Iron Bioavailability in Low-phytate Pea

Xiaofei Liu[(1)], Hank Classen[(2)], Kirstin Bett[(1)], Raymond Glahn[(3)] and Thomas D. Warkentin[(1)]*

(1) Crop Development Centre/Department of Plant Sciences, University of Saskatchewan
(2) USDA-ARS, Cornell University, Ithaca, NY, USA.
(3) Department of Animal and Poultry Science, University of Saskatchewan,
* Corresponding author: tom.warkentin@usask.ca

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Abstract:

Phytate is the main storage form of phosphorus in the seeds of most crops. Phytate is not well digested by monogastrics and it chelates iron, zinc and some other micronutrients. To increase the nutritional value of pea seeds, two low phytate lines (1-150-81 and 1-2347-144) were developed from CDC Bronco in previous research. In this study, an *in vitro* digestion/Caco-2 cell culture bioassay was used to simulate the iron absorption of peas in humans, as the cell line originated from human colon adenocarcinoma cells. The iron bioavailability of the two low-phytate lines was 1.4 to 1.9 times higher than that of three normal phytate varieties, while having the same total iron concentration. *In vivo* studies were used to evaluate iron absorption of chickens fed low phytate and normal phytate pea diets. The diets containing the two low-phytate pea lines had no significant effect on chicken body weight and hemoglobin level, compared with the diets containing normal phytate pea cultivars, however, iron deficiency was suspected in all diets used.

Introduction:

Field pea is an annual cool-season herbaceous legume crop. Canada is the leading field pea producing country. Saskatchewan is the top producing province of field pea within Canada[1]. Field pea contains high level of protein, lysine, slowly digestible starch and fibres. However, it also contains phytate, a main storage form of phosphorus in seeds, which is also a strong chelator which can bind with K, Zn, Fe and some other micronutrients. Phytate is poorly digested by human and monogastric animals, due to the limited enzyme, phytase. Iron (Fe) is an essential element for human, iron-deficiency and iron-deficiency anemia can cause poor work performance and decreased immunity.

In previous study, two low-phytate lines (1-150-81 and 1-2347-144) were developed from CDC Bronco[2], which are confirmed to have increased inorganic phosphorus level. In this paper, two studies are included to evaluate if the two low-phytate lines also have additional benefit to increase iron bioavailability.

Materials and Methods:

The first study is to determine the effect of genotype and environment on iron bioavailability in varieties differing in phytate concentration. Two low-phytate lines (1-150-81 and 1-2347-144) and other three normal phytate cultivars (CDC Bronco, CDC Golden and CDC Meadow) were tested to compare the concentration of inorganic-phosphorus, phytate-phosphorus, and total iron using modified Chen's reagent method[3], modified Wade's reagent method[4] and atomic absorption spectroscopy (AAS), respectively. The iron bioavailability was tested using the in vitro digestion/Caco-2 cell culture model[5].

The second study is to determine the effect of the pea low phytate trait on chicken performance and iron bioavailability in chicken. The same two low-phytate lines (1-150-81 and 1-2347-144) and the other two normal phytate cultivars (CDC Bronco and CDC Meadow) were mixed in the diets respectively with total 4 treatments. Birds were distributed using RCBD with six replications per treatment and 4 birds per replication. Before day 7, the birds were fed with commercial diet and the treatments (ingredients: Peas: 87.01%, Canola oil: 7.2%, Salt: 0.44%, Calcium Phosphate: 0.67%, Limestone: 2.1%, Vit/Min premix (FE free): 0.5%, Choline Chloride: 0.1%, Celite: 1.5%, DL Methionine: 0.48%) started from day 7 and end on day 35. In the end of each week the bird body weight and feed intake were recorded and blood samples were collected for hemoglobin test.

Results and Discussion:

In study one, the concentration of inorganic-phosphorus in the two low phytate lines was approximately three times greater than in the other varieties in each environment, meanwhile, the two low-phytate lines contained approximately 0.5 to 0.7 times the phytate-phosphorus concentration of the normal phytate varieties. The iron bioavailability of the two low-phytate lines was 1.4 to 1.9 times higher than that of three normal phytate varieties, while having the same total iron concentration. Also environmental effect on iron bioavailability is significant (p-value=0.0153).

The chicken study showed that there was no significant difference of bird's body weight and the hemoglobin level among four treatments. Meanwhile, the mortality rate and the sick bird rate were higher compared with other trials which were done previously. Meanwhile the hemoglobin level is around 9.1g/dl, which is also lower than 10.5 ~11.5g/dl in the study of Hoekenga et al. (2011)[6]. Based on the symptom of the sick birds, an iron-deficiency was believed that happen to the birds due to no additional iron added in diet.

So the next steps of this study will be to determine the ideal iron level in the diets for iron bioavailability comparison, and to test another objective that iron bioavailability is heritable in RILs (PR-15) (1-2347-144 X CDC Meadow).

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