学 位 論 文 の 要 旨

論文題目　Physiological and biochemical studies on growth and low phosphorus tolerance in low-phytate soybean lines
（ダイズ低フィチン系統の生育と低リン耐性に関する生理生化学的研究）

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Chapter 1. General introduction

Phytic acid (*myo*-inositol-1,2,3,4,5,6-hexa*kis*phosphate; InsP6, phytate-P), a storage form of phosphorus (P) in plant seeds, occurs mainly in the phytate anion form, and accounts for approximately 70–90% of the total seed P content. However, phytate is poorly utilized by monogastric animals, such as chickens, swine, and humans, which have low phytase activity in their digestive system. Improving P and mineral bioavailability is important for stock farming and human food security. As a result, low-phytate (LP) crops and legumes have been developed.

P is a vital macronutrient for plant growth, and soil P levels are generally high. However, despite this abundance, plant growth is limited by P uptake because the forms of P in the soil environment are relatively less available and immobile owing to the transformation of P into organic forms, such as phytates. Under P deficiency, plants can improve their root system by producing longer primary roots and more extensive lateral roots to improve their P acquisition ability. Investigations of the utilization of these potentially valuable ‘low phytate’ traits and of P utilization under P deficiency are important for the development of stock farming practices, to achieve food security, and to ensure a sustainable global P supply. This study investigated (1) the physiological and biochemical characteristics and bioavailability of soybean LP and NP lines during germination; (2) root and root nodule N fixation, phytase and phosphatase activities, and the accumulation of biomass and minerals in LP and NP soybean seedlings; and (3) the physiological and biochemical characteristics, and secretion of organic acids (OAs) and acid phosphatase (APase) in LP soybean lines in response to P deficiency.

Chapter 2. Changes in physiological characteristics, phytase activity, and mineral bioavailability of a low-phytate soybean line during germination

To determine if the germination of the LP and NP soybean lines was affected by a low phytate content, plants were grown in vermiculite the growth, phytase activity, mineral concentration and bioavailability, and mineral extractability of the seedlings were evaluated. Seedling growth did not differ significantly between the NP and LP lines before exhausting all nutrients in the cotyledon. Phytase activities at 11 to 17 days after sowing were 1.3- to 2.6-folds higher in the NP line than in the LP line. The phytate concentration decreased by 18.6% in the NP line and by 32.0% in the LP line 7 days after sowing. In addition, the phytate was hydrolyzed by the LP line 2 days earlier than by the NP line. The LP line had higher molar ratios of phytic acid to Ca, Mg, and K than the NP line. These results suggested that germination and early seedling growth are not affected by low phytate levels in seeds; however, decreased phytase activity might be related to the lower phytate content in LP seeds.

Furthermore, these results indicated that the LP line benefits from a higher bioavailability of P and macro-minerals, unlike the NP line.

Chapter 3. Changes in seedling growth, physiological characteristics, nitrogen fixation, and root and nodule phytase and phosphatase activity of a low-phytate soybean line

To clarify the 'low phytate' effects on the physiological and biochemical characteristics of the LP and NP soybean lines at the seedling stage, this study evaluated the seed P and mineral contents, seedling dry weight, carbon (C) and nitrogen (N) accumulation, nitrogen fixation, and root and nodule phytase and phosphatase activity levels at 21 days after sowing. Seedling dry weight and C and N accumulations were 31%, 38%, and 54% higher, respectively, in the LP line than in the NP line. The N fixation levels were 46% higher in the LP nodules than in the NP nodules. The phytase and phosphatase levels were 1.4-folds and 1.3-folds higher, respectively, in the LP roots than in the NP roots. The phosphatase levels in the LP nodules were 1.5-folds higher than those in the NP nodules. The mineral levels were substantially higher in the LP seeds and seedings than in those of the NP line. The HCl extractabilities of P, S, Fe, Cu, and Mn were higher in the LP seeds than in the NP seeds. These results indicated that the LP line showed superior seedling growth and N fixation relative to the NP line. The LP line showed relatively higher root phytase and root and nodule phosphatase activity levels than the NP line. These results indicate that the LP line is be better suited and more adaptable to low P conditions.

Chapter 4. Changes in organic acids and phosphatase secreted from the roots of low-phytate soybean lines under low-phosphorus conditions

To exploit and utilize these potentially valuable 'low phytate' traits, it is necessary to understand the physiological and biochemical characteristics of different LP soybean lines (i.e., LP-1 and LP-2 in the present study) as adaptations to low P stress compared with the NP cultivar "Natto Kotsubu". The LP lines showed relatively higher root dry weights and P contents than the NP cultivar. The total root exudation of LP-2 was higher than that of the LP-1 line and the NP cultivar under low-P conditions. The LP lines secreted more APase and produced more extensive lateral roots than the NP cultivar. The APase levels were higher in the roots of both LP lines under low-P conditions than in the roots of the NP cultivar. The leaf photosynthesis rate and stomatal conductance were also higher in the LP lines than those of the NP cultivar under low-P conditions. In summary, the LP lines exhibited more extensive root development, and higher total P and internal APase levels than the NP cultivar. The LP lines also showed relatively higher OA levels and APase exudation from the roots than the NP cultivar.

Chapter 5. General discussion and conclusions

These findings revealed that the LP and NP lines showed similar plant growth traits while reliant on the nutrients supplied by the cotyledon at the germinative stage. However, after the germinative stage, the LP line showed higher root and nodule mineral bioavailability, higher P-related enzyme activity levels, better physiological performance, and secreted more OAs and APase than the NP line. Moreover, LP cultivars may have a higher P-acquisition ability and thus have a higher adaptive potential to low P conditions than NP lines and the "Natto Kotsubu" cultivar, which is the breeding base cultivar of LP progeny.