 23 g DM/m² in the O2 and O4 rotations, respectively.

146 The average grain DM benefit from presence of catch crops was 102 and 36 g DM/m² in the O4 and C4
147 rotations, respectively. The largest benefits from manure and catch crops were obtained for spring barley, whereas there
148 was little effect for the barley/pea mixture (Table 4).

149 *Root N and its relation to grain N*

150 The amount of N in root was significantly affected by crop and rotation system (Table 4). On average, the amounts of N
151 in roots from the O4 and C4 cropping systems were 3.3 and 2.6 g N/m², respectively, for barley/pea, and 2.1 and 3.0 g
152 N/m², respectively, for spring barley (Table 3).

153 Mean root N was 3.0 and 2.3 g N/m² in barley/pea and spring barley, respectively. Root N in barley/pea in O4
154 increased by 0.45 g N/m² with the presence of catch crops. For spring barley, all rotations tended to have higher root N
155 when the crop was grown in systems with catch crops (Table 3). For spring barley, the root N benefits from catch crops

156 were 0.65, 0.95 and 0.60 g N/m² in rotations O2, O4 and C4, respectively. A positive correlation was observed between
157 N harvested in grain and N in spring barley roots (Fig. 1). This correlation was not found for the barley/pea crop.

158 *Particulate organic matter in soil*

159 Total POM-C contents in the O2 and O4 organic rotations were similar for soils under spring barley (Table 3). For
160 barley/pea plots, total POM-C contents were similar across crop rotations. Similarly, POM-C contents in the coarse,
161 medium, and fine fractions were similar across systems for both crops, except barley/pea, which had higher fine POM-C
162 in the conventional rotations with catch crops than without catch crops.

163 *Root C and total N recycling*

164 Root C tended to be higher under organic (O4) than conventional (C4) crop rotation for barley/pea (Table 4) and was 84
165 and 64 g C/m², respectively (Table 3). By contrast, it was similar among treatments for spring barley and averaged 72 g
166 C/m². For barley/pea, there was a higher root C in the organic (O4) compared to the conventional (C4) rotation (Table 3).
167 Overall, root C contents were similar across both crops (Table 4). Our results allow the estimation of N recycled in the
168 above- and below-ground residues of barley and barley/pea crops. For barley/pea an average amount of 119 kg N/ha was
169 returned in crop residues, composed of 89 and 30 kg N/ha in above- and belowground residues, respectively. There were
170 no pronounced differences between cropping systems in these amounts. This should be compared with an average value
171 of 65 kg N/ha in residues from spring barley, where 41 and 24 kg N/ha originated from above- and belowground
172 residues, respectively.

173 **Discussion**

174 *Productivity of cropping systems*

175 Several studies have shown beneficial effects of intercropping grain legumes with cereals, because this can increase and
176 stabilize yields compared to sole grain legume crops, and it also reduces the need for inputs (e.g., fertilizer and
177 agrochemicals for pest and weed control) compared to sole cereal crops (Hauggaard-Nielsen & Jensen, 2001; Sahota &
178 Malhi, 2012). Our results showed that grain N yields and shoot N of the barley/pea intercrop were generally considerably
179 larger than when spring barley was a sole crop. Similar results have previously been reported for intercropping of barley
180 and pea (Sahota & Malhi, 2012; Hunady & Hochman, 2014). We observed that grain DM for spring barley responded to
181 the different crop systems (organic/conventional, manure and catch crop), whereas this was not the case for barley/pea.
182 This corroborates with previous studies reporting that a grain legume-cereal mixture provided more stable yields

183 (Hunady & Hochman, 2014). Therefore, intercropping and catch crop inclusion may both contribute towards stabilizing
184 yields in organic systems and closing the yield gap between organic and conventional systems.

185 While a lack of effective weed and pest control measures has been given as the reason for lower grain DM
186 yields in organic systems (Clark *et al.*, 1998; Melander *et al.*, 2016), others report that deficient nutrient supply
187 (especially N and phosphorus) often constitutes the major limitation (Shah *et al.*, 2017). A key difference in N
188 management is related to the fact that unlike the more targeted fertilizer use in conventional systems, organic cropping
189 systems are often characterized by poor synchrony between nutrient supply and plant demand (Berry *et al.*, 2002). In the
190 current study, the effect of catch crops and the application of manure were evaluated for both organic and conventional
191 rotations. The legume-based catch crops in the organic cropping systems contributed N to the cropping systems through
192 both biological N fixation (Li *et al.* 2015b) and through retaining N by recycling this in above- and belowground plant
193 residues.

194 The highest spring barley yields (DM and N) in the organic systems were obtained when both manure and catch
195 crops were used (Table 3). Both factors contribute to crop N supply, albeit in different ways. The application of manure
196 adds a substantial amount of mineral N as ammonium, whereas the N from the catch crops is supplied over a longer time
197 through mineralization of organic matter (Olesen *et al.*, 2007; Doltra & Olesen, 2013). For spring barley, enhanced soil N
198 availability following incorporation of catch crops prior to sowing and application of manure probably explains the yield
199 similarities between the low-input organic and the high-input inorganic fertilizer based systems. These findings are
200 consistent with those from previous studies in which the use of catch crops and application of animal manure showed
201 positive effects on grain DM yields (Olesen *et al.*, 2007; Chirinda *et al.*, 2010). Moreover, the larger grain yields from O2
202 compared with the O4 rotation suggest a positive response to the inclusion of grass-clover green manure in the rotation
203 (Olesen *et al.*, 2007). Nonetheless, it is important to note that prior to 2005, the current conventional plots had not
204 received fertilizer or manure and did not include a catch crop. It is reasonable to assume that this system was, to some
205 extent, depleted in readily degradable organic N when converted to conventional management.

206 *N inputs by roots*

207 No significant differences in root N content were observed between crop rotation systems, but there was a tendency for
208 higher values for the barley/pea intercrop compared to spring barley. In addition, the inclusion of catch crops in organic
209 systems led to higher shoot and root N input. This is simply a consequence of the presence of more plant biomass in

210 systems where pea and catch crops were included, and this indicates a larger N accumulation capacity in organic systems
211 with inclusion cereal-legume crops and catch crops.

212 Grain DM yields have been reported to closely relate to the growth and development of root systems in high
213 yielding cropping systems (Wang *et al.*, 2014). However, Hu *et al.* (2018) found no relationship between shoot and root
214 biomass of cereals when comparing organic and conventional systems. This lack of difference in root biomass and root C
215 between cropping systems of different intensities was also found in our study (Table 3). We found a linear relationship
216 (Fig. 1) between root N and grain DM for spring barley, but not for the barley/pea intercrop. This relationship was caused
217 by differences in N concentration in roots of different systems (15.0 g N/g DM in spring barley versus 17.9 g N/g DM in
218 barley/pea), rather than differences in biomass (data not shown). The lower N availability in the organic systems was thus
219 found to result in lower N concentration in root biomass, which would delay net N mineralization of these residues
220 (Thomsen *et al.*, 2016).

221 *Residual N effects of spring barley and barley/pea*

222 The productivity of organic cropping systems depend on the recycling of nutrients through crop residues. We observed
223 considerably higher amounts of N in residues following barley/pea compared with spring barley. This may be expected to
224 enhance the risk of N leaching losses during autumn, but such differences in N leaching between spring barley and
225 barley/pea were not found by Askegaard *et al.* (2011). Other previous studies (e.g., Bergström & Kirchmann, 2004)
226 show that when legumes instead of ryegrass are included in cropping systems, leaching of N increase considerably over
227 the short-term. In the case of our study, it is likely that barley/pea contributes to the long-term fertility of the cropping
228 system rather than short-term losses. These temporal aspects of N release are affected by the N concentration and C:N
229 ratio of the crop residues (Jensen *et al.*, 2005), and the root N concentrations of both barley/pea and barley roots were so
230 low that they would likely cause net N immobilization.

231 *Crop management implications on POM*

232 The amounts of POM-C strongly depend on organic matter inputs, which are influenced by cropping systems and
233 management practices. In this study, if we consider all crop residues, there were no pronounced differences in average
234 organic matter inputs between different management systems (data not shown). Fine POM-C (250-53 μm) was only
235 significantly higher in soils under barley/pea in the organic rotation that included catch crops and manure, and in
236 conventional systems that included catch crops, than in the conventional system that excluded catch crops. As fine POM
237 is a biologically and chemically active part of the easily decomposable organic matter, the high amount of fine POM-C in

238 rotations including catch crops may be the reason for the higher soil respiration from rotations, including catch crops, as
239 reported in a previous study conducted on the same experiment (Chirinda *et al.*, 2010). However, this higher soil
240 respiration may be associated with higher N mineralization that increases the risk of N loss through leaching, which thus
241 emphasizes the need for maintaining the use of catch crops to retain N in the system.

242 As observed by Marriott & Wander (2006) for a study conducted on nine farming system trials, similar POM-C
243 (>53 μm) was observed in organic and conventionally managed systems. We also found little difference between
244 cropping systems in POM-C. The POM-C may therefore not in itself be a good indicator of soil fertility, but may have to
245 be supplemented with indicators that better reflect nutrient contents and nutrient availability, in particular N
246 mineralization potential.

247 **Conclusions**

248 Our results suggest that manure application and inclusion of catch crops improves crop N supply and reduces differences
249 between yields in conventional and organic rotations, resolving some of limiting factors of organic systems such as low
250 N supply. Although application of manure in organic systems reduces differences in yields between conventional and
251 organic systems, limitations in availability of manure from livestock farms may challenge this option for enhancing N
252 inputs in organic systems. The observed positive correlation between root N and grain N for spring barley suggests that
253 high yielding cereal crops may be contributing more N to soil fertility. These findings suggest that grain N can be used as
254 a proxy for N in root N residues and thus the amount of residual N available for subsequent crops. Overall, targeted
255 management of manure application and catch crop inclusion combined with knowledge on root derived residual N should
256 be able to improve crop yields and sustainability of organic cropping systems.

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259 **Acknowledgments**

260 E. Córdoba has a contract from IFAPA (III Programa de Incorporación de Personal Investigador) and co-financed by the
261 European Social Fund (Programa Operativo FSE de Andalucía 2007-2013). The authors gratefully acknowledge the
262 support of the RowCrop project funded under Organic RDD2 by Danish Ministry of Environment and Food, and the
263 FertilCrop project funded under Core Organic Plus by Danish Ministry of Environment and Food.

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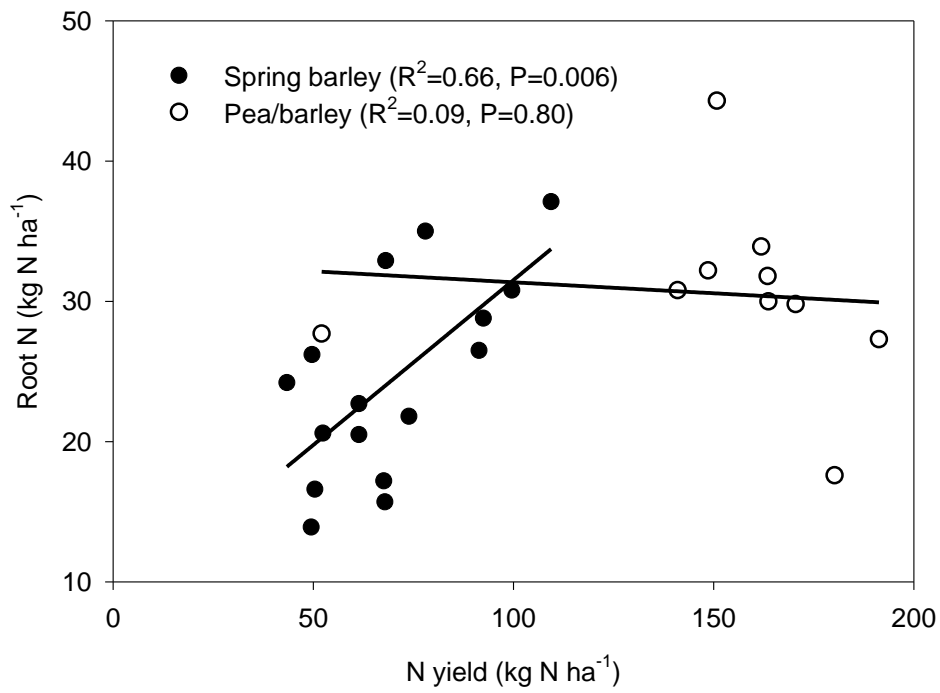


Fig. 1: Relationships between root and grain nitrogen for pea/barley and barley only cropping systems for samples collected in 2012. The statistics are shown for linear regressions of root N on N yield.