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Phosphorus Metabolism during Ripening of *Glycine max.* (L.) Merril.

S. K. Arora and B. M. Gandhi

Punjab Agricultural University, Hissar, India

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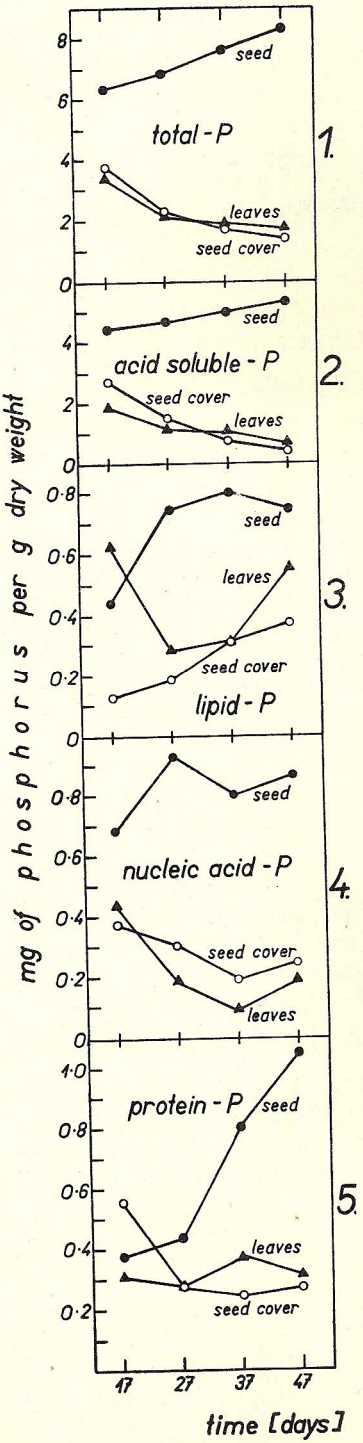
Abstract. The seeds, seed covers and leaves, taken after 17, 27, 37, and 47 days after tagging of flowers of soybean, were analysed quantitatively for their contents of different phosphorus fractions. Total phosphorus content increased in seed cover and leaves, there was a gradual decrease during ripening. All the phosphorus fractions i.e. acid soluble — P, lipid — P, nucleic acid — P, and protein — P were found to increase with maturity in seeds whereas in case of seed covers the content of acid soluble — P, nucleic acid — P and protein — P decreased but a marked increase was observed in lipid — P. In leaves during ripening, all the phosphorus fractions decreased except protein — P which was found to be almost constant. In lipid — P an increase was observed during later stages of maturity.

Phosphorylated compounds play an extremely important role in variety of reactions in seeds. The seeds receive a large share of phosphorus accumulation in plants, which is mostly in the form of phytin or inositol hexaphosphate and also as phospholipids, nucleoproteins, nucleic acid, phosphorylated sugar and other nucleotides. In light of the importance of various phosphorylated compounds in metabolism, different workers have studied the changes in phosphorus compounds during the ripening of Bengal gram (Verma and Lal 1966), wheat (Ergle and Morton 1963), oat (Albaum and Umbreit 1943), cotton (Ergle and Eaton 1957), cherry seed (Olney and Pollock 1960) and lupines (Večer and Matoško 1965). But no work has been reported on the ripening of soybean. In continuation with our previous work on phosphorus metabolism of soybean during germination, it seemed desirable to study the changes in various phosphorylated compounds during ripening of soybean and results have been presented in this paper.

Material and Methods

Cultivation and Sampling Technique

For ripening study, the sowing of seeds of soybean variety Pb. No. 1 was done in the fields of Punjab Agricultural University, Hissar. The open



flowers were tagged and samples for the present study were drawn periodically only from pods set on tagged flowers. Tagging of flowers was done during the peak flowering period. Samples of pods and leaves were collected after 17, 27, 37 and 47 days of tagging. The pods were separated into seed covers and seeds. The samples of seeds, seed covers and leaves were dried at low temperature (70°) and ground in a grinder and stored in glass stoppered tubes for further analysis.

Estimation of Phosphorus Fractions

The following estimations were carried out in duplicate and recovery was tested in each case and results were calculated on that basis.

Total Phosphorus

Total phosphorus in the plant material was estimated according to the method of FISKE and SUBBA ROW (1925) after a prior wet digestion.

Extraction procedures for phosphorus fractions:

The various plant parts were extracted according to the modified procedure of SCHNEIDER (1945).

Acid Soluble Phosphorus

20 mg of the plant material was thoroughly homogenized with 0·2 N perchloric acid with the help of pestle and mortar. The extract was taken in a centrifuge tube and kept in ice water for 15 minutes. Then it was centrifuged. The clear supernatant was removed and residue was again extracted twice for 15 minutes in ice cold 0·2 N perchloric acid and clear supernatant removed. The clear acid soluble extracts were combined and analysed for total — P, after drying and digestion with triple acid.

Lipid Phosphorus

The acid insoluble residue was extracted three times at room temperature with ethanol : ether : chloroform (2 : 2 : 1, v/v) for 10 minutes each. The clear extracts after centrifugation were combined and analysed for total P, after drying and digestion with triple acid.

Nucleic Acid Phosphorus

The defatted residue left was washed with ice cold 5 per cent trichloroacetic acid for 5 minutes and then extracted with an additional portion of 5 per cent trichloroacetic acid at 90° for 15 minutes, and centrifuged. The clear extracts was removed and residue was again extracted with 5 per cent trichloroacetic acid. The extracts were combined and analysed for total — P after drying and digestion with triple acid.

Protein Phosphorus

The hot trichloroacetic acid insoluble fraction was suspended in 1 N sodium hydroxide for 10 minutes. It was then placed in a boiling water bath for another 10 minutes. After cooling it was centrifuged and the clear extract was then analysed for total — P after drying and digestion with triple acid. The final residue left was analysed for total — P after digestion with triple acid.

Results and Discussion

The samples of seed, seed cover and leaves taken after 17, 27, 37 and 47 days after flowering of soybean plants were analysed quantitatively for their contents of different phosphorus fractions. The results obtained are tabulated in Table 1 and shown graphically in Fig. 1 to 5. In seeds, during ripening, total phosphorus increased whereas in seed cover and leaves the decrease was observed. A perusal of various fractions of phosphorus (Table 1) shows that acid soluble — P, lipid — P, nucleic acid — P and protein — P also increases in seed during ripening with concomitant decrease in seed covers and leaves. In seeds, lipid — P decreased during later stages whereas in nucleic acid a decrease (37 days) was followed by a slight increase. The

TABLE 1

CHANGES IN PHOSPHORUS FRACTIONS during ripening of soybean mg/gm (on dry weight basis)

Plant Part	Days after flowering	Total-P	Acid soluble-P	Lipid-P	Nucleic acid-P	Protein-P	Residual-P
Seed	17	6·38	4·56	0·44	0·69	0·38	0·19
	27	7·00	4·75	0·75	0·94	0·44	0·13
	37	7·75	5·06	0·81	0·81	0·81	0·19
	47	8·38	5·38	0·75	0·88	1·06	0·13
Seed cover	17	3·81	2·75	0·13	0·38	0·56	0·09
	27	2·31	1·50	0·19	0·31	0·28	—
	37	1·69	0·81	0·31	0·19	0·25	0·06
	47	1·41	0·44	0·38	0·25	0·28	0·06
Leaves	17	3·38	1·88	0·63	0·44	0·31	0·06
	27	2·09	1·13	0·28	0·19	0·28	0·09
	37	1·81	1·00	0·31	0·09	0·38	—
	47	1·69	0·63	0·56	0·19	0·32	—

increase in acid soluble — P shows that during ripening, phytin is being accumulated. ROWEN and TURNER (1957) also reported an increase in acid soluble — P, and JENNINGS and MORTON (1963) also reported the formation of phosphoprotein during wheat grain ripening.

In seed covers, the content of acid soluble — P, nucleic acid — P and protein — P, decreased and lipid — P increased. The contents of nucleic acid — P and protein — P, increased during later stages. It is interesting to point out that the lipid — P, goes on increasing in seeds as well as in seed covers. The results are at variance with those of VERMA and LAL (1966). It might be due to the fact that soybean is also a rich source of oil.

In view of the fact that the seed cover attains its full size at a very early stage of seed development and since the levels of total and other phosphorus fractions in the seed cover, even at early stages of seed development, are much lower than in the young seed of the same age, and at subsequent changes in these levels of phosphorus fractions in the seed cover are quite insignificant as compared to those in the developing seed. It is suggested that at least a major part of the nutrients from the plant parts are translocated directly into the developing seeds, without being distributed or stored in pod covers. Similar results were observed by VERMA and LAL (1966) during their study on development of Bengal gram seed.

A decrease in the amount of phosphorus in seed covers, during ripening points to another interesting implication, that in the initial stages of seed development, the seed cover takes active part in the seed metabolism, but this activity diminishes with the progress of maturity and the seed cover gradually assumes the role of a protective layer for the embryo and cotyledon.

The content of various phosphorus fractions, showed many variations in leaves during seed maturation, acid soluble — P, lipid — P, nucleic acid —P and protein — P, decreased in the later stages of maturity.

The change in the different phosphorus fractions in relation to total phosphorus present in different parts shows that the acid soluble — P as a percentage of total phosphorus declines slightly over the period of sampling in seeds, whereas in seed covers and leaves, it decreases sharply. The lipid — P and protein — P as a per cent of total — P increases in all parts during ripening. Nucleic acid — P, increases in the seed in the beginning and attains a constant value at the later stages of maturity. In seed covers, nucleic acid — P, as per cent of total — P, increases while in leaves it decreases in initial stages and increases later on.

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S. K. ARORA, B. M. GANDHI, Pandžabská zemědělská universita, Hissar, Indie: **Metabolismus fosforu při zrání soje *Glycine max.* (L.) MERRIL.** — Biol. Plant. **12** : 139—143, 1970.

Byly prováděny analýsy na obsah fosforu v několika základních frakcích v semenech, osemení a v listech odebraných 17, 27, 37 a 47 dnů po odkvětu. Celkový obsah fosforu v semenech se během zrání zvyšoval, zatím co v osemení a listech se postupně snižoval. Ve všech frakcích, tj. v kyselinorozpustné frakci, lipidické frakci, frakci fosforu nukleových kyselin a ve frakci fosforu proteinů se v semenech během zrání obsah fosforu zvyšoval. V osemení se obsah fosforu snižoval ve frakcích fosforu rozpustného v kyselinách, fosforu nukleových kyselin a proteinů, zatím co výrazné zvyšování obsahu fosforu bylo pozorováno ve frakci lipidní. V listech se obsah fosforu postupně snižoval ve všech frakcích vyjma frakce fosforu proteinů, kde obsah fosforu zůstával na téměř stejné úrovni. Ve frakci fosforu lipidů bylo pozorováno zvýšení obsahu fosforu v pozdějších fázích zrání.