Iron Bioavailability in Low Phytate Pea

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Abstract:

The objectives of this study are to determine the effect of genotype and environment on iron bioavailability in a set of five pea varieties differing in phytate concentration using the *in vitro* digestion/Caco-2 human cell assay (Glahn 2009), to determine whether iron bioavailability in field pea is heritable by evaluating recombinant inbred lines (RILs) differing in phytate concentration using *in vitro* digestion/Caco-2 human cell assay, and to determine the effect of the pea low phytate trait on chicken performance and iron bioavailability in chicken. In a previous study, two low phytate pea lines (1-2347-144 and 1-150-81) were developed from CDC Bronco at the Crop Development Centre, University of Saskatchewan (Warkentin et al. 2012). As a powerful chelator of iron, phytate can reduce the iron bioavailability in diets. The low phytate peas may have increased iron bioavailability compared to the normal phytate peas. In the first objective of this project, the iron bioavailability of pea seeds of the two low phytate lines, their parent CDC Bronco and two other popular pea varieties in western Canada (CDC Meadow and CDC Golden), derived from 3 replicate field experiments conducted in 2009 and 2010 at SPG (Saskatchewan Pulse Growers land), Outlook and Rosthern, were assessed using the *in vitro* digestion/Caco-2 cell culture. The result shows that the iron bioavailability of the two low phytate lines is significantly higher than the other three normal phytate varieties, although their iron concentrations have not significant difference. The low phytate line 1-2347-144 and CDC Meadow were crossed to develop RILs.

Preliminary results:

In the first objective, soluble iron concentration was tested using atomic absorption spectroscopy (AAS). Soluble iron concentration is not equal to iron bioavailability, because the highly soluble form of iron, like mono-ferric phytate, can have little availability. In each environment (2009 SPG, 2009 Outlook, 2009 Rosthern and 2010 Rosthern), there was no significant difference in iron concentration between these five varieties (Table 1).

Table 1. Iron concentration (ppm) of 5 varieties grown in field trials at SPG, Outlook, and Rosthern in 2009, and at Rosthern in 2010.

Variety	2009 SPG	2009 Outlook	2009 Rosthern	2010 Rosthern
1-150-81	42.7 a	39.3 a	40.9 a	38.1 a
1-2347-144	40.6 a	37.4 a	40.9 a	40.4 a
CDC Bronco	43.1 a	34.9 a	40.0 a	39.4 a
CDC Golden	44.7 a	35.2 a	44.2 a	42.1 a
CDC Meadow	44.2 a	38.4 a	43.7 a	40.5 a
Mean	43.1	37.0	41.9	40.1
$LSD_{0.05}$	5.3	3.8	3.1	4.1

The inorganic phosphorus concentration was determined using modified Chen's reagent method (Chen et al. 1956). Table 2 shows that the concentration of inorganic phosphorus in the two low phytate lines was approximately 3 times greater than the concentration in the other varieties in each environment.

Table 2. Inorganic-P concentration (µg/gm) of 5 varieties grown in field trials at SPG, Outlook, and Rosthern in 2009, and at Rosthern in 2010.

Variety	2009 SPG	2009 Outlook	2009 Rosthern	2010 Rosthern
1-150-81	67 a	89 a	67 a	89 a
1-2347-144	64 a	82 a	61 a	83 a
CDC Bronco	19 b	20 b	19 b	19 b
CDC Golden	22 b	22 b	21 b	20 b
CDC Meadow	25 b	22 b	24 b	19 b
Mean	39	47	38	46
$LSD_{0.05}$	10.4	6.6	9.4	7.9

Phytate concentration was tested using Modified Wade's reagent method (Gao et al. 2007). Table 3 indicates that the two low-phytate lines contained approximately 0.5 to 0.7 times the phytate concentration of the normal phytate varieties.

Table 3. Phytate concentration (µg/gm) of 5 varieties grown in field trials at SPG, Outlook, and Rosthern in 2009, and at Rosthern in 2010.

Variety	2009 SPG	2009 Outlook	2009 Rosthern	2010 Rosthern
1-150-81	72 b	73 b	87 b	101 c
1-2347-144	64 b	67 b	85 b	94 c
CDC Bronco	129 a	146 a	142 a	154 ab
CDC Golden	142 a	145 a	146 a	163 a
CDC Meadow	133 a	137 a	150 a	141 b
Mean	108	114	122	131
$LSD_{0.05}$	17.9	8.2	11.8	9.4

Figure 1 below indicates that iron bioavailability of the two low-phytate lines (1-150-81 and 1-2347-144) was 1.4 to 1.9 times higher than the other three normal phytate varieties (LSD = 4.3, 2.52, 2.96, 2.23 respectively).

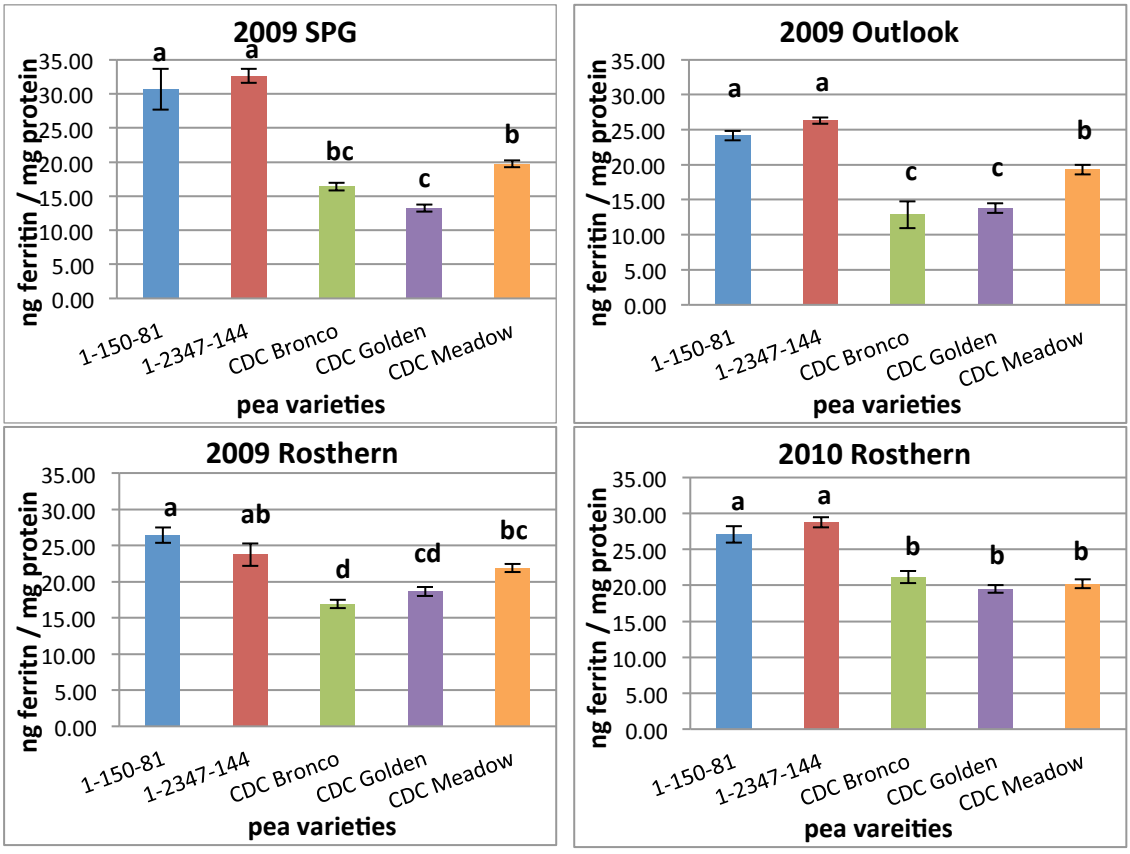

Figure 1. Mean iron bioavailability (ng ferritin/mg protein) of 5 varieties grown in field trials at SPG, Outlook, and Rosthern in 2009, and at Rosthern in 2010.

In conclusion, filed pea lines 1-150-81 and 1-2347-144 had lower phytate concentration, higher inorganic phosphorus concentration, and greater iron bioavailability in human cells

than normal phytate varieties. In this case, low phytate is a technique for biofortification of the pea crop.
In the future study, the iron bioavailability of RILs developed from a cross between 1-2347-144 and CDC Meadow will be tested, and the iron bioavailability of low phytate and normal phytate pea varieties will be conducted in an *in vivo* chicken feeding study.

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