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Communication

A New Approach for Improving the Nutritional Quality of Soybean (*Glycine max* L.) with Iron Slag Coating

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Abstract: Iron slag, a byproduct of the steel manufacturing process with a high amount of iron (Fe), magnesium (Mg), manganese (Mn) and zinc (Zn), was used as a seed coating material to improve soybean nutrient quality and maintain yield during cultivation. Soybean yield (grain, aboveground, roots) did not differ significantly from the non-coated seeds, but nutrient concentration in soybeans, such as nitrogen, magnesium and manganese, were significantly increased in the iron-coated treatment, by 6%, 20% and 17%, respectively, than in the non-coated seeds. The application of iron slag as a protective seedcoat improved the nutrient concentrations of soybean seeds after harvest and maintained a good yield, implying that the material could be applied worldwide to improve the nutritional quality of soybeans in large scale production.

Keywords: magnesium; manganese; iron seed coating; n fixation

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1. Introduction

Soybean [*Glycine max* (L.) Merr.] is one of the most important crops as it is one of the main sources of vegetable oil and protein [1,2]. Global soybean yield has continuously increased over the last century, caused by agricultural practice improvement and genetic engineering of high-yielding cultivar, but soybean yield has still not reached a plateau [3,4]. In order to meet the increasing food demand due to high population growth, yet facing limited area for agricultural lands, the improvement of high nutritional-yielding soybean remains a challenge. For example, multiple agricultural management techniques were implemented to achieve these aims, which include increasing nitrogen fixation, optimizing carbon utilization, adjusting soybean development process, and improving photosynthetic efficiency [1,5,6].

Seed-coating technology is among the modern technologies used to increase soybean yield, and it has developed rapidly during the past two decades, specifically for large-seeded agronomic and horticultural crops [7]. In some seed coating techniques, several materials such as nutritional elements, plant growth regulators, chemicals, fertilizers, and pesticides are added to the seeds using adhesive agents to increase seed germination and performance [8]. For example, rice seeds were characterized as resistant to sparrow attack and seed-borne diseases when seeded onto the puddled soil surface by broadcasting, row seeding, or hill seeding after the pre-germinated seeds were granulated using a mixture of reduced iron (Fe) powder and calcined gypsum [9]. Rust (Fe oxide), the oxidation product of reduced Fe, served as a binder which oxidized Fe in the coating layer and was composed of 0.97 to 0.99 kg kg^{-1} amorphous Fe oxides and 0.01 to 0.03 kg kg^{-1} a, b, g-Fe oxyhydroxide and magnetite. It was assumed that amorphous Fe oxides operates as a binder that does not dissolve in water, preventing the collapse of the coating in water. Aside from seed coating, slag-based silicon fertilizers have beneficial effects on the growth and disease resistance of rice. For example, slag-based fertilizer used in Si-deficient paddy soil improved both rice productivity and brown spot resistance, in which the immobile silicon deposited in host cell walls and papillae sites acted as the first physical barrier for fungal penetration, while the soluble Si in the cytoplasm enhanced physiological or induced resistance to fungal colonization [10].

The iron-slag seed coating technique is the one of most popular soybean coating methods in South Korea, for it is able to reduce bird-inflicted damage on the seeds before germination due to the hardness of soybean skins [11]. Soybean iron-slag coating does not only reduce bird-inflicted damage on newly sown soybean, but may have a positive impact on soybean growth characteristics and nutrient contents because of the high micronutrients present in the raw iron-slag materials such as magnesium, manganese, zinc, and others. We report a new finding on the potential use of iron slag as a soybean seed coating material, specifically its ability to improve the nutrient concentrations of soybean seeds at harvest. The objective of this study was to investigate the effect of an iron-slag coating technique on the soybean growth characteristics during cultivation, and the changes in nutrient contents of soybean seeds after harvest.

2. Materials and Methods

2.1. Soybean Coating

Soybean seeds (cv. Merr) were coated with iron coating material (Segi Global, South Korea) using a seed coating machine (BG-SC200, Hwangso machinery Inc., Seoul, South Korea). The soybean was coated with the iron coating material (Segi Global, Seoul, South Korea) using a seed coating machine (BG-SC200, Bulls, Seoul, South Korea). One (1) kg of soybean seeds was totally mixed with a 30-mL binding material (Saechong, Thiram 26.5%, Hankooksamgong, Seoul, South Korea) using a coating machine, and then mixed with 130 g of iron coating material for 30 min with water spraying to prevent exothermic reactions. The coated soybean seeds were air-dried in a well-shaded area for three days and sprayed with water two times a day.

2.2. Soybean Cultivation

The greenhouse experiment was conducted in Gyeongsang National University Farm, Jinju, South Korea, in which the composite soil (fine, silty, mesic typic Endoaquepts) was placed (15 kg dried soil) into Wagner pots (1/2000 na size) with river sand mixture (70% soil, 30%, ww^{-1}) for easy sampling of root and root nodules. A total of 12 experimental pots were installed, with 6 for seed-coating and 6 for no seed-coating. For each pot, three coated seeds were directly sown at the 5 cm soil depth on 15 June 2021, and harvested on 20 October 2021. Fertilizers were applied similarly for each treatment before sowing, following the standard fertilizer recommendation for soybean cultivation in Korea at the rate of $N{:}P_2O_5{:}K_2O$ (30:30:32 kg ha^{-1}), [12]. Urea, superphosphate, and potassium chloride were used as fertilizers for N, P_2O_5, and K_2O, respectively.

Analysis of Soil, Plant Tissue and Statistical Properties

Fresh soil was collected before and after soybean cultivation and dried at 105 °C for 72 h using a dry oven, and then sieved with a 2 mm sieve. Chemical properties of soils and iron coating material were determined as follows: soil carbon and nitrogen were analyzed by CHNS Analyzer (CHNS-932, Leco, Benton Harbor, MI, USA), and the exchangeable micro nutrients (Fe, Mg, Ca, Mn, and Zn) were extracted using 1 M ammonium acetate (pH 7.0) solution (1:5 = soil:solution, ww^{-1}) and quantified using ICP-OES (PerkinElmer Model Optima 4300 DV, Shelton, CT, USA). The SAPD readings were taken with a chlorophyll meter (SPAD-502, Minolta, Japan) with a 30 day interval from 20 days after seedling. The plant dried (60 °C for 72 h) weight was measured after harvest for above biomass, soybean yield, and root biomass. The plant chemical properties were also analyzed after oven-drying and then acid digestion of plant tissue samples was performed for the plant

C/N and micronutrient contents, respectively. Statistical analysis was carried out using the SPSS 23 (IBM SPSS, Chicago, IL, USA) with a significance level of $p = 0.05$ and different letters denoting significant differences at the $p < 0.05$ level between treatments. Principal components analysis (PCA) was conducted to explore the effects of the iron coating technique on multivariate crop parameters (yields and micronutrients) in the R statistical program (version 4.1.3), using the Devtools Package.

3. Results

3.1. Properties of Soil and Iron Coating Material

The composite soil represented the typical Korean upland soil, which contained 8.2 and 0.65 g kg^{-1} carbon and nitrogen contents, respectively (Table 1). Before the study, the extractable micronutrient concentrations were 1.3, 51.3, 7.9, and 6.4 mg kg^{-1} for Fe, Mg, Mn, and Zn, respectively. Iron seed coating material had significantly higher extractable micronutrient concentrations than the composite soil. For example, iron seed coating material contained 121, 2.3, 5.5, and 65.8-times the amount of extractable nutrients than the experimental soil for Fe, Mg, Mn, and Zn, respectively (Table 1). After soybean cultivation, there were no significant differences ($p > 0.05$) in terms of soil properties between the control and coated soil treatment. The coated soil showed lower extractable Mg and Mn concentrations but not significant ($p > 0.05$) against the control after soybean cultivation.

Table 1. The results of carbon, nitrogen, and extractable cation concentrations in before-experiment soil, seed coating agent, and after-experiment soil.

Materials	C	N	Fe	Mg	Mn	Zn
	(g kg^{-1})		(mg kg^{-1})			
Before experiment						
Experimental soil	8.2	0.65	1.3 ^{b}	51.3 ^{b}	7.9 ^{b}	6.4 ^{b}
Coating material	-	-	158.3 ^{a}	120.2 ^{a}	43.8 ^{a}	421.2 ^{a}
After experiment						
Control soil	7.9	0.66	1.3	50.7	7.8	6.2
Coated soil	7.7	0.69	1.3	49.7	7.7	6.3

Different letters indicate significant differences at level of $p < 0.05$ between materials before experiment.

3.2. Crop Growth Properties and Soybean Nutrition Contents

Plant yield properties did not differ significantly ($p > 0.05$) between the no coating and coating treatments. For example, soybean grain, above ground biomass, and root biomass were slightly higher with iron-slag seed-coating treatment, but did not statistically differ from the control (Figure 1). However, the SPAD value and grain nutrient concentrations were significantly ($p < 0.05$) higher in the seed coating treatment than in the non-coated seeds (Table 2). The SPAD value did not differ at 26 DAS in the seed coating treatment, but the latter gave significantly ($p < 0.001$) higher SPAD value than the control at 72 DAS. Nutrient concentrations in the soybean grain varied significantly ($p < 0.05$) in the iron-coated, which were higher by 20%, 15%, and 6% for Mg, Mn, and N, respectively, than the non-coated seed treatment. Soybean Mg concentration was highly correlated with Mn concentration, root biomass, and SPAD value (72 DAS) and correlated negatively with Ca and Zn concentrations in the soybean grain after harvest (Figure 2).

Table 2. Result of SPAD and grain nutrition contents under different seed coating methods.

Treatment	SPAD		Fe	Mg	Ca	Mn	Zn	C	N
	26DAS	72DAS	(mg kg^{-1} grain)					%	
Control	41.7	22.2 ^{b}	170	2285 ^{b}	2266	26 ^{b}	49	48.5	3.02 ^{b}
Coated	41.3	37.2 ^{a}	173	2749 ^{a}	2169	30 ^{a}	49	48.0	3.20 ^{a}

Different letters indicate significant differences at level of $p < 0.05$ between control and coated treatment.

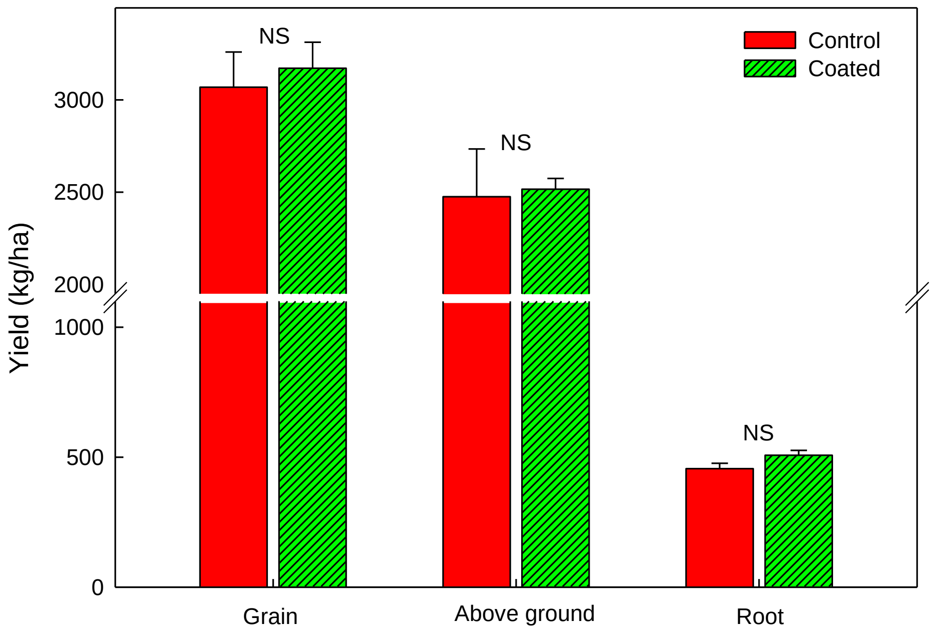

Figure 1. Changes of yields (grain, above ground, and root) resulting from different seed coating methods. NS means not significantly differing between seed coating methods ($p > 0.05$).

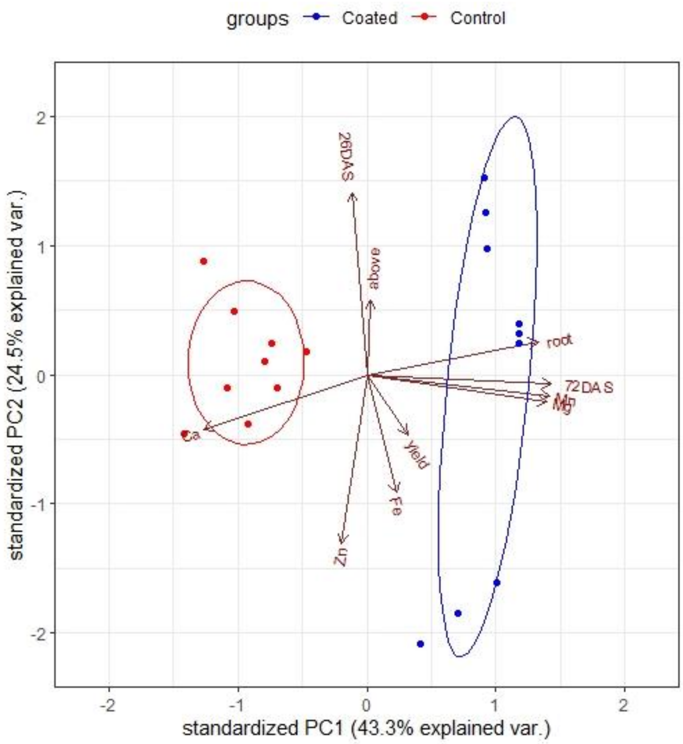

Figure 2. Principal components analysis of soybean yields and nutrition properties between seed coated and control treatments. Above; above ground biomass, root; root biomass, yield; soybean grain yield, 26DAS; SPAD value of 26 days after seedling, 72DAS; SPAD value of 72 days after seedling.

4. Discussion

Seed coating methods are widely used worldwide with many advantages such as protecting soybean seed [13], reducing bird-eating damage [14], and increasing cold tolerance [15]. The soybean yield has been increased by various seed coating methods in many studies, but our study showed no significant differences in soybean yield between the seed-coated and non-seed coated treatments (Figure 1). As soybean was cultivated in the greenhouse, no damage was inflicted by the birds during the seed germination stage, which could be unavoidable if the soybeans were directly cultivated in the field as in the previous studies in Korea [11]. However, the iron coating method significantly increased nutrient concentrations in soybean such as Mg, Mn, and N compared with the non-coated seeds (Table 2), which might have been impacted by the high Mg content of the iron-slag coating material. Magnesium plays a particular role in the transport of carbohydrates from leaves to sink organs, as enhanced starch accumulates in source leaves and lower starch concentrations are found in the sink organs [16,17]. In particular, phloem loading of sucrose is an active and carrier-mediated function driven by proton pumping ATPase in the plasma membrane on the sieve tube cells [18], and the ATPases involved have an absolute requirement for Mg [19]. Magnesium affects not only the utilization, but also the synthesis of the ATP [20]. Peng et al. [21] reported that Mg promoted soybean growth and increased Mg concentration under N-limited conditions mainly derived from the soybean's higher ability to nodulate. Magnesium is reported to be important in the metabolism of rhizobia by facilitating the alteration of carbohydrate partitioning and transport into nodules, and its supply in the plants altered neither nodule structure nor Mg homeostasis, but remarkably promoted nodule enlargement, resulting in an increase in the number of big nodules [21]. In biological fixation, Mg influences nodule formation [22,23], with an important role in the process of photosynthesis being involved in enzymatic reactions, enabling them to perform properly, including urease and nitrogenase, which are involved in N fixation [24,25]. The addition of Mg together with other nutrient elements contributed to increase nodulation as well as the yield and quality of soybean seeds inoculated with *Bardyrhizobium* sp. [26,27]. Hence, the high Mg concentration in the iron-slag coating material used in this study increased soybean nodule activity in the soil, originally with low N content, and then increased N fixation and N content in the soybean grain. The iron coating method was normally used to prevent bird eating damage, but our finding found that the material is a potential source of nutrients and increases nutrient quality such as N and Mg during soybean cultivation.

5. Conclusions

Seed coating methods are important tools used to increase soybean yield and prevent crop losses due to bird-inflicted damage, fungal diseases, heavy worm infestations, and others. Iron-slag coating slightly increased soybean yield but did not significantly differ from the non-coated seed treatment. However, nutrition concentrations significantly increased in the coated seeds, by 20%, 15%, and 6% for Mg, Mn, and N content, respectively, in comparison with the non-coated seeds. In several countries, nutritional supplements are applied individually to crops to enhance microelement nutrition. Based on our findings, iron-slag coating can be one of the solutions implemented to improve the nutrient quality of soybean, specifically Mg, aside from ensuring no yield loss.

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