CHAPTER THREE

Moisture Content:

The moisture level in the seeds and pod covers of the C.cajan and P.tetragonolobus were studied at different developmental stages after anthesis.

Successful fruiting depends on a number of external and internal factors. Assuming that functional pollen and ovules are produced, successful fertilization and fruit development is under the influence of the following environmental factors.

(1) Temperature
(2) Height
(3) Nutrient level
(4) Moisture content/level

Out of the above factors moisture level plays important role in pod development. Moisture level in different types of cells depends upon their age, physical environment and stages of growth.

The influence of low atmospheric moisture levels or rather relative humidity, on pod set is connected with high temperature. Excessively high relative humidities at or close to saturation levels are adverse, frequently resulting in abscission of pods in addition to favouring invasion by pathogenic organisms. In the case of the

groundnut (and perhaps also the other geocarpic pulses such as Voandzeia and Kerstingiella), moisture levels may influence normal development of the pod.

In groundnut the carpophores produced after fertilization of the ovules normally penetrate the soil vertically for several centimetres and then the fruit develops in a more or less horizontal zone. If the carpophore does not reach the soil or fails to penetrate it, it usually withers in some spp. of groundnut, especially Valenica, one capable of developing reduced pods only when relatively humidities are fairly high.

Once pod and seed developments are well advanced, an adequate moisture level is required to mature seed. If the moisture supply is curtailed, particularly during the latter half of growth, the seed production will not be able to mature normally and will be more or less shrivelled at harvest. In addition to failure of natural rainfall, excessive competition for water by needs or unduly high drop, plant population can cause problems in this respect. There is no doubt that available water supply during the latter part of the crop season may be a decisive factor determining yield in both the quantitative and qualitative senses.

The pod of P.arvensu increases first in length and width and then in wall thickness, attaining its maximum fresh weight before the contained seeds become really active in laying down storage reserves (Flinn and Rate, 1968). After this, pods lose dry matter

and nitrogen steadily, final drying out being accompanied by a quite rapid loss of chlorophyll and photosynthetic capacity (Flinn and Rate, 1970).

P. Valgaris, L.CV. Senole pods removed from the plant, continued their development when incubated in suitable conditions of moisture and temperature and seeds continued to grow and develop pods and seeds and passed through an apparently normal developmental sequence to dryness. Seed growth was at the expense of pod dry wet reserves. Losses of dry out were parallel to dry out gain by seeds in detached pods and in pod cylinders containing seeds. The transfer activity was apparent only with the period 10-30 days after anthesis (DAA) with maximal activity between 15 to 20 DAA. This period corresponds to maximum pod growth and the attainment of maximal dry weight. Seeds are in only the early phase of seed growth at this time. No dry weight transfer was observed at developmental stages beyond 10-30 days after anthesis when normal senescence dry weight losses in pods became evident and seeds were in the later phase of seed fill, pod and pod cylinders remained green succulent over the transfer period, later passing through yellowing and drying phases characteristics of normal development. Dry weight transfer was dependent on funicle integrity and was readily detectable in pod cylinders after 7 days incubation. The dry weight transfer activity may contribute to continuing nutrition of seeds under conditions where the normal assimilate supply to seeds becomes limiting plants reduced seed yields but allowed persistance of seed maturation processes

such that all seeds developing to dryness were capable of germination.

Singh et al. (1987) reported that, as the seed of C. cajan reaches moisture below 46 per cent during drying the quality was reduced in terms of their germibility. The seeds got aged on the mother plant itself after physiological maturity.

From Table-2.1 it was evident that the fresh weight seeds and pod cover in C. cajan from 10 days to about 35 days after fertilization. Thereafter fresh weight declined rapidly as the seeds desicated. Dry weight of seeds and pod cover increased steadily reaching a constant value at about 35 days after fertilization. Between 10 and 25 days after fertilization, pod weight increased rapidly suggesting that much of the substrate available for fruit formation was channelled into the pod that time.

When pods started drying and seeds reached their complete maturity, there was a significant reduction in the moisture content. In P. tetragonolobus a measurable amount of water was lost from senesing seeds and pod covers. The pod fresh weight started its rapid increase about 10 days after anthesis and maximum was attained at about 45 days in P. tetragonolobus and at about 35 days in C. cajan.

Maximum fresh weight was attained at 25 days in C. cajan and 45 days in P. tetragonolobus and declined thereafter as the seeds matured and dried. Luthra et-al. (1983) noted that, the increase in seed dry matter observed between 30 and 35 days in C. cajan

and 40 and 45 days in P. tetragonolobus after anthesis. It was probably due to loss of dry matter in the pod wall. The moisture percentage in seed was increased till 30 days and then decreased. The pod wall gained more dry matter as compared to seed during initial stages of pod development. Flinn and Pate (1970) and Khanna Chopra and Sinha (1976) also concluded that the increase in dry matter of seeds of pea and mustard at later stages of pod development was due to loss of dry matter in the fruit wall.

Murry (1990) has observed that maturation of Soybean CV. Anoka fruit from final dry weight accumulation, when pods were still green to the terminal stage of desication when pod walls were brown and seeds were yellow. Fruit development was analysed in terms of water content, conductance as measured by dye flow and water potential of fruit parts. When pod walls and seed coats lost all greenness, dye flow into fruit stopped first at the seed coats, followed by the funiculus, pod wall and pod wall vascular bundles. As the pod wall cavity dried in response to pod wall dehydration, major seed dessication started and was also reflected by decreasing water potentials. The finding that during dessication the water potentials of pod walls were more negative than that of the water potential of seeds suggested that seed moisture evaporated into the pod wall cavity, was absorbed by the pod wall membrane, and then was evaporated from the pod wall into the atmosphere. This conclusion

TABLE-21: Growth, development and moisture content of C.cajan and P.tetragonolobus pods at different developmental stages.

Day after anthesis	Length of Pods cm		Breadth of Pods cm		Fresh wt. 100^{-1} pods C.cajan		Dry wt. 100^{-1} pods C.cajan		Moisture % in C.cajan		Fresh wt. per P.tetra.		Dry wt per pod P.tetra.		Moisture % in P.tetra.	
	C.cajan	P.tetra	C.cajan	P.tetra	Seeds	Pod covers	Seeds	Pod covers	Seed	Pod covers	Seed	Pod covers	Seed	Pod covers	Seed	Pod covers
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
5	-	03.38	-	0.28	-	-	-	-	-	-	-	0.58	-	0.03	-	79.75
10	5.1	11.16	0.40	1.19	5.50	25.55	01.06	5.25	77.39	79.45	0.81	2.43	0.15	0.26	44.00	89.25
15	6.0	17.45	0.45	2.18	5.70	30.00	01.05	5.42	88.14	81.93	2.72	9.86	0.36	0.77	65.50	92.19
20	6.9	21.74	0.50	2.58	5.32	39.50	01.05	6.40	83.83	78.81	8.10	16.63	1.35	1.36	50.00	91.87
25	7.6	22.35	0.56	3.30	14.70	30.20	03.16	7.17	80.26	71.47	20.17	30.16	4.09	3.05	32.00	89.89
30	8.2	22.75	0.60	3.45	29.20	23.10	15.52	8.38	78.50	68.96	30.15	32.65	7.35	4.22	31.00	87.09
35	8.9	22.75	0.80	3.45	30.10	22.10	18.20	8.50	65.36	62.09	42.70	34.72	11.64	5.25	26.50	84.89
40	9.0	22.75	1.20	3.50	24.02	09.00	20.32	8.94	15.40	00.66	54.10	41.34	18.14	6.61	19.80	84.01
45	-	22.60	-	3.50	-	-	-	-	-	-	92.41	45.95	41.23	9.58	12.40	75.86
50	-	22.50	-	3.50	-	-	-	-	-	-	83.57	30.69	41.15	9.47	10.30	69.14
55	-	22.50	-	3.50	-	-	-	-	-	-	62.32	18.54	41.17	9.65	5.10	47.95
60	-	22.30	-	3.45	-	-	-	-	-	-	43.28	11.28	41.17	8.71	5.00	22.78

was supported by results showing delay of pod wall and seed dessication by high humidity. High humidity delayed pod wall yellowing and prevented browning.

Dutt and Thakurta (1939) studied different stages of cajanus seed and concluded that, pre-resting seeds (i) were fresh before being dried, resting (ii) those that had been dried to less than 12 per cent moisture. Post-resting (iii) seed had been dropped water rapidly with decreasing moisture.

Table-2.2shows dry matter percentage worked out by different cultivars by Kadam et al. (1983). Comparing values given in Tables 2.1 and 2.2 moisture percentage in seed C. cajan is higher than values given in Table 2.2. Pod covers of C. cajan and P. tetragonolobus show higher moisture percentage than the values given in Table No. 2.2.

TABLE-2.2: Accumulation of dry matter in P.tetragonolobus seed

Stage of maturity Days after Flowering	DRY MATER (%) CULTIVAR		
	Srilanka	Nigeria	Indonesia
40	16.46	17.58	17.08
50	20.64	31.44	30.36
60	32.64	44.39	40.79
70	45.31	49.1	43.92
80	86.98	86.46	84.98

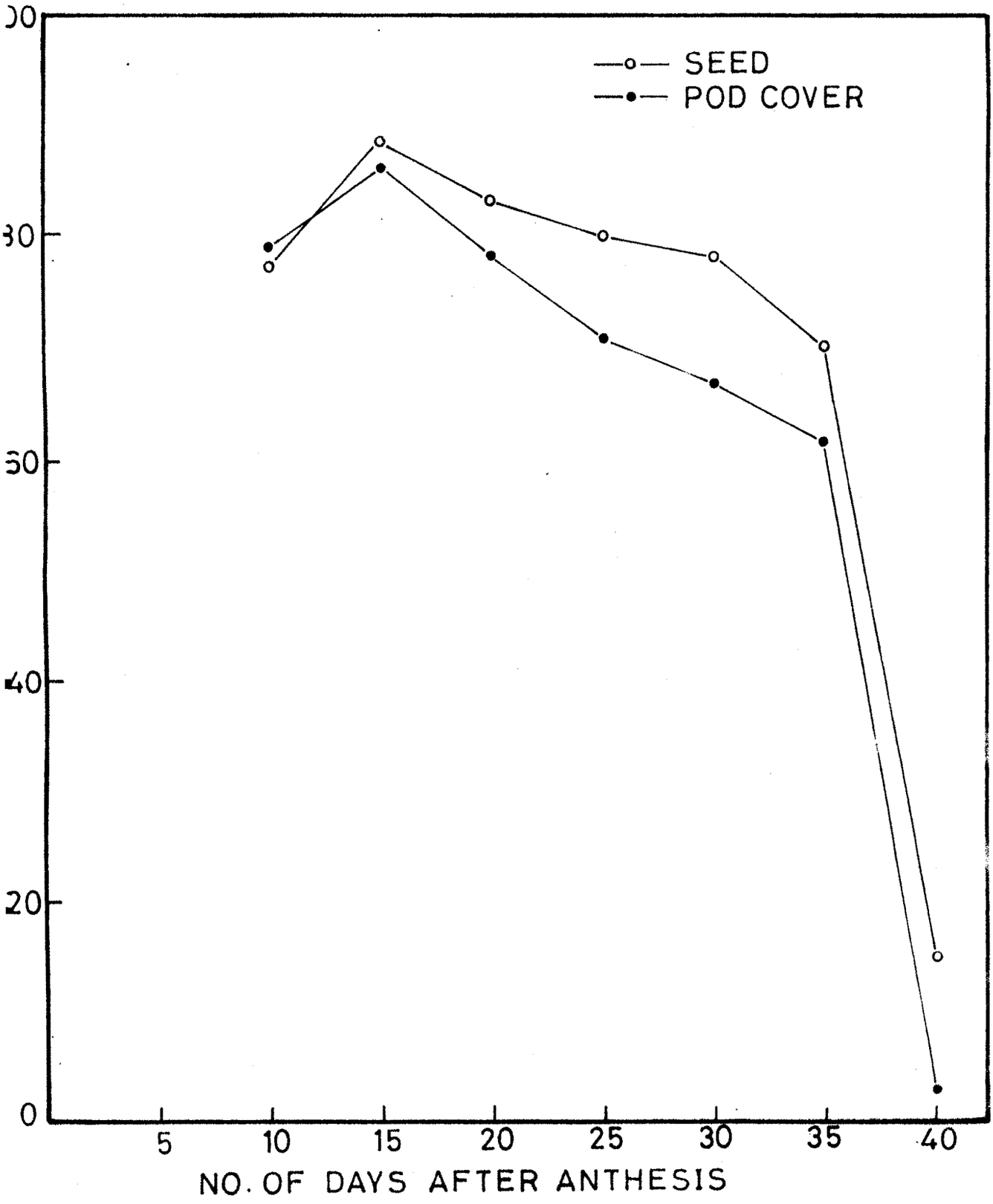

FIG. 2.1 MOISTURE PERCENTAGE IN SEED AND POD COVE OF C. cajan DURING POD DEVELOPMENT.

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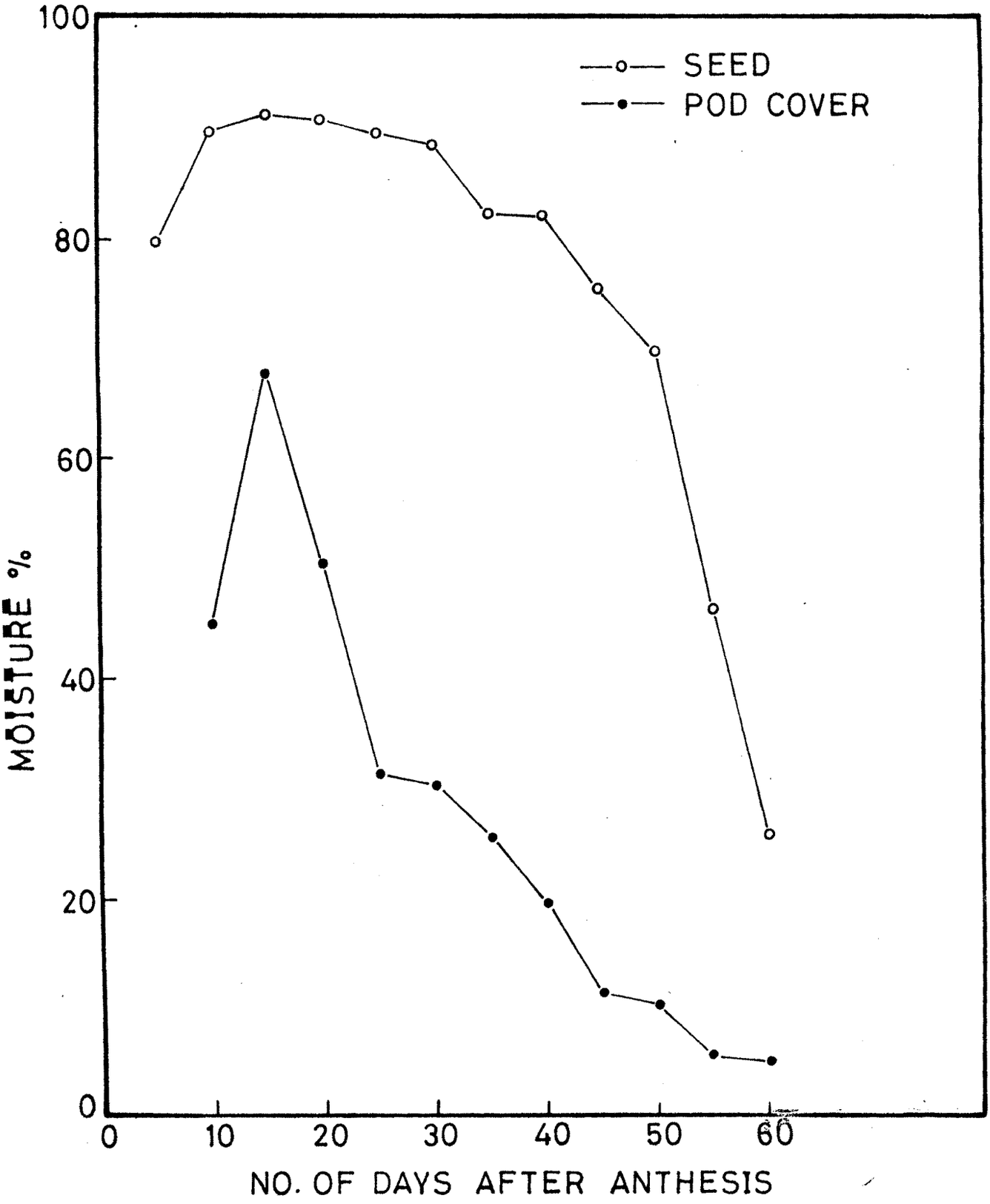

FIG. 2.2 MOISTURE PERCENTAGE IN SEED AND POD COVER OF P. tetragonolobus DURING POD DEVELOPMENT.

NITROGEN METABOLISM

(i) Total Nitrogen:

Legumes are considered as self-supporting in respect of their nitrogen requirement due to symbiotic nitrogen fixation. Sufficient N is necessary for proper root growth and nodule formation which play an important role in symbiotic nitrogen fixation. Pulses can fix nitrogen as much as 45 to 217 Kg/ha (Annoymus, et al., 1971).

The roots of higher plants absorb N from the soil, in the form of nitrates (NO_3). Nitrate nitrogen is not directly used by plants, it is reduced to ammonia with the help of enzyme nitrate reductase and nitrite reductase.

Nitrogen is known to play a vital role in three major aspects of yield: (i) Formation of vegetative structures for nutrient absorption and photosynthesis. (ii) Formation of reproductive structures and determination of sink strength. (iii) The production of assimilates to fill the economically important sink. Nitrogen is primarily derived through symbiotic nitrogen fixation. The limitation of nitrogen in pods may be due to low nodule activity and leaf nitrogen. Nitrogen accumulation in the leaves and stem ceases after the onset of pod development approximately 70 days after planting (Tonn and Weaver,

et al., 1981).

As the supply of nitrogen increases the production of protein in the plant also increases which helps the pods to grow in size. So that large surface of the pod cover is available for photosynthesis. Thus, the proper range of nitrogen supply for many crops and the amount of pod cover area available for photosynthesis is roughly proportional to the amount of nitrogen supplied.

Nitrogen is essential for plant growth. When plant is grown in nitrogen deficient soil, it shows deficiency symptoms like, stunned growth, chlorotic foliage and reddish colouration of stems. Reid and Cox et al. (1973); Reid and Yord et.al. (1958) showed that the nitrogen deficiency affects fruit, pod and kernal development in peanut to fulfil nitrogen deficiency of plant 40 million tons of nitrogen fertilizers are produced per year and it is rising by 2 million tons per year.

Forms and Fate of Nitrogen in Plants:

Most plants derive nitrogen required for their metabolism from the soil solution. There are three main sources of inorganic nitrogen, viz., soil organic matter, atmospheric nitrogen and nitrogen fertilizers. During decomposition of organic matter, excess NH_4^+ is released which is not utilised by microbes and subsequently oxidised by autotropic bacteria to NO_2^- and then NO_3^-. Nitrites do not accumulate but accumulate temporarily under special conditions such as, when pH

is more than 7 and NH_4OH together, inhibit the NO_2-oxidizers. When atmospheric nitrogen is fixed, the first form of combined nitrogen to appear is NH_4^+ and most of it is immediately assimilated into organic forms, so that very little is exuded to soil. In spite of fertilizer nitrogen, soil supplies major source of fixed nitrogen to the plant.

Decomposition of Organic Matter:

N_2 **Fixation**

Organic matter —Mineralization→ HN_4^+ —Nitrification→ NO_3^- —Denitrification→ H_2, H_2O

HN_4^+ —Immobilization→ Organic matter

NO_3^- —Nitrite reduction→ HN_4^+

NO_3^- —uptake plants reaching ground water

The amount and type of organic matter and presence of microbial population and conditions favourable for microbial activities, are essential for availability of nitrogen to plants. Under optimal conditions the microbial activity is favoured for plant growth, but the microbes usually bear a wider tolerence range than plants. The direction of nitrogen transformation process is dictated by C:N ratio in the soil. However, nitrogen limitation for example after the addition of excess carbon in the form of organic material with less than 1.5 per cent N, results in the net immobilization of nitrogen until the C:N ratio is lowered to 20-30 when net nitrogen mineralization is reinitiated (Broadbent, 1973).

Under aerobic condition the rate of nitrification is faster

than nitrate reduction and mineralization is faster than immobilization (Broadbent, 1968). As a consequence there is often a continual conversion of organic nitrogen to nitrate, with or without accumulation of ammonia. Excess water in the soil will obstruct the growth of aerobic microbial population, e.g., fungi and actionymycetes.

The important forms of inorganic nitrogen in plants are nitrates, nitrites and ammonia. The percentage of these three may differ considerably but in general very rare nitrte is accumulated, while concentration of ammonia is relatively low between 0.004 M and 0.01 M (Hewitt et al, 1957). Thus, nitrate remains the principal source of nitrogen for most higher plants growing under normal field conditions in fertile soils. When the soil is moist, well aerated and temperature is favourable, then nitrification of ammonia is rapid and plant growth is vigorous (Russell, 1960).

Nitrogen plays major role in the determining processes in all crops. Williams et al. (1979) studied the nitrogen uptake characteristics in a groundnut cultivar Egret found that leaf and stem nitrogen contents decreased from early kernel growth in groundnut and remained fairly constant until maturity. Reddy et al. (1981) observed that in groundnut nodule number and dry weight of plant increased from seedling stage till maturity. But the nitrogen content in the foliage was decreased from flowering, while it increased in the productive parts. Thus, kernel nitrogen is derived from foliage, which was accumulated essentially from the dinitrogen fixation of

the nodule. However, the amount of nitrogen removed by the pod was proportionally lower to the quantity of nitrogen in C. cajan and P. tetragonolobus are dinitrogen fixation and soil nitrogen. Accumulation of nitrogen in vegetative and reproductive parts depends on nitrogen availability for the plant, either by rhizobial nitrogen fixation or soil nitrogen. The nodule mass increases with growth of plant and leaf nitrogen decreases from flowering stage till fruit fruit formation stage. This decrease in nitrogen may be due to either reduced nitrogen fixation or cue to sink strength for nitrogen accumulation or even both.

Total nitrogen uptake and fixation in C. cajan and P. tetragonolobus are generally increased with crop duration but there are substantial differences between cultivars within a maturity group. Early maturing determinate fix little nitrogen (maximum 7 Kg N ha^{-1}. Indeterminate early and medium maturing cultivar fixed more nitrogen (27 to 55 Kg N ha^{-}). Late maturing cultivars estimate the fixed nitrogen range from 13 to 69 Kg N ha^{-1A}. (Kumar, Rao J.V.D.K and P.J. Dart (1987). Ziena et al. (1987) had reported that, the chemical composition during the pod development of faba bean seed changes during the development of which was affected by pods (nodular, middle and terminal position. D. Van, 1990 showed that the uptake and redistribution of nitrogen and photosynthesis in legumes such as peas, V. faba, V. ungulculata, L. albus and cereals during seed filling, the uptake and fixation of nitrogen decreased. Nitrogen was redistributed to seeds to form the vegetatve parts. There is linear

relationship between nitrogen concentration and rate of photosynthesis at low but not at high nitrogen level.

Nitrogen Metabolism During Fruit Ripening:

Nitrogen metabolism during fruit ripening is characterised by a transient increase in respiratory activity. This has been termed as respsiration climacteric and usually preceds the softening and visible ripening of the fruit. The perod appears to be associated with metabolic changes and fruits have been extensively studied with a view to establshing both the cause of the respiratory burst and the nature of the changes in metabolism which occurs during ripening process.

Brevendan et al. (1977) reported that, the increase in nitrogen supply to soybean crop during flowering and pod set, increases the yield due to reduction in flower drop, when nitrogen was supplied at the time of flowering.

Brevendan et al. (1977) observed that, plants which receive higher nitrogen rate during flowering had significantly more pods than those which receive the lowest nitrogen rates. He also reported that nitrogen treatments had no effect on seed size or seed number per pod, but increase in yield was achieved as a result of increase in the number of pods per plant and number of seeds per pod.

The important nitrogen at the time of flowering is also supported by Streeter et al. (1978) by studying the nitrogen starvation

in soybean at various stages and its effect on yield. He reported that the starvation of the nitrogen at the end of flowering or during early pod formation or seed formation, resulted in decrease in individual seed and nitrogen concentration. Longer period of nitrogen stress causes major reduction in pod and seed number.

Nitrogen assimilation in soybean declines during seed development and new assimilation is not sufficient to meet seed needs (Thibodeus P.S., 1975). Jeppson and Johnson et al. (1978) and Leihere and Rotcosky et al. (1982) reported that 50 to 90 per cent of nitrogen found in mature seed, alongwith leaves, stems and pods is important contribution to the pod of reserves available to seeds. They account for upto 30 per cent of the nitrogen that is mobilized to soybean seeds and the pods begin losing nitrogen earlier than stems or leaves (pods are temporary) sink for nitrogen which can be stored as earlier as either soluble compounds or proteins.

The changes in nitrogen concentration with growth upto 140 days after sowing in plant tops, roots and nodules of C.cajan cultivars of different maturity groups evidenced that the young plants had the highest nitrogen concentration around 60 days after sowing. In plant tops, nitrogen concentration declined 40 days after sowing. The root nitrogen content declined rapidly after 20 days but was quite stable after 60 days. Bain et al. (1966) have noted that in peas (P.sativum) there was an initial deposition of nitrogenous materials in the endosperm, following anthesis but their components were

deflected and nitrogenous materials accumulated extensively in the developing embryo.

Kumenko et al. (1962) observed that when seeds and pods of Vinca faba were compared at the milky, waxy stages of ripening with fully ripe pods, there was decrease in total nitrogen similar to that in the leaves. The pod cover accumulated nitrogen compounds upto 21-28 days after anthesis with their subsequent distribution to the developing seeds of Pigeon pea (Kharta, Sidhu et al. 1987). Pinegia, Klimenka et al. (1961) had described that nitrogen content of vegetative mass of crop like pea was as follows. Stroma N 1.25-1.99 per cent, extractable N 0.88-1.99 per cent and protein N 0.65-2.16 per cent. Kalenov et al. (1983), put forth that, the development of early stage of soybean seed, total nitrogen accumulation is identical, however after that time and upto maturation the relative total nitrogen and protein content of early stage of seed development is greater. In final stage of maturation protein nitrogen increases from 32.4 per cent to 88.4 per cent.

Wiewlor Kowski et al. (1960) found that, during ripening of lupin seeds the rate of nitrogen synthesis is higher than carbohydrates or lipids, mainly due to increase in globulines. Nitrogen exchange between developing cowpea fruit and the parent plant showed that 96 per cent nitrogen of the fruit was incorporated into the seeds (Pate, Atkins, 1986).

From the observation Table-23 it is evident that, nitrogen

content in the seeds and pod cover of developing C.cajan and P. tetragonolobus pods was not stable. In the initial stage of development, nitrogen content in seed was less, but later there was incredible increase in it in the seeds of C.cajan and P.tetragonolobus till the end of maturity. However, pod cover showed a gradual decrease in total nitrogen content during later phase of pod development.

Flinn and Pate (1968) studied the biochemical and physiological changes during maturation of field pea (Psum arvense) fruits. They noticed that during the early phases upto 35 days after anthesis the fresh weight, dry weight and total N^2 of pod exhibited certain fluctuations. There was overall increase in all these quantities until final maxima were attained after 35 days. A net loss of dry matter and nitrogen from pod occurs during the next phase, 35 days after anthesis until seed maturity. Rauf et al. (1978) studied the changes in protein nitrogen and soluble nitrogen during pod development in C.arientinum, P.sativum and V.faba. He noticed that in C.arientinum there was no significant change in protein nitrogen upto 35 days after anthesis. Thereafter it declined, whereas in pea and V.faba, the level of protein N per pod increased upto 35 days after anthesis and thereafter it showed a steep decline. The soluble N level of the pod in Cicer attained a maximum level by 25 days after anthesis and then showed a decline. In pisum, the soluble 'N' level increased in the 1st phase and thereafter decreased relatively slow in the second phase and rapidly in the 3rd phase. In V.faba soluble content increased upto 35 days after anthesis and this was followed by depletion.

Thus, legume spp. seems to differ in time course of N accumulation in pods and its retranslocation.

It was suggested by Pate et al. (1977) that legume pods might act in trapping, proceeding and exporting nitrogen from pods to the seed through xylem.

From the present results a gradual decrease in total nitrogen level in the pod cover of both the plants C.cajan and P.tetragonolobus with concomitant and rather a rapid increase in the total nitrogen content of the developing seeds it is suggested that pod cover is involved in the translocation of nitrogenous substances to the developing seeds in the pod. Thus the nitrogenous material received from vegetative part of the plant is transferred (at least partly) to developing seeds via pod cover in C.cajan and P.tetragonolobus.

TABLE-23: Changes in total nitrogen content of the developing pods of C.cajan and P.tetragonolobus.

Days after anthesis	TOTAL NITROGEN 100^{-1} g DRY WEIGHT			
	C.cajan		P.tetragonolobus	
	Seed	Pod cover	Seed	Pod cover
10	3.00	1.40	1.25	7.60
15	3.10	2.25	-	-
20	3.35	2.15	2.00	5.90
25	3.50	2.00	-	-
30	3.65	1.80	4.80	3.90
35	3.90	1.65	-	-
40	4.10	1.50	7.40	4.00
50	-	-	-	-
60	-	-	7.43	0.80
70	-	-	7.60	0.80

TABLE-2.4 Changes in nitrogenous constituents at different stages of seed development of winged bean on dry weight basis.

Cultivar	Stages of maturity days after flowering	Total nitrogen Percentage
Srilanka	40	4.30
	50	4.36
	60	4.45
	70	5.34
	80	5.64
Nigeria	40	4.49
	50	4.60
	60	4.75
	70	4.75
	80	5.36
Indonesia	40	3.86
	50	4.16
	60	4.60
	70	4.95
	80	5.05

Nitrogen content in seeds of C.cajan as compared with different cultivars by Kadam et al. (1982) is low. Seed nitrogen of P.tetragonolobus seeds of different maturity periods is higher than our findings according to Table-2.3. Nitrogen content in pod cover of P.tetragonolobus according to Table-2.3 is higher than the findings according to different cultivars given in Table-2.4.

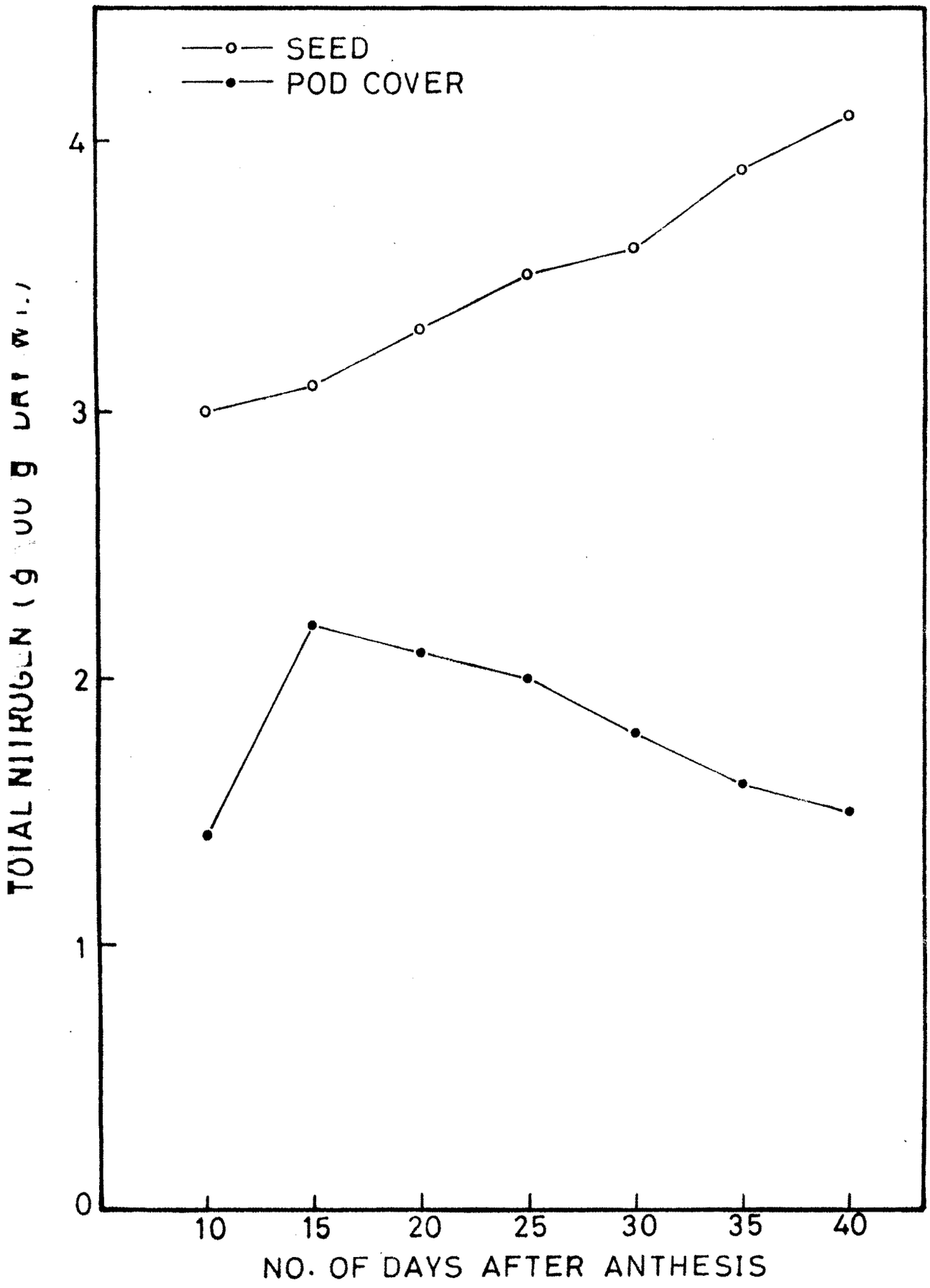

FIG. 2·3 TOTAL NITROGEN IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

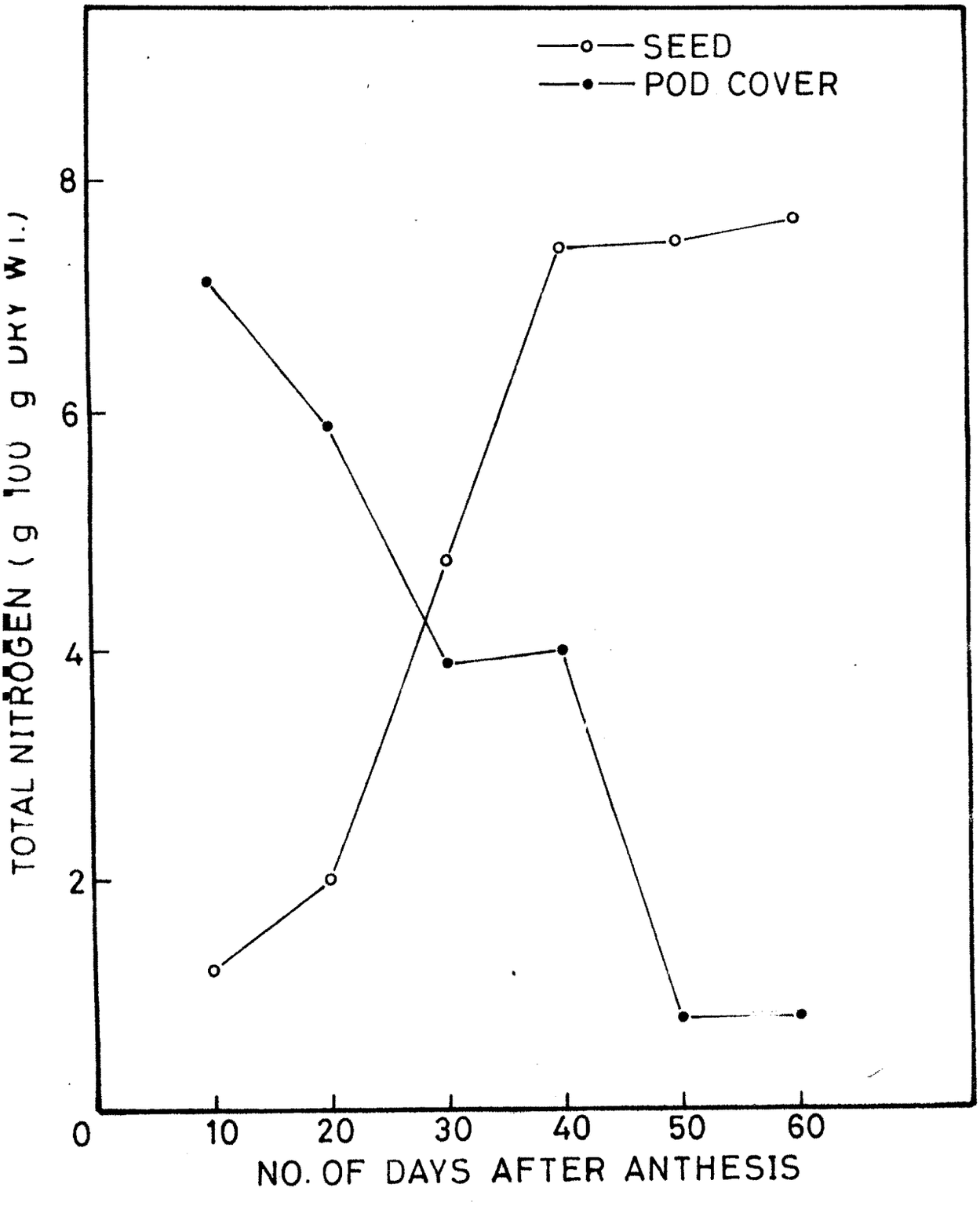

FIG. 2.4 TOTAL NITROGEN IN SEED AND POD COVER OF P. tetragonolobus DURING POD DEVELOPMENT.

NITROGEN METABOLISM

(ii) Enzymes of Nitrogen Metabolism:

Williams et al. (1987) summarised their observations on groundnut and stated that nitrogen accumulation is fast during productive growth, while nitrogen fixation was less during this phase as compared to earlier growth. The nitrogen metabolism in plants is mediated through number of enzyme systems, which catalyse various steps of nitrogen assimilation in different plant tissues. This enzyme system includes the enzymes of nitrate reduction (Nitrate and Nitrite reductases), ammonia assimilation (glutamate dehydrogenase, glutamine synthatase, and glutamate synthase) and of biosynthesis and interconversion of various aminoacids whch are the building blocks of proteins. Gopal and Rao (1982) studied the behaviour of enzyme nitrate reductase during germination of groundnut seedlings maximum enzyme activity occurred in cotyledons after two days of germination and in embryo axis after 6 days of germination. The GDH and GOT activities were considerably high before seed formation and declined as the seed led itself for further stages of pod development.

The rate of pod growth was slow after peg penetration. So, demand for amino acids was low during this period. The level of enzyme GDH and GOT increased from 40 days onwards with faster rate of pod growth. GOT activity reached its maximum at 60-70 days after anthesis followed by a decline at maturity, while GDH activity continued to increase resulting in the highest level at maturity.

High activity of these enzymes at the later stages of development suggested the requirement of amino acids for photosynthesis in the fast developing seeds. The above observations indicate that both GDH and GOT play an important role during pod development in groundnut.

(a) Nitrate Reductase (NR):

Crop plants can take up NO^{-3} or NH_4^+ and assimilate them. Assimilation of N into organic molecules was dependent on the reduction of NO^{-3} by the nitrate reductase enzyme in plant tissue (Neyra and Hageman, 1975).

Nitrate reductase is one of the important enzyme systems in plant nitrogen metabolism. It brings about reduction of nitrate to nitrite. Usually the level of this enzyme is very high in the leaves. Nitrate reductase many times is correlated with photosynthetic activity of leaves and photosynthesis provides the reducing power to this enzyme.

Enzyme nitrate reductase has high molecular weight varying from 2,30,000 to 5,00,000 and the molecular weight depends on the organisms in which it occurs. It contains several prosthetic groups such as FAD, Cytochrome-557 and molybdenum which are the main constituents of this enzyme. It has been observed that, the enzyme activity depends on numerous intrinsic and external factors and this enzyme is substrate inducible. Sinha and Nicholas (1981) indicated that, nitrate reductase catalyzed reactions involving various

electron donars. Further nitrite is reduced by catalytic enzyme nitrite reductase. Burstrom (1943, 1945) worked on young wheat leaves and came to the conclusion that, nitrite reductase is closely linked with photosynthesis and source of energy is light.

Nitrate reductase is found only in mesophyll cells and not in bundle sheath cells of C4 plant leaf. Highest nitrate reductase in root epidermis of maize presents symbiotic pathway for iron transport (Ruffy et al., 1986). The molecular weight of enzyme so far estimated in the eukaryotic plants is about 1000,000 in N.crassa.

(a) Electron Donar Specificity:

Enzyme system includes a reduced pyridine mucleotide (NADPH or NADH) as an electron donar (Evans and Nanson, 1953), a flavin adenine dinudeotide (FAD) as prosthetic group and molybdenum as an activator. Electrons are passed from reduced pyridine nucleotide to FAD giving reduced FAD. Stoy (1956) had shown that, photochemically reduced riboflavin was more effective than even NADH as an electron donar in wheat. The electron is transferred from $FADH_2$ to oxidised Mo, resulting in a reduced molybdenum, which in turn passes electrons to nitrate, reducing it to nitrite. Campbell (1988) considered NR as a multicentral redox enzyme, because it contains several internal electron carriers which are reduced during catalysis. Thus NR can be considered as, "mini electron transport chain". NR have two components:

(1) Smaller - contains FAD and could use NADH to reduce Ferricynide.

(2) Larger - contains Mo and could use reduced methyl vialogen (MYH) to reduce nitrate.

(b) Distribution of the Enzyme in the Plant:

Beevers and Hageman (1969) had observed presence of NR in seed cotyledons, roots, shoots and leaves. It is also present in flower buds (Jones and Sheard, 1972) and in embryos (Rajven, 1958). Reproductive parts of C.cajan possess a capacity of NR.

(c) Role of Molybdenum and Iron:

Evans and Nason (1953) investigated that molybdenum acts as prosthetic group. In soybean nitrate reductase, Mo weakened the bonds between two nitrogen atoms in nitrogen, for reduction. Iron is essential for binding the nitrogen molecule to the enzyme molecule.

Pericarps of legume fruit do not only assimilate carbon autotropically but nitrate also. The presence of NR in pods of legume has been demonstrated in vivo and vitro by Schlesier and Muntz (1974).

It is evident from the values recorded in Table-15 that nitrate reductase activity in the seeds and pod cover of C.cajan was different at different developmental stages. NR activity in the leaves was found to be low between 5 and 10 days after anthesis. It was increased

later till 25 days after anthesis and as the pod senescence started it declined. NR activity in pod cover was less as compared to that in the seeds of C.cajan indicating vigorous nitrate reduction in the seeds of this plant.

Schlesier (1976, 1977c) have stated that, NR activity in field bean pods started at 24 days to 31 days after flowering. It was doubled and about 5 times rise in it was evident in pea. Fruits detached 6 weeks after flowering increased their nitrate reductase activity even 25 times after 60 hr of organ culture in pure mineral nutrient solution containing nitrate. The quantity of nitrate uptake depends on the presence of other compounds, e.g., sucrose. In vicia fruits nitrate uptake and nitrate reductase activity of pericarps decreased with increasing sucrose concentration.

(b) Nitrite Reductase:

The formation of nitrite, requires the transfer of two electrons to nitrate (NO_3^-) and in the formation of hyponitrite, 2 electrons are transferred to nitrite.

Mayer and Schulze (1884) proposed a scheme for nitrite reduction, in which hyponitrite and hydroxylamine functioned as intermediates. Heber and Purczeld (1977) have suggested that, HNO_2 rather than NO_2 which was transported through chloroplast membrane. Thus, HNO_2, in the chloroplast stroma is reduced to NH_3. This reaction takes place outside the thylekoid membrane.

Sinha and Nicholos (1981) indicated that nitrite is reduced by catalytic enzyme nitrite reductase. Hegeman et al. (1962) further demonstrated that, plant extract, when fortified with reduced benzyl viologen as electron donar, were capable of stoichiometrically converting nitrite into ammonia. It was believed that, the reduction of nitrite to ammonia was catalyzed by one protein, namely, nitrite reductase in which siroheme, the iron porphyrin prosthetic group of the enzyme functions in the transfer of six electrons (Murphy et al., 1974). There was no free intermediates.

$$NO^{2-} + be^{-} + 84^{+} \quad NH_4^{+} + 2H_2O \ (AG = 124 \text{ K cal})$$

Extensive studies have been made to determine the physiological electron donar for nitrite reductase. It was investigated that chloroplast could catalyze the reduction of nitrite when strengthened with an additional soluble enzyme component. Subsequently, it was shown that ferredoxin (Losada et al., 1963) could replace reduced violgen dyes as electron donar for the enzymic reduction and thus it appears that illuminated chloroplasts functioned in nitrite reduction by producing a source of reduced ferrodoxin.

Nitrite reductase activity in the seeds and pod cover of C.cajan was more during maturity phase of pod development was declined as pod started senescence. The pattern of NiR activity in both fruit parts was thus identical. Not only this but it is also clear that behaviour of both these enzymes in the seeds and pod cover also followed the same pattern of initial increase during early

phase of development followed by a linear and continuous decline towards senescence or full maturity of pods.

TABLE-2.5: Nitrate reductase and Nitrite reductase activity in developing seed and pod cover of C.cajan.

Days after anthesis	Nitrate reductase (mg $NO_3 g^{-1} hr^{-1}$)		Nitrite reductase (mg $NO_2 g^{-} br^{-1}$)	
	Seed	Pod cover	Seed	Pod cover
10	0.09	0.25	0.07	0.07
15	1.90	1.35	0.16	0.24
20	1.68	1.26	0.38	0.21
25	1.43	1.09	0.21	0.17
30	1.34	1.01	0.18	1.14
35	1.26	0.84	0.15	0.12
40	1.09	0.76	0.07	0.04

It is evidenced that Ammonia is a final product of nitrate reduction which is an intermediate through which inorganic nitrogen from the soil is brought into organic combination. It was thought that reductive amination reaction was catalyzed by enzyme glutamate dehydrogenase Ammonia combines with L-ketoglutarate in the presence of glutamate dehydrogenase in a reductive amination to produce glutamate. First the experiment was carried out with $^{15}NH_3$ with the yeast candida ytukus (Sims et al., 1968).

$$2\text{Ketoglu.} + {}^{+}NH_3 + \begin{matrix}NADH^{+}\\ \text{or}\\ NADPH^{-}\end{matrix} \xrightarrow{\text{Glutamate dehydrogenous}} \text{Glutamate} + \begin{matrix}NAD\\ \text{or}\\ NADP\end{matrix} + H_2O$$

Mostly the enzymes are located in Mitochondria utilize NADHI Given et al. (1970) reported that $NADPH^{+}$ gluctamic dehydrogenase is located in the chloroplast Vicia faba and have been suggested to function extensively in ammonia incorporation in leaves. Since the enzymes glutamate dehydrogenase is located in Mitochondria it plays a catabolic role rather than anabolic one. Tempest et al. (1970) worked with Acrobacter aerogenes (Enderobacter acrogenes) and found the presence of enzyme glutamate synthase in them. This enzyme when preceded by glutamina synthetase would allow an alternative route with the synthesis of glutamate. Pathway is as follows:

$$NH_4^{+} + ATP + \text{Glutamate} \xrightarrow[\text{synthetase}]{\text{glutamine}} \text{Glutamine} + ADP + P.$$

$$\text{Glutamine} + \text{L-oxoglutarate} + NAD(P)\ H^{+} \xrightarrow[\text{synthase}]{\text{glutamate}} 2\ \text{glutamate} + NAD(P)$$

More studies on glutamine synthase and glutamate synthase from bacteria have shown that this might be an effective pathway of ammonia assimilation rather than glutamate dehydrogenase.

Glutamate synthase has been observed in non-photosynthesis plant cells (Dougall & Bloch, 1976) in pea roots (Miflin and Lea, 1975) and in developing cotyledon (Beevers and Storey, 1976), Muflin and Lea (1974) have shown that glutamin synthetase and glutamate

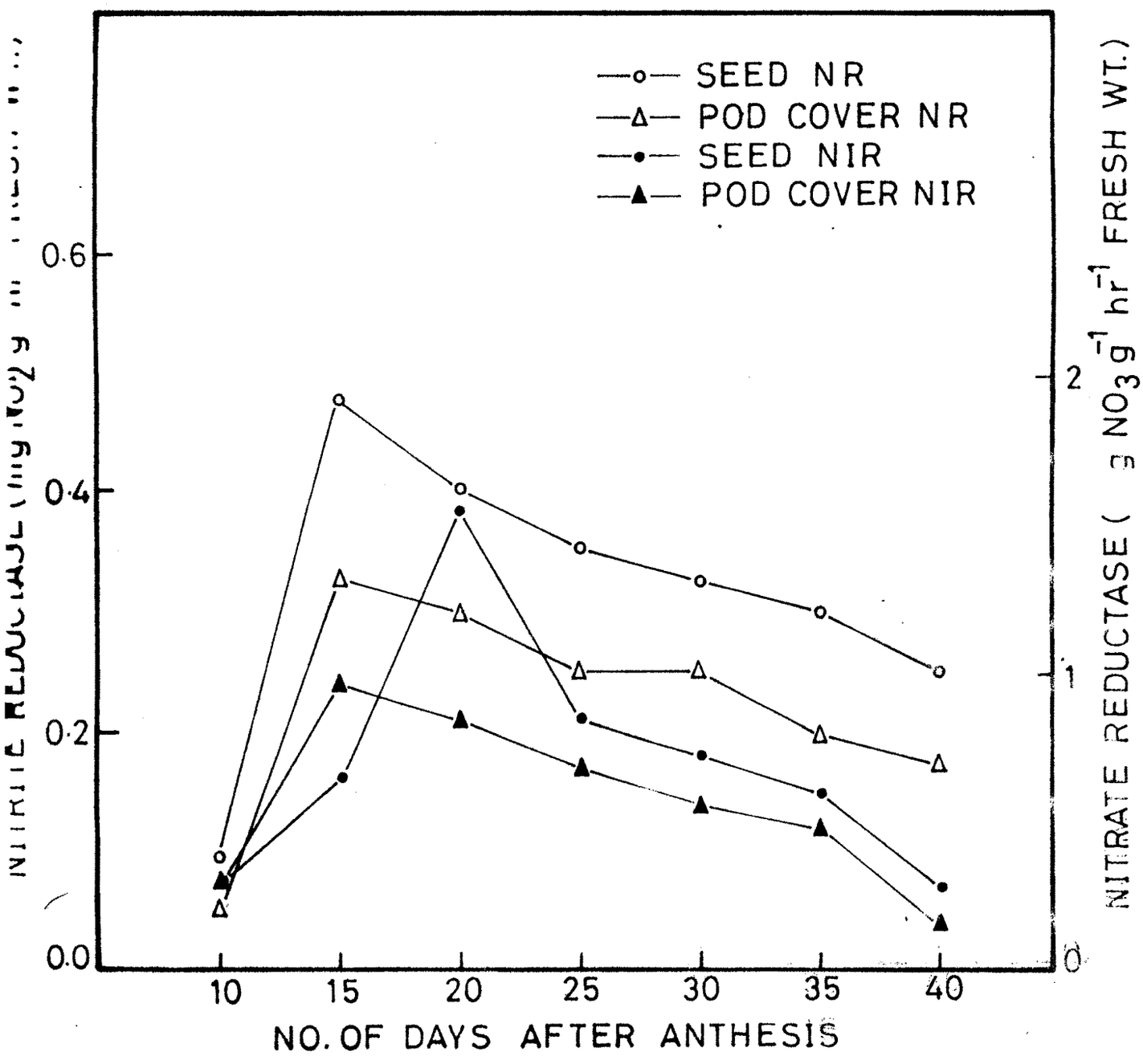

FIG. 2.5 NITRATE AND NITRITE REDUCTASE IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

synthatase is located in chloroplast which show ammonia assimilation.

Once assimilated into amino group, nitrogen may subsequently be distributed among the variety of metabolits in plant cells during transamination reactions. Transamination is catalyzed by an amino-transferase. Aminotransferases are located in the cytoplasm, chloroplasts, mitochondria and microbodies. They play a major role in the biosynthesis of various protein and non-protein amino acids and the process in many instances hinges upon the capacity to form the appropriate precursor. These reactions also include the reactions of photosynthetic carbon metabolism, glycolytic pathway, pentose phosphate pathway and the tricarboxylic acid cycle. Bryan (1976) has listed eightynine different enzymes participated in this process.

PROTEIN

Proteins During Pod Development:

Research on crop improvement in the past few years has been primarily concerned with increasing yield, little emphasis being placed on nutritional equality. In view of various problems of protein calorie, malnutrition, more importance must be given to the production of crop varieties having higher protein content and better quality. Pulses are the major source of protein in the predominantly cereal-based Indian diet. They contain two or three times as much protenous cereals.

C.cajan which comes next to Chickpea in area under cultivation,

is an important food legume not only in India but also in African countries. In pea, there is a close correlation between protein activity and protein breakdown in pods during seed development. Storey and Beevers (1977) have studied the changes in pod proteins during seed development.

If pod proteins contribute to the nutrient needs of seeds then total protein levels show decline during seed development. In levels of soluble protein extracted from pods and seeds beginning, 2 to 3 week after flowering respectively. In maize the protein accumulation in the endosperm appears to be biphasic with an initial accumulation occurring between days 15 and 30 after anthesis and a subsequent second deposition occurring between 30 days and maturity.

In developing peas the proteins accumulate rapidly between days 12 and 25 following anthesis. Beevers and Poulson (1972) studied protein synthesis in cotyledons of P.sativum, length index changes in cell free amino acid and incorporation capacity during seed development and maturation.

In bean (Vicia faba) Millered Millard et al (1971) studied legumin synthesis in developing cotyledons and observed that initial protein accumulated in the cotyledons are the albumins whereas during later stages of seed development the globulins, vicilin and legumin are deposited. The RNA content of the endosperm declined during second phase of protein accumulation and seed maturation.

In tomatoes, apples, peas and contalou peas e.g., there is no increase in protein during ripening while in bananas and avocades there is no change in this component (Sacher, 1973). It has been shown by several authers that during the ripening period there is shift in the pattern of protein within the tissue and even in those fruits showing no net increase in protein content there may be an enhanced incorporation of amino acids (Clements, 1970).

Danielsson (1952) has studied seed development in pea and has shown that the bulk of the reserve protein in developing pea cotyledons consists of two globulins, vicilins and legumin. These two reserve proteins not only have different solubility, sedimentation coefficient constant and aminoacid compositions, but also are laid down at different rates during development. However, their characters as reserve protein are shown by their disappearance during germination. No enzymatic activity is known to be associated with the reserve globulines. The non-particulate enzymes are presumed to constitute the greater part of albumin fraction. The albumin fraction is small fraction of the total nitrogen of the pea and changes slowly during development and germination. The protein bodies disappear more rapidly during germination than the globulin. Thus rupture of the bodies probably proceeds extensive hydrolysis of the globulin reserves. Protein bodies occur widely in cotyledons and endosperm of both starch bearing and oil bearing seeds. Some of the protein bodies contain crystalline inclusions of inorganic salts, that they are probably surrounded by membranes that their formation commences only during

the later stages of ripening of the seed, and that they swell coalesce and disappear early in germination, since these early observations in germination of maize have been studied by many workers. Thus the protein granules of maize endosperm are of the major site of zein storage. Duvick (1961), Altschul et al. (1961), Varner and Schidlovsky (1963a) have shown the major site of the globuline storage. During germination of the peanut the protein bodies are 5-10μ in diameter in the resting seed, swell and develop activities, fragment and disappear (Bagley et al., 1963). These changes occur between 4 and 9 days of germination and coincide with the most rapid disappearance of acid insoluble protein.

It has been known for a long time that pea seed protein consists of two major fractions, a water soluble albumin fraction and a salt soluble globulin fraction (Osborn and Campbell, 1898; Osbor, 1926). Albumins accumulate progressively throughout seed development and comprise many different proteins (Fox et al., 1964) whose relative amount changes during the maturation of the seed (Flinn and Pate, 1968). At maturity albumins are present in about two thirds of the quantity of the globulins (Verner and Schidlovsky, 1963; Beevers and Poulson, 1972).

Developing seeds of pea (Pisum sativum) were examined by Chatterjee et al. (1980) for changes in the fresh weight, dry weight, protein content and the content of amino acids in the ethanol soluble (free pool) and ethanol insoluble (proteins) fraction, at different intervals. At early stages the synthesis of protein was

not as rapid as the translocation of aminoacids from the other plant parts. This was followed by a phase of active synthesis of protein and other storage reserves, as evidenced by the accumulation of dry matter. In the final phase only protein synthesis continued at a slower rates, until maturity. At early stages, all the essential aminoacids were present in desirable amounts or even in slightly higher amounts. The nutritive quality of the unripe seeds were better than that of ripe seeds.

Boulter (1986) studied the synthesis of the storage proteins, legumin, convicilin and vicilin in developing pea seeds. It was found that in the period of about 24 days from flowering to maturity, vicilin accumulates initially faster than legumin, although the synthesis of the latter persists longer to 14 to 20 days respectively. Both vicilin and legumin increase in the cotyledon cells from 8 to 14 days due to a burst of transcriptional activity in nuclei of 8 to 10 days old cotyledons. It is suggested that the protein content of pea seeds could be increased by regulating the transcription to start earlier or continue longer. The synthesis of the 50 kilodalton complex of vicilin subunits dominated the early stages of protein accumulation but was a negligible proportion of the total incorporation in the later stages of seed development by Spencer, 1981. ? or Spencer et al, 1981 ?

Results and Discussion:

From the observation Table-2.6 it was evident that the protein

content of seeds and pod covers in C.cajan and P.tetragonolobus increased during their development. The protein content of C. cajan seed varied from 18.38 to 25.63 per cent dry weight in the case of P.tetragonolobus the protein content of the seeds ranged from 7-42.5 per cent dry weight and that in pod cover varied from 42.5 to 22.4 per cent dry weight. From the observation it was also evidenced that protein content of the seeds increased as the number of days increased from anthesis while in pod cover it was decreased during the later stages of pod development.

The winged bean has been reported to contain 29 to 30 per cent proteins. The proteins of dry hard seeds of winged bean have comparatively low digestibility than other legume seeds. Studies of Ekpenyong and Barcher's (1978) had shown that proteins from mature pods had 73.8 per cent digestibility whereas raw seed proteins had 67.3 per cent. Seed globulines of C.cajan were purified and characterized. About 78 per cent of the seed protein was salt soluble out of which 61 per cent were globulines which were further separated into 3 fractions. The α fraction was insoluble at pH 4.7 and consisting two substantial fractions β and γ were soluble at pH 4.7. The protein consisted of subunits which are not held together by covalent disulphide linkage (Krishna, 1978; Gopala et al., 1978).

Singh et al. (1988) studied physiological maturity of pigeon pea seeds. The effects of harvesting C.cajan at different seed moisture levels (16-66 per cent) were also studied by them. They found

that the total amount of protein did not vary greatly with seed maturation. However, the contents of individual protein did vary. The free amino acid content was maximum at 38.46 per cent moisture. Asana (1968) determined the nutritive value of C.cajan seed and it was found to be quite high with about 20 per cent level, but the seed produced digestive disturbances when fed to mice, over a long period as the major dietory constituents. Samuel (1978) reported protein synthesis and accumulation in bean cotyledons during their growth. It was observed that accumulation of 50 per cent of all protein synthesized took place in this tissue during the following 14 days.

The formation of storage protein during development of P.vulgaris seeds was studied in Lee (1987) using gel electrophoresis. The protein content of the seeds increased gradually during development with rapid deposition of storage protein beginning at 18 days after flowering. At 30 days after flowering the-vicilin like protein became predominant and remained so throughout maturation. Legumin like protein was synthesized at a later stage of development (between 30 and 50 days after flowering). The vicilin-like protein became predominant and remained so throughout maturation.

Ziena et al (1987) investigated chemical changes during the development of Vicia faba as affected by pod and seed position. The chemical changes during the development of faba bean seed position inside the lower pods (nodular, middle and terminal) were studied. Faba beans were analysed at regular intervals during development

for the following constituents: dry matter, total nitrogen, non protein nitrogen, crude ether etc.. The chemical changes for the three zones exhibited the same trend but occurred at different rates; seeds grown on the upper zones had relatively lower cosntent of protein, crude fiber, ash and ether extract than those grown on lower and middle zones. The rate of upper zone is higher than the other two zones. Very slight differences in chemical components were observed among the three seed positions.

Protein level in pea pods reached a maximum at 3 weeks after flowering and then declined by about 37 per cent between weeks 3 and 6. This was the period when a majority of seed proteins accumulated. Raccake (1957b) observed that in ripening pea seed, protein content of the whole seed and the embryo of C.cajan increased continuously throughout the period of study. In seed coat there was a decrease in protein content.

From the present observations it appears that C.cajan and P.tetragonolobus protein contents probably increased at the expense of soluble nitrogen which is in agreement with the observations made by Bission and Jones (1932). Studies of Vigna radiata by Sengupta (1983) stated that main protein constituents of the pulses are the storage proteins, which are deposited at a markedly increased rate upto about 1/3 of the development cycle of the seeds. There was preferential loss of an abundant storage protein from soybean pod during seed development. A temporary vegetative storage protein,

composed of similar 25 kilodalton and 27 kilodalton subunits, was found to be abundant in soybean (Glyain max L.) leaves, stem, pod, flower (Paul and Staswick, 1989). Baikhim (1981) investigated changes in protein complex in ripening seeds of soybean protein complex in ripening seeds of soybean varieties with different vegetative periods.

Development in early 10 days of soybean variety, total protein nitrogen content is greater. Protolytic enzyme inhibitors which affect protein nutrient value, were absent in early seed development but after 23 days seed inhibitor activity increased, reaching a maximum at the end of the vegetative growth.

Storey et al (1978) have detected that changes in the weight and chlorophyll, free aminoacid and protein of developing and senescing, vegetative and reproduction organs of Bisum sativum were measured and the protyolytic activity in the extracts from the senescent leaf and subtended pod was followed in relation to these changes. Protein content decreased in the aging of leaf and pod, as the leaf and pod increased, the leaf protein content decreased. In contrast, protyolytic activity in the subtended pot increased, while the protein level decreased. The proteolytic activity in the extracts from the aging organs was greater than the rates of protein loss. The proteolytic activity of leaf and pod extracts was greater on protein prepared from the respective organ than on non-physiological substrates. Proteolysis was increased by two mercaptoethanol and ethylene diamine tetra-acetate

but was not influenced by addition of ATP to the reaction mixture.

Kalenov et al. (1983) studied development on early 10 days Kumsomalka soybean variety, total nitrogen accumulation is identical. However, after that time and upto maturation the relative total soluble and protein nitrogen content of early 10 days and 20 days of maturation prior and foud that accumulation of above in kumsomalka is greater than that of kumsomalka. In the final stage of maturation protein N increases from 32.4 per cent to 88.4 per cent and 25.7 per cent to 83.1 per cent of the total early nitrogen in early 10 days.

Saxena and Sheldrake (1980c), Singh et al. (1981a) have studied on growth of chickpea pod and stated in both seed and pod wall the percentage of nitrogen and total protein is the highest at first and then declines with the growth of pod. Most of the proteins are synthesized during 25-35 days after anthesis (Srivastava et.al., 1981).

Kumar et al. (1978) analysed chickpea seed during germination and reported changes in the proteins during a 12-day germination, accumulation of subunits of the proteins was negligible during the germination of seed. According to Sun et al. (1978) in bean cotyledons 50 per cent protein is accumulated during the following 14 days of germination.

Hill et al. (1974) reported that the accumulation of major

protein component during seed development and maturation, was more within short period of time during seed development in many legumes. Millard et al. (1971) have described that accumulated proteins are few in kind but they constitute a high proportion of the total protein in mature seed. Thus, the developing seed has been generally recognised to have potential for studies on quantitative and qualitative in regulation of protein synthesis. The seed proteins were extracted beginning at 12 days after flowering and supernatant of the 31,000 gm spin was studied. At all stages of maturity except 12 days after flowering at least 90 per cent of total protein extracted in the crude homogenate, was recovered in the supernatent fraction. At 12 days after flowering only 90 per cent of extract protein was recovered. The relative low level of total protein found at the earliest stages is apparently less than 5 per cent of the total protein of the mature seed had been accumulated by 12 DAF. Protein accumulation was very rapid between 12 and 28 DAF and then declined ceasing at the onset of seed dessication. Thereafter actual protein levels decreased slightly. At very immature stages in seed development (12 DAF and 17 DAF) much of the protein was widely distributed over the pod. Shrivastava (1989) observed that in developing chickpea cotyledon, protein accumulation was between 25 and 35 days after anthesis. He also indicated more deposition of high molecular weight proteins in the later stages of maturity.

We had seen that the protein content in seeds continuously increased but in pod cover it was declined. We have already seen

that protein contents decrease during senescence of leaf and as pod cover is a leaf, protein content of pod cover decreased during maturation of pod. Michael (1936), Wallgiehn (1967) have evidenced declination of protein and RNA during maturation and senescence. In nineteenth century it was first suggested that the proteins in plant cells were in a dynamic state, being continually broken down and resynthesized. A continuous breakdown of protein to their constituent aminoacid is evident. Resynthesis of new proteins is required to incorporate loss of protein from senescing tissue concerned. Such imbalance between the rate of protein synthesis and rate of degradation is caused by slowing of the rate of protein synthesis or an increase in the rate of degradation.

From the observation of Table-2,6 it would be seen that during pod development of C.cajan and P.tetragonolobus, protein contents increased till later phase of development. It was observed that protein accumulation during earlier stage of development was less. Adverse observations were found in the case of pod covers. Protein contents in pod covers were decreased during later phase of pod e.g., 30 days after anthesis.

According to observation Table-2,6 the highest values of proteins in C.cajan and P.tetragonolobus are at the maturity period in seeds of C.cajan, the highest protein value being 25.63 gm 100^{-1} g dry weight 40 days after anthesis. In pod cover of C.cajan steady decrease in protein content is seen. In P.tetragonolobus seeds protein levels

are 42 gm 100^{-1} g dry weight, while in pod cover it decreases. P.tetragonolobus have higher protein level than C.cajan seeds.

If the above values are compared with protein fractions in chickpea and pigeon pea (Singh and Jambunathan, 1982) it will be found that whole seed protein in chickpea and pigeon pea is less than seed proteins of P.tetragonolobus.

TABLE 26: Total proteins in developing seeds and pod covers of C.cajan and P.tetragonolobus.

Days after anthesis	TOTAL PROTEINS gm 100^{-1} g DRY WEIGHT			
	C.cajan		P.tetragonolobus	
	Seeds	Pod covers	Seeds	Pod covers
10	18.38	8.75	7.00	42.50
15	19.39	14.06	-	-
20	20.94	13.44	11.20	33.04
25	21.88	12.50	-	-
30	22.81	11.25	26.80	21.84
35	24.38	10.31	-	-
40	25.63	9.38	41.40	22.40
50	-	-	41.60	04.46
60	-	-	42.50	04.48

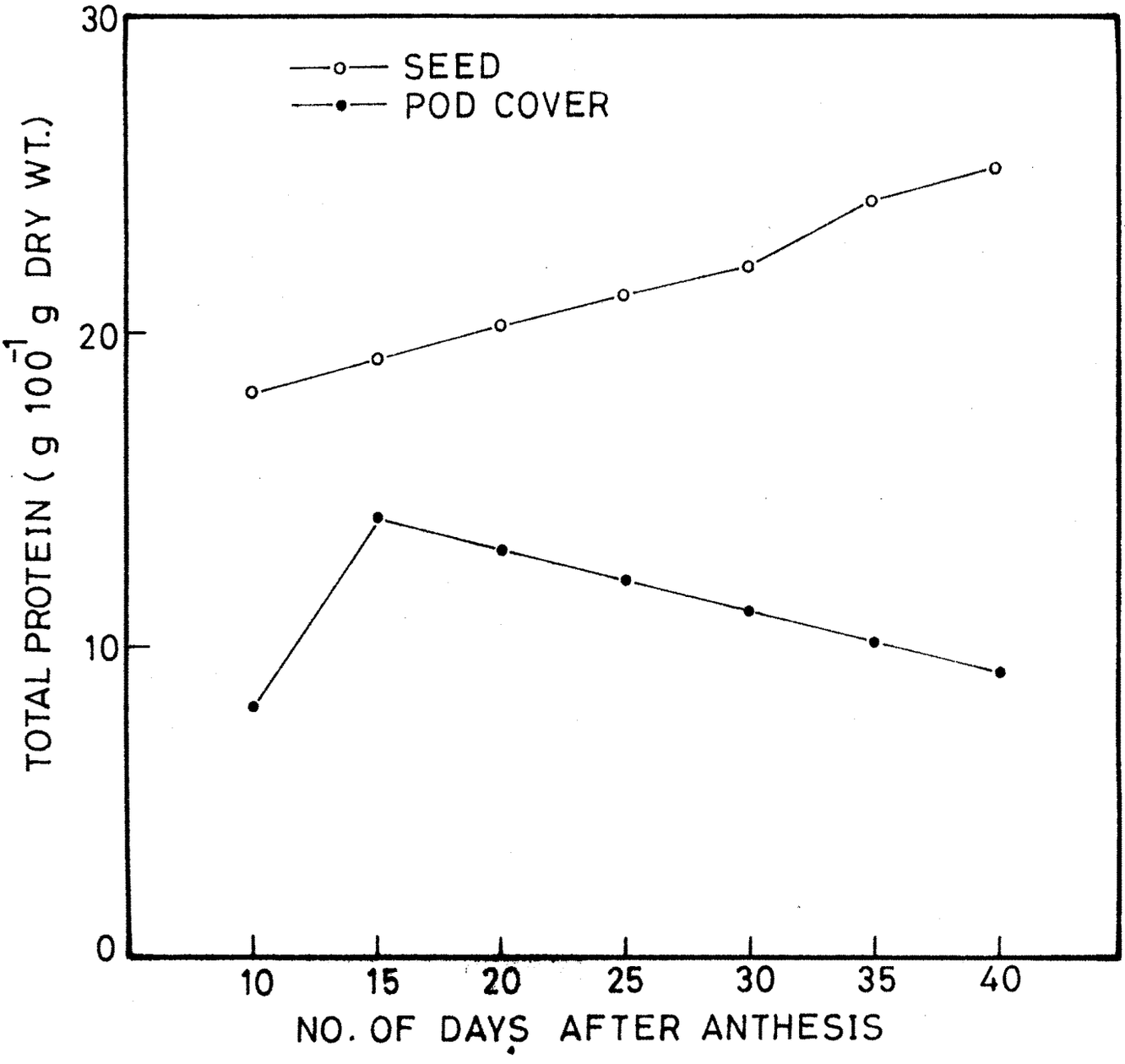

FIG. 2·6 TOTAL PROTEIN IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

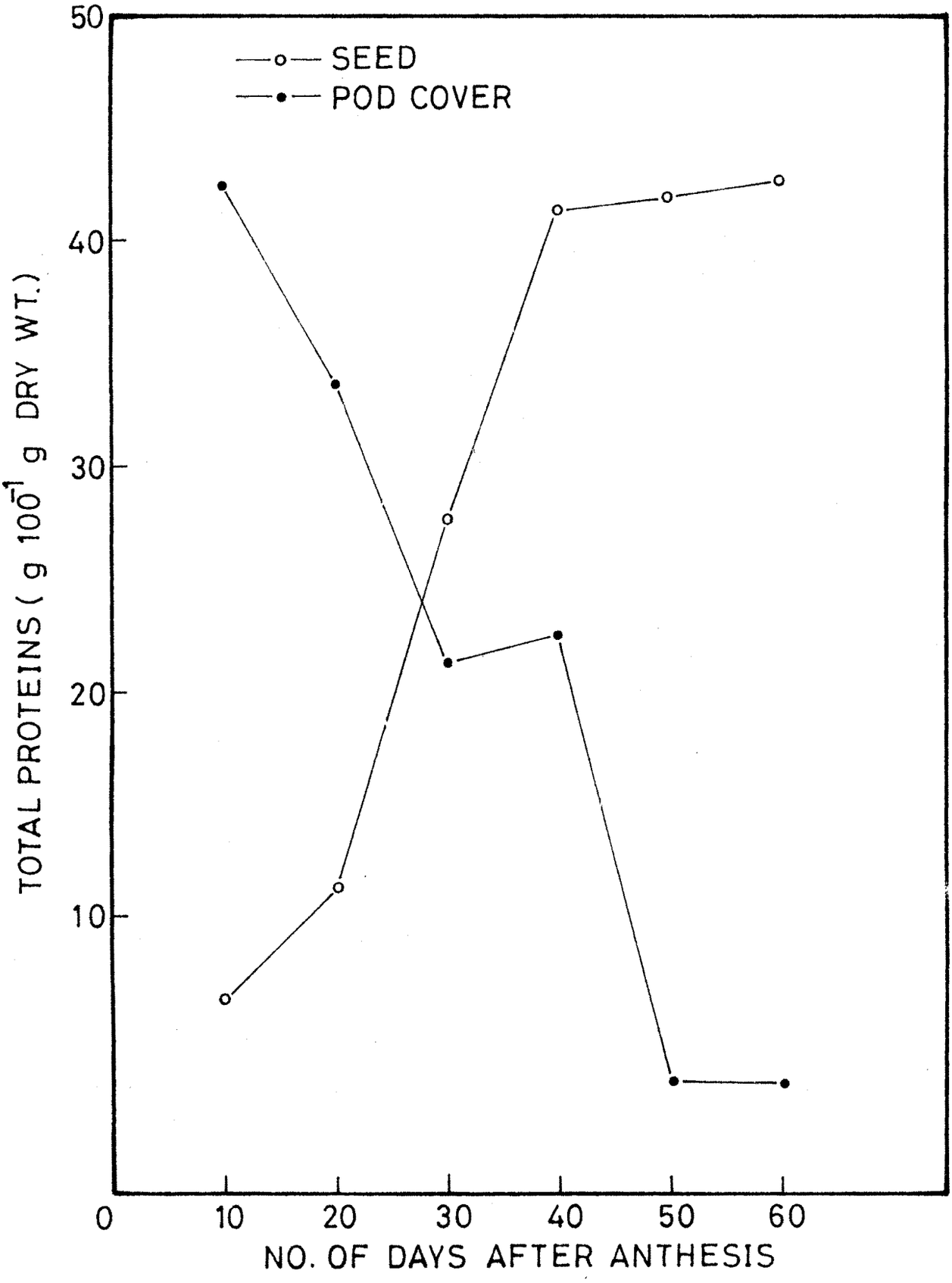

FIG. 2·7 TOTAL PROTEIN IN SEED AND POD COVER OF *P. tetragonolobus* DURING POD DEVELOPMENT.

TABLE-2.7: Distribution of protein fractions in different components of chickpea and pigeon pea.

Legume/component	Protein (%) (N X 6.25)
Chickpea	
Embryo	52.1
Cotyledon	24.8
Seed coat	16.4
Whole seed	21.3
Pigeon Pea	
Embryo	99.6
Cotyledon	22.2
Seed Coat	4.9
Whole Seed	20.5

PROLINE

Proline is one of the 20 amino acids which form building-blocks of proteins. It is widely distributed in different plant discs. In stressed green leaves glutamate is generally considered as the major carbon donar for protein synthesis (Barnett and Naylor, 1968; Morris et al., 1969). According to Parker (1958) and Suzuki (1982) for storage of nitrogen in trees like black locust and white mulberry proline is an important component. Young parts of a plant synthesizes less proline than old one.

Water often limits crop growth and development. The plants

response to water stress is relative to its metabolic activity, morphology, storage, organ growth and yield potential. Water stress affects chlorophyll synthesis. Enzyme nitrate reductase should reduce activity, but some hydrolytc enzymes (amylase) show increased activity. The level of proline as compared to other amino acids is increased under stress.

Some plants may have different degrees of drought resistance depending upon the ecological conditions of plants and their adaptation. Mukherjee et al. (1982) tried to find out relation between proline accumulation in plants and ecological habitats of those plants.

Karadge and Chavan (1979, 1981) have studied the proline content in the leaves of Arachis hypogia and their values are very close to those in P.tetragonolobus. They further (1983) stated that the leaves, stem and roots of Sesbania aculeata and S.grandiflora contain very high level of proline. Murumkar (1984) observed little proline in the roots of chickpea. Boadess et al. (1978) have reported that proline dependent O_2 uptake subject to respiratory control ADP/O_2 ratios with proline as substrate were intermediate between ratios obtained with exogenous NADH and Malate pyruvate as substrate.

Isotope studies showed proline metabolism to be dependent on O_2 but not NAD. Swamy et al. (1990) stated that proline contents decreased in leaf disc of cow pea during senescence in darkness. Treatment with calcium chloride or abssicic acid also affected the proline level.

According to Steward et al. (1966) sugars play a role in proline accumulation in wilted leaves. He further stated that sucrose is precursor of proline via α-ketoglutarate and glutamate. Tan and Hollorn (1982) studied proline accumulation in stressed seedling leaf and its association with a number of physiological responses introduced by water deficit in 14 wheat cultivars. It was noted that there was a positive correlation between sugar and proline contents in water stressed wheat leaves. Roger (1986) noticed that if soil moisture was depleted it caused an increase in the levels of sugars which led to accumulation of free proline. If the sugar is involved in the formation of proline then there is decline in sugar content.

ROLE OF PROLINE IN PLANT METABOLISM:

Proline incorporates into peptide linkage serving as a precursor to peptidyl bound to hydroxy proline and it accumulates when some plants are exposed to diverse biological and environmental stresses. It is important to study various roles played by proline in plant metabolism.

(1) Amino Acid Metabolism:

In the biosynthesis of various amino acids L-proline plays an important role. Steward et al. (1977) had invented that ^{14}C proline can be easily converted into amino butyric acid, alanine, aspartic acid, glutamine and glutamic acid in turgid tissues of Hordeum vulgare.

Vallee et al. (1977) observed that when proline is externally supplied to apical tissues or leaf of tobacco plants after floral induction then there is increase in the free glycine concentration in these plant parts. Due to different environmental conditions (light, dark and varying concentrations of CO_2 and O_2) the exogenous proline appeared to modify reaction of glycolate pathway.

(2) Nitrogen Pool and Chlorophyll Synthesis:

Britikov et al. (1970) observed that proline plays a role of nitrogen source in leaves and young inflorescence of Chenopodium album L. It was further observed that proline may be utilized for Chlorophyll synthesis (Duranton and Mille, 1962; Perdrizet, 1963).

Bengston et al. (1978) suggested that proline may serve as a reserve substance for the synthesis of Chlorophyll upon the relief of stress. This conclusion was based on observations that chlorophyll formation, which took place in fully turgid Triticum aestivum L. leaves were pretreated with a water stress. They found that the longer the stress treatment, the greater effect on chlorophyll formation.

(3) Substrate for Respiration:

Britikon et al. (1965) studied germination in Anirrhinum pollen and suggested that proline could serve as a respiration substrate in this germination. Britikon and Linskens (1970) further examined the effect of proline on O_2 uptake by spinach leaves and styles

of maize and petunia. They concluded that proline stimulated normal tissue during respiration and eliminated the inhibitory effects of glutamic acid - Keto-glutrate, citrate and malate on O_2 uptake.

(4) Cell Wall Biosynthesis:

In cell wall biosynthesis proline plays important role. Steward and Chang (1963) suggested that externally proline was first incorporated into various soluble proteins which could be separated electrophoretically and then with greater hydroxylation into the more insoluble protein that constituted to the bulk of protein of the cell and its organells. The hydroxy-proline was associated with wall. King and Bayley (1965) identified hydroxyproline in the cell walls of Hellanthus tuberosus tubers, Pea stems and oat coleoptiles 70 per cent of hydroxyproline was found in green leaves of broad bean associated with cell wall and 30 per cent was in cytoplasm (Puztai et al., 1971). Proline participated in the cell wall formation.

(5) Growth and Yield:

1975

Proline is essential in Zea Mays mutant (Gavazzi et al.). They further suggested that essential amino acid is not synthesized by mutant but proline was the only component which was required to bypass the lethal growth stage and allow growth to the normal seedling. Prell (1977) observed proline content in leaves of 10 days old tomato plants and cumulative fruit yield of 175 and 192 days old plants.

(6) Twining Habitat:

Molin (1977) observed that in twining shoots of Periploca gracea L. proline was longitudinally distributed and accumulate maximum in the internode No. 3 and were different in the concave and convex halves; a transverse gradient existed. Proline level decreased during rolling of barley leaves and proline may play an important role in this process.

(7) Bathurst (1954) observed the pollen of grass species for 17 amino acids, amino-nitrogen, aminonic urea, glutanine, asparginie total soluble nitrogen and total nitrogen. Britikov and Musatova (1964) have invented soluble proline content from pollen and Pistil and further determined that free proline content was less in pistil than that in pollen.

(8) Drought Resistance:

Drought has generally been accepted as deficiency of available soil moisture which produces water deficit in plants sufficient to cause a reduction in growth. The drought lowers the water status and leaf water potential of various plant parts and in particular, the leaves. The water stress brings about marked decrease in relative water content of the leaves, which further leads to proline accumulation.

Patil et al. (1984) were unable to find out relations between

water content and free proline in five maize genotype and observed that there was marked increase in proline contents in all genotypes under drought. During February, March, April and May moisture tension is high as compared to monsoon months and a general increase in proline content.

Relationship Between Leaf Potassium and Calcium Status and Free Proline Level:

Anatomical as well as physiological functions of the pericarp resemble those of leaves. K^+ and Ca^{++} are essential micronutrients for their metabolism. K^+ generall forms weak complexes in which it is readily exchangeable (Wynjones et al., 1979). K^+ plays a major role in plant metabolism, particularly in photosynthesis, respiration and ATP generation. The K^+ reduces water doses by transpiration (Brag, 1972). It also plays a vital role in stomatal movements; opening occurs as the turger pressure in guard cell is increased by increasing concentration of K^+ in the cells. Ben-Zioni et al. (1971) have suggested that K^+ is a mobile element which helps upward translocation of nitrate in the entire plant. Transport of nitrogen through phloem is also medicated by K^+.

Ca^{++} is a larger divalent cation; calcium uptake rate is restricted and is coupled to metabolic processes. Calcium is less mobile from cell to cell and in the phloem it is very low. Ca^{++} functions mainly outside the cytoplasm in the apoplast. It is essential to regulate membrane permeability and strengthening of cell walls. Calcium with

pectate forms calcium pectate in the cell wall. It is also important in determining the susceptibility of the tissue to fungal infections and in the ripening of fruits. The pollen tube growth is also dependent on the presence of Ca^{++} in the growth medium (Mascarenhas and Machlis, 1964).

Calcium deficient plants show increase in respiratory rates and enhances the net rate of protein synthesis (Faust and Klein, 1974). Ca^{++} protects cell membrane under various stress conditions. It increases the activity of amylase phospholipase and ATP-ase (Wyn Jones and Lunt, 1967). ? / Wyn-Jones et al 1967 ?

Ca^{++} concentration in plants varies from 0.1 to 5.0 per cent dry weight, depending on growing conditions, plant species and plant organ. Mineral constitution in the leaves show seasonal variations. Mitchel (1936) showed that K^{+} content was decreased from June to October while the Ca^{++} content increased sharply. It was increased from May onwards and reached its peak in December (Jones and Parker, 1951). K^{+} was increased during November to March.

Proline level was maximum in Eucalyptus leaves in the month of March. In the months of July and August free proline and Ca^{++} levels were very low in the leaf tissue. According to the above investigations it is difficult to correlate mineral element status with free proline in the leaves as both nutrient accumulation and proline accumulation are regulated by a number of factors.

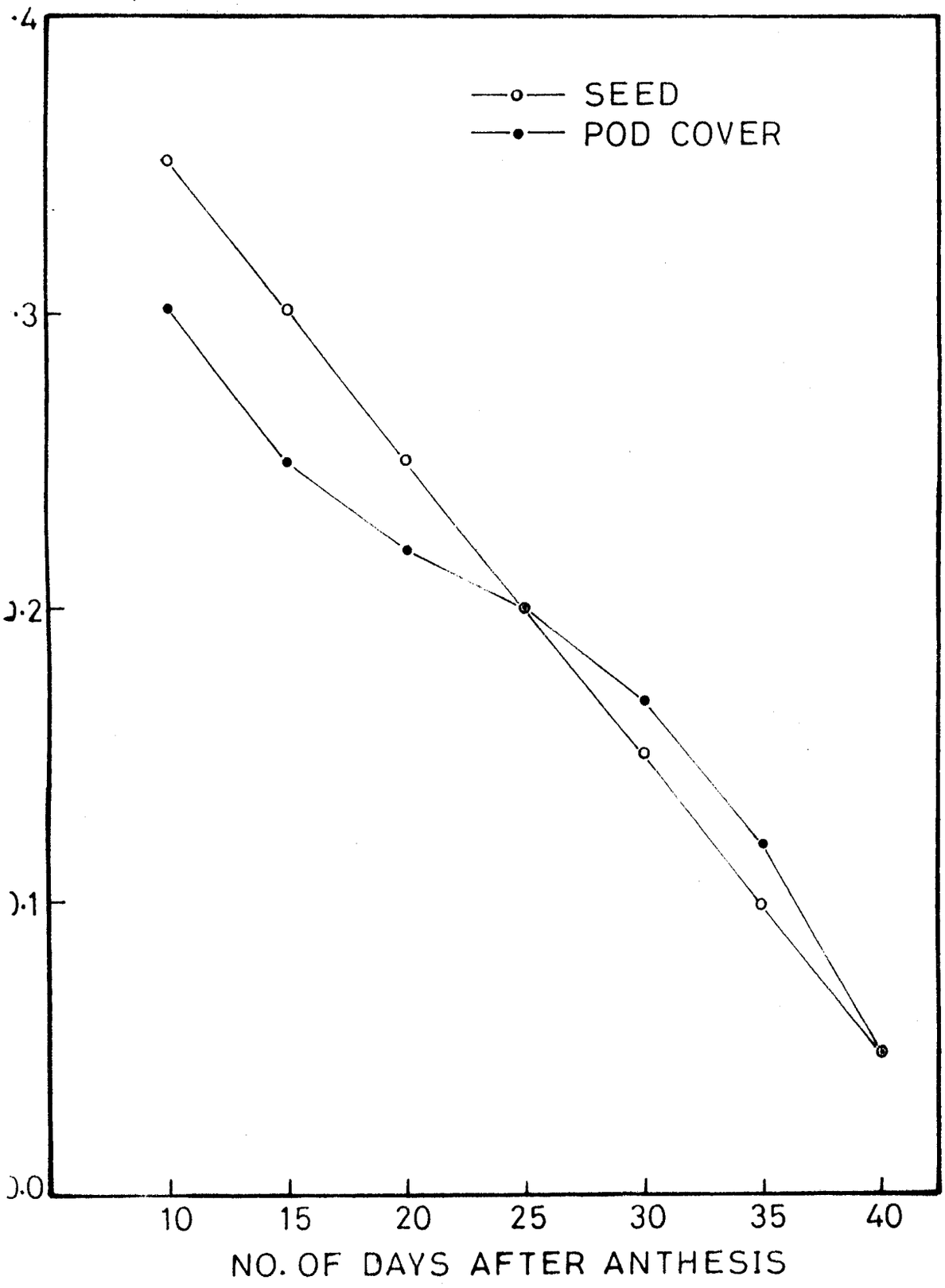

FIG. 2·8 FREE PROLINE IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

TABLE-2.8 : Proline content of C.cajan seed and pod during their development.

Days after anthesis	Proline g 100 g^{-1} Dry Weight	
	Seed	Pod cover
10	0.35	0.30
15	0.30	0.25
20	0.25	0.22
25	0.20	0.20
30	0.15	0.17
35	0.10	0.12
40	0.05	0.05

From Table-2.8 it was evidenced that proline content was high during early stage of pod development. As the pod develops, moisture content decreases. Comparing moisture content in seed and pod development seeds shows more accumulation during early days of pod development. In pod cover proline content 30-35 days after anthesis was more than seed.

CHLOROPHYLLS

Results and Discussion:

The chlorophylls present the principal class of pigments responsible for light absorption in photosynthesis and are found in all photosynthetic organisms. There are a number of different

types of Chlorophylls, such as Ch.. a, b, c, and e. Chlorophylls 'a' and 'b' are predominant in higher plants.

Luthra et al. (1983) analysed photosynthetic rates and enzyme activities of leaves, developing seeds and pod covers of C.cajan.

The rates of dark respiration, photosynthetic carbon dioxide fixation and activities of ribulose 1, 5 disphosphate carboxylase, PEP carboxylase, glycollate, oxidase and NADH-malate dehydrogenase were studied at weekly intervals after anthesis in substending leaves, developing seeds and pod covers of C.cajan. On the fresh matter basis, leaves photosynthesized 4 to 12 times and 7 to 9 times more than the seeds and pod covers respectively. Leaves exhibited higher rates of respiration than the seeds and pod covers. The activity of each enzyme, except malate dehydrogenase was in general higher in pod cover than in the leaves and seeds. Thus CO_2 metabolism of the pod cover plays a significant role in seed development of C.cajan.

Singh and Pandey (1980) studied the production and distribution of assimilates in chickpea. According to them leaves are the main photosynthetic organs and pod cover photosynthesis was not a significant source of assimilate for seed development. However, pod cover contributed a significant amount of photosynthate to its own growth. The leaf axil of the pod was the major source of photosynthates to the pod (Pandey et al., 1978). Bangal et al. (1983) have

reported that, the pod cover contributed to grain development during later growth period.

Pigments in C_3 and C_4 plant species were studied by Holden (1973). He stated that, ratio of chlorophyll a/b ranges from 3.1 to 5.6 for C_4 dicot plants while the range for C_3 dicotyledons was 2.5 to 3.7. C_4 plants are relatively deficient in Chlorophyll 'b'. Holden (1973) further observed that, Chlorophyll 'b' content was similar tin C_3 and C_4 grasses on fresh weight basis but value of Chlorophyll 'a' in C_4 grasses is more than 30 per cent. Sestak (1966) has shown difference between photosystem I in C_3 and C_4 plants, and he observed that, photosystem I in C_4 plants is governed by Chlorophyll 'a' which is more active than C_3 plants. Chlorophyll a/b ratio in C_3 plants is lower than in C_4 plants. Value of chlorophyll a/b ratio in the case of C.cajan was from 0.01 to 1.70 in seeds and from 0.56 to 1.97 in pod cover, which is relatively less than the value of C_3 plants. From the above findings it can be concluded that photosynthesis in seeds and pod cover of C.cajan is probably similar to that in C_3 plants and not in C_4 plants.

Chlorophyll content varies from plant to plant and species to species. Karadge and Chavan (1980) have recorded 280 to 230 mg chlorophylls 100^{-1} g fresh tissue and chlorophyll a/b ratio 2.25 to 2.72 in groundnut. According to Deshpande (1981) chlorophyll content of C.cajan leaf is 392.74 mg 100^{-1} g fresh tissue with Chlorophyll a/b ratio 3.37.

The loss of Chlorophyll from fruit may be synchronous with ripening or it may occur only in earlier stages of ripening. The pigment change occurs mainly in the Chloroplasts, of course, converting them from green Chloroplasts with grana into Chromoplasts with more disperse thylakord membranes. Dodge (1970) showed that the shape of chloroplast in Betula leaves changes with a decrease in its volume. Danuta (1976) reported that during ripening of fruits, the thyllakoids are disrupted, stroma lamella get degraded and accumulation of osmophilic globules takes place. These changes affect the photosynthetic electron transport and photophosphorylation.

As the number of days increased after anthesis, there was a decrease in the level of Chlorophyll in seed and pod cover of C.cajan (Table-2.9). Green pod changes to yellow particularly during later stages, probably because of increased accumulation of anthocynine, and other flavocoides. Chlorophyll loss may be due to increase in Chlorophyllase activity in the fruit.

The timing of Chlorophyll degradation in senescence leaf may not always correlate with other aspects of its functioning. Thomas and Stoddert (1977) have stated that the loss of Chlorophyll during leaf senescing maynot be inseparably linked to the overall process of senescence. Recent work with Phaseolus valgaris and Hordium vulgare (Brown and Woolhouse, 1982) indicated that the degradation pathway of chlorophyll may involve an hydroxylation at C_{10} to yield Chlorophyll a-1. Observation of Chlorophyll a, content rises, passes

through a maximum and then decreases. It is not a stable degradation product but is an intermediate which is itself degraded. Hydroxylation was the first step in Chlorophyll breakdown. The subsequent steps in the degradation of Chlorophyll are unknown.

It was evidenced that the Chlorophyll content of the C.cajan pod covers increased to maximum at 20 days after anthesis and then it declined very rapidly (Table-29). The Chlorophyll content of the seeds, however, was maximum only at the 10 days' stage. Later it was continuously decreased with age of the seed. Flinn and Pate (1968) also observed that Chlorophylls were lost very rapidly during the final phase of pod drying.

From the observation it was also evident that the Chlorophyll a:b ratio in pod cover was always higher than that in the seeds of C.cajan during their development and with the development this ratio was continuously or rather uniformly decreased with increase in the age of pods (Table-29). The observations indicate that Chlorophyll 'a' was affected relatively more than Chlorophyll 'b' in both seeds and pod cover.

TABLE-2.9: Changes in chlorophyll content of C.cajan seeds and pod cover during their development.

Days after anthesis	Chlorophyll in seeds (mg 100^{-1} g fresh weight)				Chlorophylls in pod cover (mg 100^{-1} g fresh weight)			
	Chl. a	Chl. b	Total Chl.	Chl. a/b	Chl. a	Chl. b	Total Chl.	Chl a/b
10	19.08	6.74	25.82	1.71	8.60	9.90	18.50	1.97
15	14.20	5.34	19.54	1.32	12.40	7.84	20.24	1.71
20	09.12	3.26	12.38	0.79	14.95	7.58	22.53	1.58
25	05.31	1.73	07.04	0.61	05.57	4.27	09.84	1.31
30	02.28	1.59	03.87	0.20	04.56	3.60	08.16	0.87
35	01.11	0.87	01.98	0.01	02.02	2.67	04.69	0.56
40	-	-	-	-	-	-	-	-

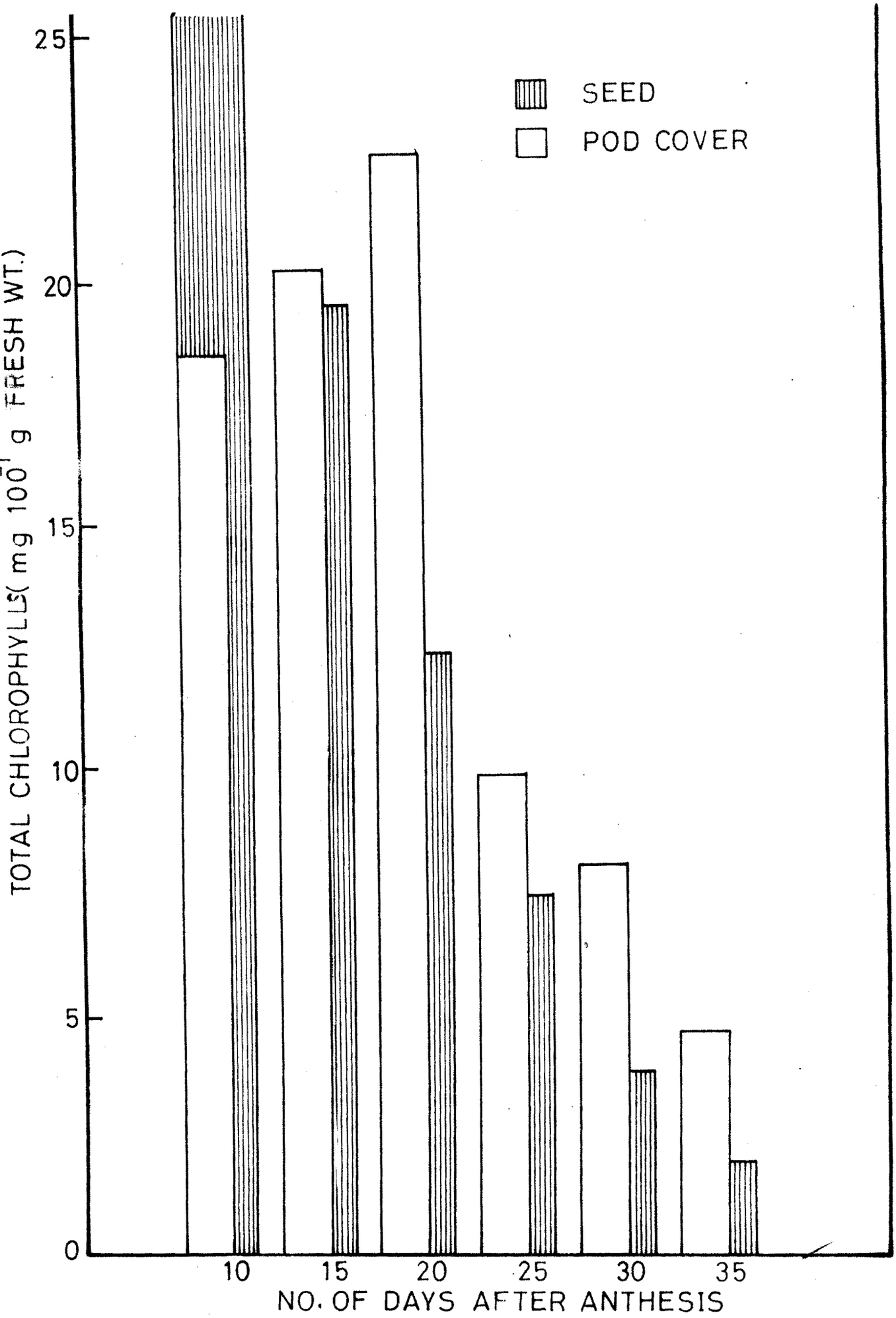

FIG. 2.9 TOTAL CHLOROPHYLLSIN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

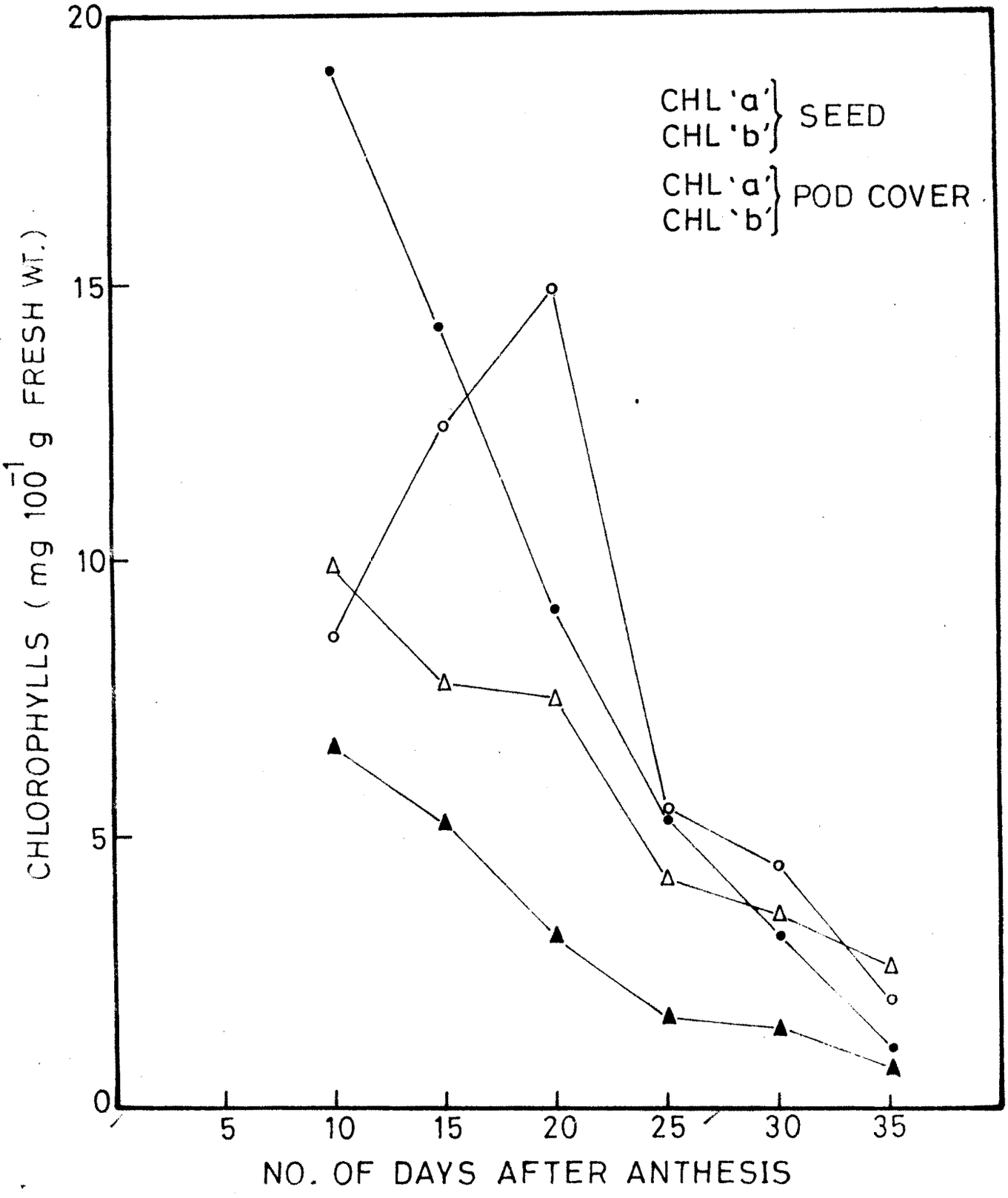

FIG. 2·10 CHLOROPHYLL IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

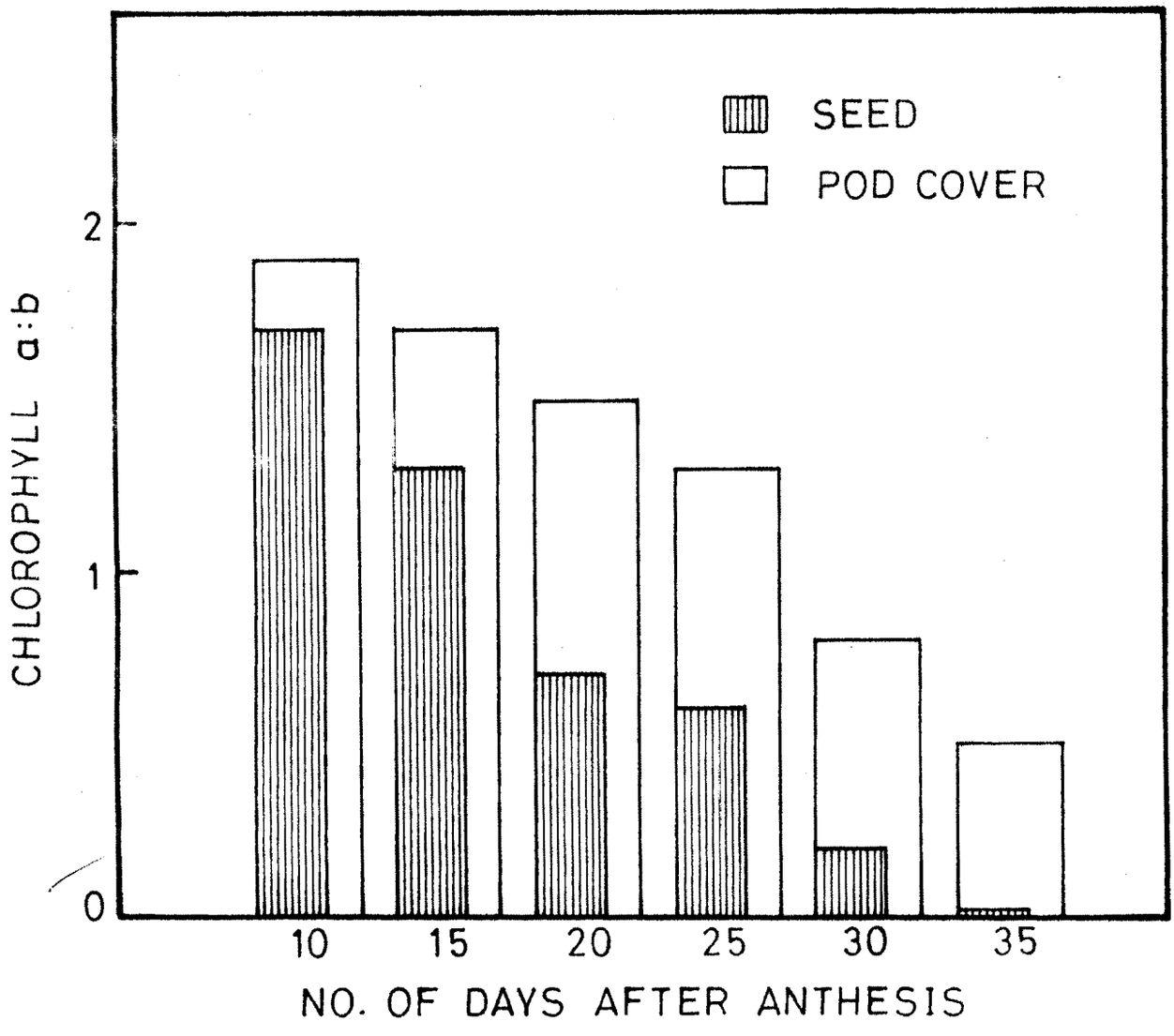

FIG. 2·11 CHLOROPHYLL a:b IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

CARBOHYDRATES

Carbohydrates provide the main source of energy for respiration and show marked changes. Kidd and West (1930) showed that about 16-20 per cent of carbohydrate reserves of apples was utilized between harvesting and death by fungal disease whether the apples were stored at 2.5°C, 10°C, 22.5°C. Fidler (1960) concluded that production of CO_2 by apples could be accounted for by the combine loss of carbohydrates and acid.

Chickpea (Cicer aretinum), Pigeon pea (Cajanus cajan) and Bonavist bean (Dolichos lablab) legume seeds provide food of high nutritive value to both human and their domestic animals. They are particularly valuable in some tropical and sub-tropical regions, where there is an acute storage of animal protein, e.g., the tre-tre fly zone in South Sudan. Their carbohydrates especially the unavailable carbohydrates receivec little attention. Pritchard et al. (1973) defined unavailable carbohydrates as the difference between total and unavailable carbohydrates. The unavailable carbohydrates consisting of water-soluble fractions and three insoluble structural fractions "hemicellulose", "cellulose" and "lignin".

Dunphy (1972) has carried out the carbohydrate analysis at various stages of development in soybean. He reported that substantial quantities of labile carbohydrates were accumulated in the leaves, petioles and stems prior to seed development and water utilized in the seed growth. Kadam et al. (1981) have reported that winged

Kadam & Salunkhe, 1981

beans (seeds) contain 25 to 45 per cent carbohydrates. Sugar, and total sugars in the seeds of C.cajan and P.tetragonolobus increased after 30 days of anthesis while the level of insoluble carbohydrates was increased 25 days after anthesis (Table-).

In the pod cover of C.cajan and P.tetragonolobus the level of reducing sugars was increased from 10 to 30 days after athesis and later decreased; while the amount of total sugars and insoluble carbohydrates was increased from 10 to 40 days after anthesis and during later developmental stages it was decreased. As we know, carbohydrates are the sight for respiration. The carbohydrate content of pod cover is more than that of seeds. Pod cover plays a major role in respiration of pod and higher level of carbohydrates it is in consistance. Carbohydrates from pod cover are probably translocated to seed for its further development. As pod becomes senescent and dry there is decline in the photosynthetic activity due to decreased activity of photosynthetic enzymes like RuBP case and PEP case.

Starch During Pod Development:

Starch is most abundant in legume carbohydrates and varies from 24 per cent in wrinkled pea to 56.5 per cent in Pinto bean, Soybean, Lupine and Winged bean have the lowest starch content (0.2 to 0.5 per cent). Pinegina and Klimenko (1961) reported 38.1-60.4 per cent starch in seeds and vegetative parts of pea. According to Sajjan and Wankhede (1981) carbohydrate composition of Winged

bean shows 36.2 per cent of starch. Garcia and Palmer (1980) determined starch content in different cultivars of Winged beans based on iodine staining and enzymatic determination. They reported that there was no starch in five cultivars of Winged bean. However, in most other food legumes starch is the predominant carbohydrate. The discrepancies observed in these earlier two reports may be due to the method of extraction or determination of starch. Hulme (1958) found that percentage of starch in the fresh weight of apple fruit increases to a maximum during development on the tree and then decreases till at harvest when very little starch is present.

Bhamri and Malik (1983) traced starch sugars and reducing sugars, free amino acids in the pod wall and seeds of P.vulgaris at different developmental stages. It was found that starch content was increased till maturity while total sugars were increased till 12 days after anthesis.

Bhamri et al. (1983) studied the biochemical changes in seeds and pod cover of Phaseolus vulgaris and traced the level of starch, sugars and reducing sugars in seed and pod wall at different developmental stages. He concluded starch content increased till maturity; while total sugar increased till 12 days after anthesis. Smith (1974) studied synthesis of Nucleic acid protein and starch in cotyledons of Pisum arvense and stated that the full development of grain was evident in the cotyledon cells, 20 days after anthesis, so that the main period of starch accumulation over the period 20-35

days was essentially a filling and maturation of already formed grains. Therefore, little starch is laid down although cell walls become impregnated with hemicellulose. Flinn and Pate (1968) observed in Pisum sativum starch accumulation usually continues until seeds are fully ripe and during the early stages of its synthesis and substantial drop occurs in the level of sucrose and other sugars in the seeds.

Khatra et al. (1987) studied the pattern of dry matter accumulation in developing fruit parts of early and late maturing Pigeon pea. They also compared dry matter accumulation in Pigeon pea early 15 days and late 21 days in the pod wall, seed coat and seed throughout the reproductive development. Significant water loss and dry matter accumulation in the fruit parts began much earlier in early 15 than in late 21. The pod wall accumulated starch reducing sugars and nitrogen compounds upto 21-28 days after anthesis with subsequent distribution to the developing seed. The pod wall had lost a significant amount of dry matter when the seeds reached maximum dry weight, fruit of early 15 days accumulated more photosynthates than late at all comparable stages.

Sugars:

The occurrence of oligosaccharides of raffinose family of sugars in food legumes is well documented (Salunkhe, 1982). These sugars (varbasco, stachyose and raffinose) are important in nutrition because they cause flatulence in man and animals. Claydon (1975) observed that raw matured winged beans cause abdominal pains

indicating the presence of flatulence factors. Garcia and Palmer (1980b) anc Sajjan and Wankhede (1981) reported that the level of oligosaccharides in winged bean seeds was about the same as found in soybean.

Sugar level increases during development except during the first week in both apples and pears, fructose is the predominant sugar, while lesser amount of glucose and sucrose and relatively small quantities of the corresponding heritals are present. Significant changes in carbohydrate constituents have been reported during pod and seed development in chickpea seeds by Singh et al. (1981b). They observed that during grain development percentage of soluble sugars decreased upto 28 days after flowering and then remained constant. During this process starch content increased upto 28 days. As starch content increased, soluble sugars declined during 14-28 days. After anthesis this period was known as period of intense biochemical activities. Sinha and Prasad (1978) reported the three soluble sugars, namely, sucrose, fructose and glucose to be completely utilized by A.flavus in infested seeds. According to Dign Peng, Shi-Jeon Sung, C.C. Mack (1989) in developing seeds of lima bean sucrose metabolism was dominated by sucrose synthetase pathway but in seedling embryos both the sucrose synthase pathway, acid invertase were active. UDPase activity was low and seemingly insufficient to complete for UDP during sucrose metabolism in seed development ol germinatior.

Changes in carbohydrates during ripening of tropical and sub-tropical fruits are of similar nature (Biale, 1960). Bananas are harvested before they are edible and during ripening there is a decrease in starch and increase in fructose, glucose and sucrose in fruit pulp. Hemicellulose of the pulp declines from 9 per cent to 12 per cent as reserve carbohydrate in banana metabolism. Dunielson (1959) reported that sugar concentration in the water phase of ripening pea seeds was constant during the greater parts of the maturation process and then fell rapidly.

The sucrose concentration of the pod wall, seed coat and cotyledons remained constant at moderate and high total crop respiration rates. Adenylate energy charge remained relatively constant in the pod wall, seed coat and cotyledons with changes in fruit respiration. Sucrose transport to the fruit took priority over the maintenance of reserve sucrose pool size. Remobilization of seed starch only occurred when the sucrose pool of the pod wall was not maintained. Accumulation of starch and reducing sugar in pod wall takes place upto 21-28 days after anthesis (Khatra et al., 1987).

Bidhan Chandra (1987) found that the embryo of legumes attained physiological maturity much before the pods become fully mature. Studies on physiology of seed development in V. mungo at different days after anthesis have been initiated with a view to understanding the role of starch accumulation in embryo development and increase in seed dry out in this crop. Large number of flowers were tagged

on the day of pod opening. Pods were harvested at 4, 8, 12, and 16 days after anthesis and increase in starch and decline in sugars till maturity (Yoshida et al., 1972).

Bidhan Chandra (1987) observed that soluble sugars in Urad bean increased upto 8 days after anthesis and then showed a sharp decline after 12 days and decreased further till maturity. However, a rapid increase in starch content was observed from 8 days after anthesis to maturity. The increase in starch content accompanied by decrease in soluble sugars during the same developmental period indicated that soluble sugars were utilized for the synthesis of starch. Since increase in seed dry weight parallels the accumulation the accumulation of starch, accumulation of starch is responsible for increase in dry weight of the seeds during development and maturation. Similar results were obtained with Chickpea (Singh and Jambunathan, 1982).

Sharma and Pant (1978) analysed six varieties of pigeon pea (C.cajan) and found them to contain 53.59 per cent starch and 2.4 per cent soluble sugars (dry weight). During their germination the degradation of starch was mainly due to activities of phosphrylase and α-amylase. Two distinct phases of starch depletion were recognised, first slow during which phosphorylase activity was at its maximum and second rapid which coincided with the maximal activity of α-amylase.

Sharma and Pant (1978) further stated that the change

in dry weight of pigeon pea varieties consisted a slow phase of decrease during initial 6 days and fast phase thereafter. The starch content varied between 53-59 per cent of dry weight. Starch was found to be degraded slowly during the intial 2 days followed by its rapid utilization. the residual starch on 12th day was only 3-9 per cent of the original starch content. The content of soluble sugars present was very low. On the second day it was maximum, decreased slowly upto 4th day followed by a rapid decline. Dian-Peng et al. (1989) found that in developing seeds of lima bean sucrose breakdown was dominated by the sucrose synthase pathway, but in the seedling embryo, both the sucrose synthase pathway and acid invertase were active, UDPase activity was low and seemingly insufficient to compete for UDP during sucrose metabolism in seed development or germination. In contrast, both acid and alkaline pyrophosphatases were active in seed development and germination. The set of adaptive enzymes identified in developing seeds were sucrose synthatase, PPi-dependent phosphofructokinase, plus acid and alkaline pyrophosphates and the adaptive enzymes identified in germinating seeds including the same set of enzymes plus acid invertase. The set of maintenance enzymes identified during development in the dry seed, and during germination were UDP glucopyrophosphorylase, neutral invertase, ATP and UTP depencent fructokinase, glucokinase, phosphoglucomutase, ATP and UTP-dependent phosphofructokinase and sucrose synthatase. Bhivere (1984) worked on P.vulgaris and recorded that reducing sugar content was lowered during the latter phase of pod development.

the level of non-reducing sugars increases upto senescence of pod drying.

In seeds of starch storng legumes such as P.sativum, P.vulgaris, Vicia faba starch granules are accumulated in cotyledons. Starch degradation takes place in living storage cells. In the case of germination starch storing cotyledons can convert carbon from heterotropy to photosynthetic competence during development of seedlings. Reserve starch is mobilized from the inner part of the granule. Harris (1976) reported that there was apparently little breakdown at the outer surface. During legume germination reserve starch degradation is accompanied by an increase in extractable activities of starch metabolizing enzymes such as endoenzyme, amylase and phosphorylase; further X-glucon biosynthesis and formation of an additional population of starch granules was observed during degradation. In P.vulgaris and Vicia faba the number of reserve starch granules per cell remained constant during the early stages of germination and small particles increased by one or two orders of magnitudes. Large and small starch granules are located in perenchyma of cotyledons.

The present observation Table-~~11~~ 5.4 2.10 evidenced that reducing sugars, total sugars and starch are more between 15 to 25 days after anthesis and then rapid decline in C.cajan pod cover. Accumulation of reducing sugars in pod cover is less while starch and reducing sugar accumulated till the maturity in seed of C.cajan.

P.tetragonolobus seed showed rapid increase in starch

TABLE-7.10: Reducing sugars, total sugars and starch in the seed and pod cover of C.cajan and P.tetragonolobus during pod development.

Days after anthesis	Carbohydrates in C.cajan g 100^{-1} g fresh weight						Carbohydrates in P.tetragonolobus g 100^{-1} g fresh wt.					
	Seed			Pod cover			Seed			Pod cover		
	Red sugar	Ins. sugar	Total sugar	Red sugar	Ins. sugar	Total sugar	Red sugar	Ins. sugar	Total sugar	Red sugar	Ins. sugar	Total sugar
10	0.60	5.2	1.2	0.40	3.0	0.90	0.32	3.20	0.72	9.19	7.4	5.81
15	0.28	6.0	1.6	0.42	3.0	0.85	-	-	-	-	-	-
20	0.40	6.1	1.5	0.45	3.2	1.00	0.21	4.46	0.66	6.42	7.09	7.95
25	0.45	7.4	1.5	0.36	3.1	1.00	-	-	-	-	-	-
30	0.49	7.6	1.7	0.33	2.8	0.92	0.22	9.26	0.75	10.21	12.69	7.04
35	0.52	8.0	1.6	0.28	2.4	0.97	-	-	-	-	-	-
40	0.62	10.0	1.0	0.22	2.1	0.25	0.23	14.48	1.33	6.57	8.78	7.00
50	-	-	-	-	-	-	0.23	16.74	1.65	0.97	5.51	1.41
60	-	-	-	-	-	-	0.23	20.8	2.62	0.31	2.48	1.10

TABLE-~~12~~ 5.2 2.11: Carbohydrate of food legumes.

Name of legume	Carbohydrates in Percentage	
	Starch	Total sugar
Winged bean seeds	-	3.4
Smooth peas	36.9-48.6	5.3-8.7
Wrinkled peas	24.0-36.6	10.[illegible]-15.1
Great Nort. beans	44.0	9.9
California small White bean	57.8	7.7
Red kidney beans	31.9-47.0	8.0
Navy beans	27.0-52.7	5.6-6.2
Pinto beans	51.0-56.5	6.7
Pink beans	42.3	-
Black eyed beans	41.2	-
Black gram	32.2-47.9	3.0-7.1
Bengal gram	37.2-50.0	3.5-9.0
Mung bean	37.0-53.6	3.9-7.2
Red gram	40.4-48.2	3.5-10.2
Soybean	0.2-0.9	5.3
Broad bean	41.2-52.7	3.1-71.
Lentil	34.7-52.7	4.2-6.1
Cowpea	31.5-48.0	6.0-13.0
Lupine seeds	0.3-3.5	7.4-9.5

From Reddy, N.R., Pierson, M.D., Sathe, S.K. and Salunkhe (1984).

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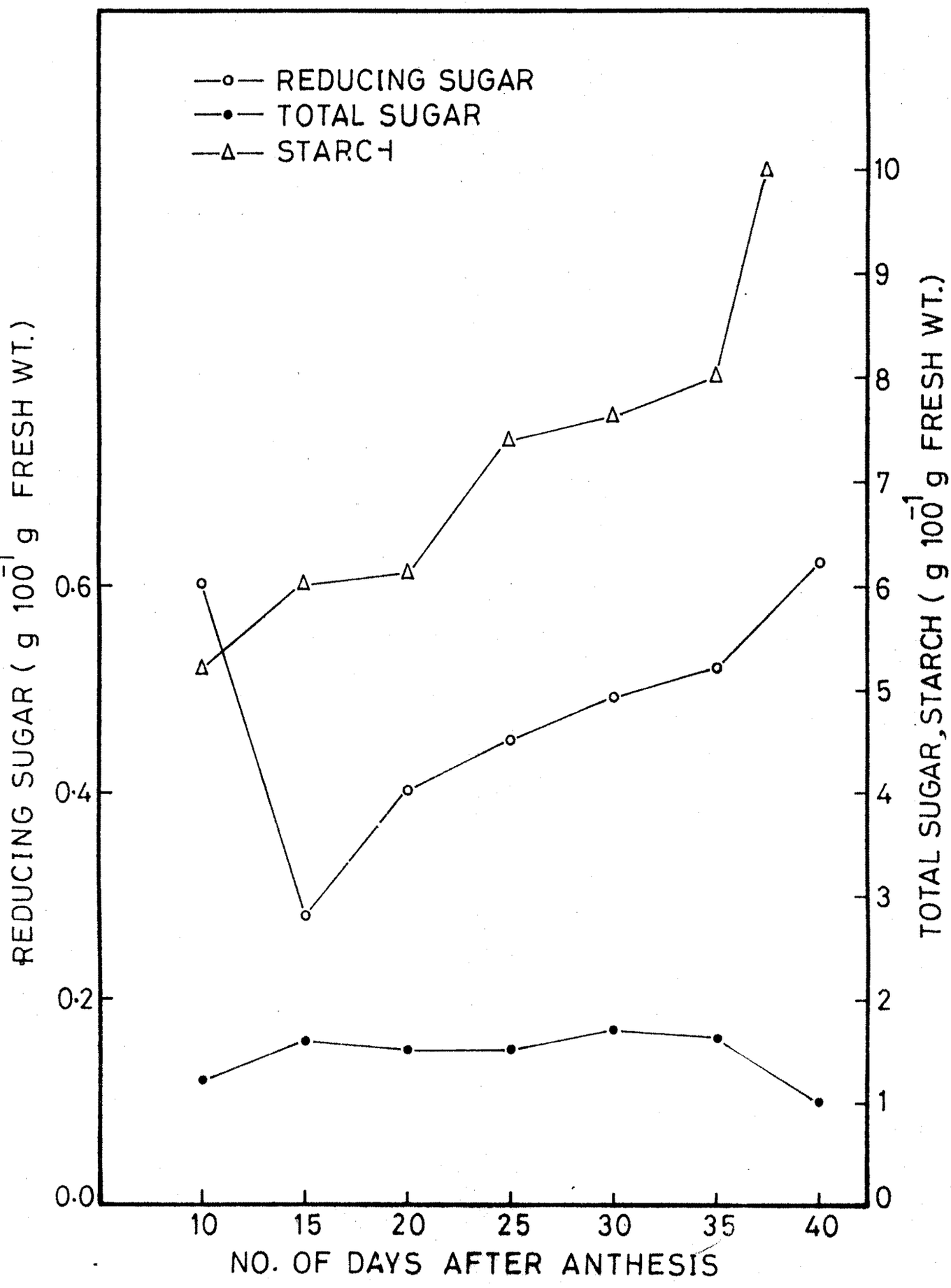

FIG. 2·12 REDUCING SUGARS TOTAL SUGARS AND STARCH IN THE SEED OF C. cajan DURING

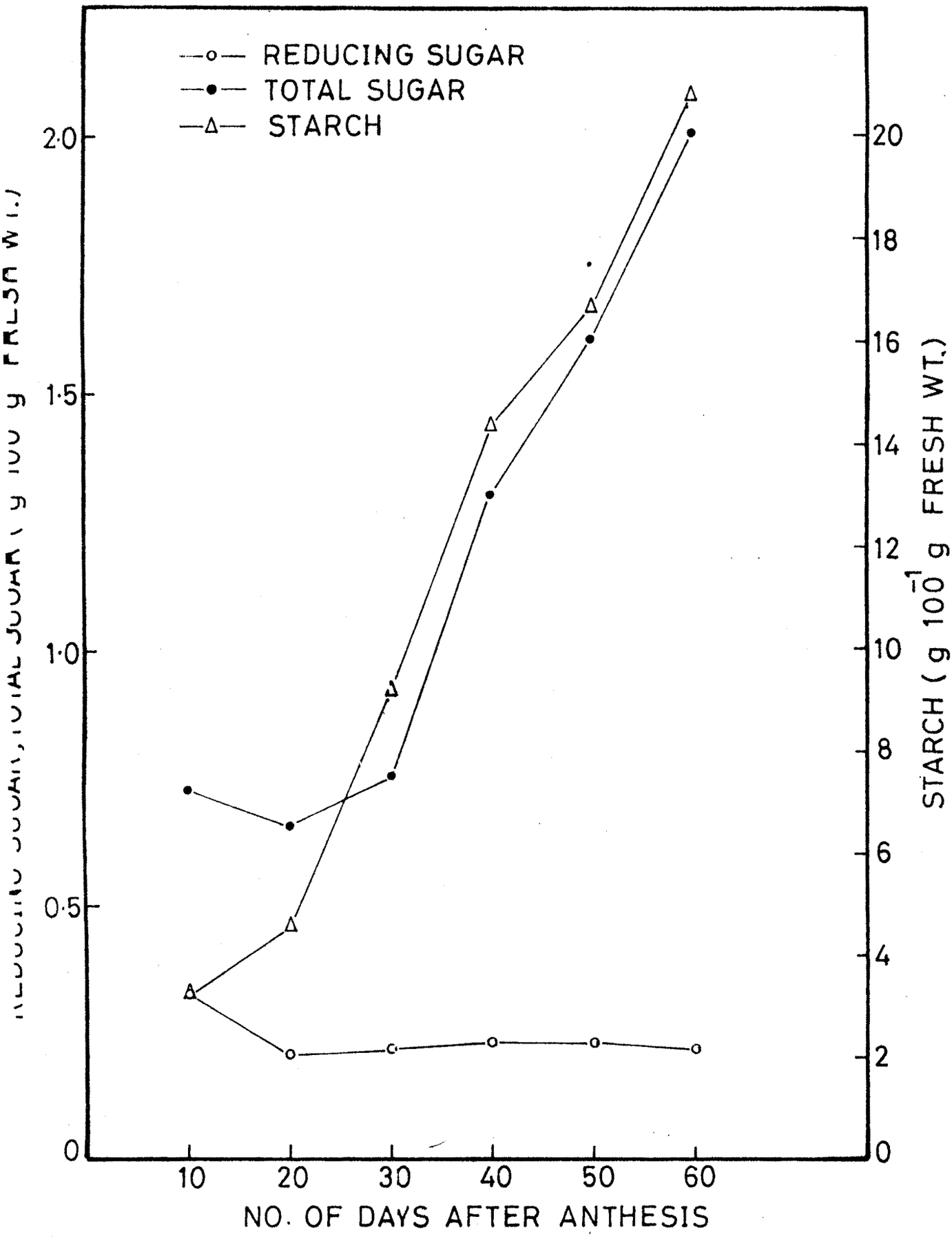

FIG. 2.13 REDUCING SUGARS TOTAL SUGARS AND STARCH IN THE SEED OF P. tetragonolobus DURING POD DEVELOPMENT.

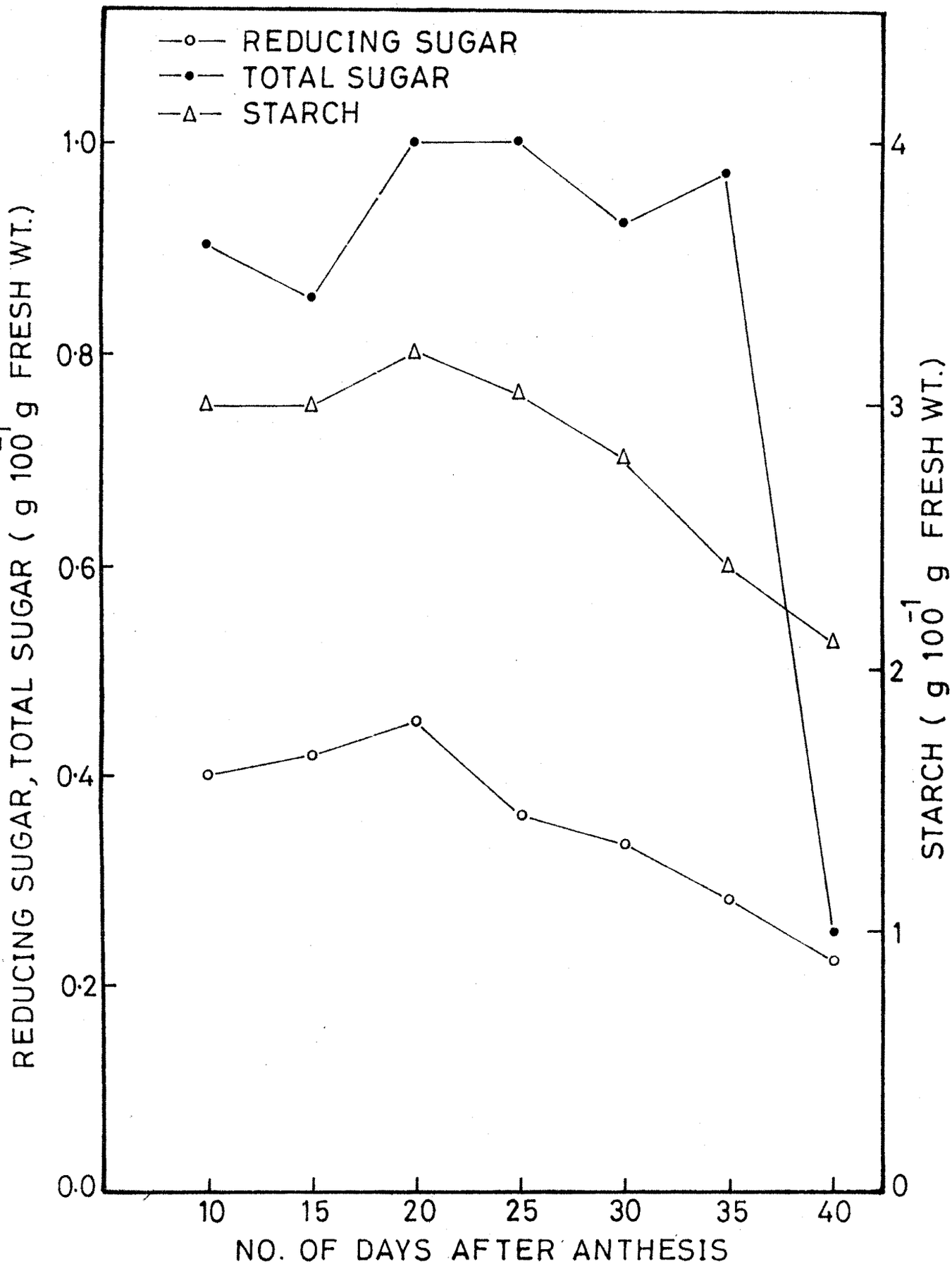

FIG. 2·14 REDUCING SUGAR TOTAL SUGAR AND STARCH IN THE POD COVER OF C. cajan DURING POD DEVELOPMENT

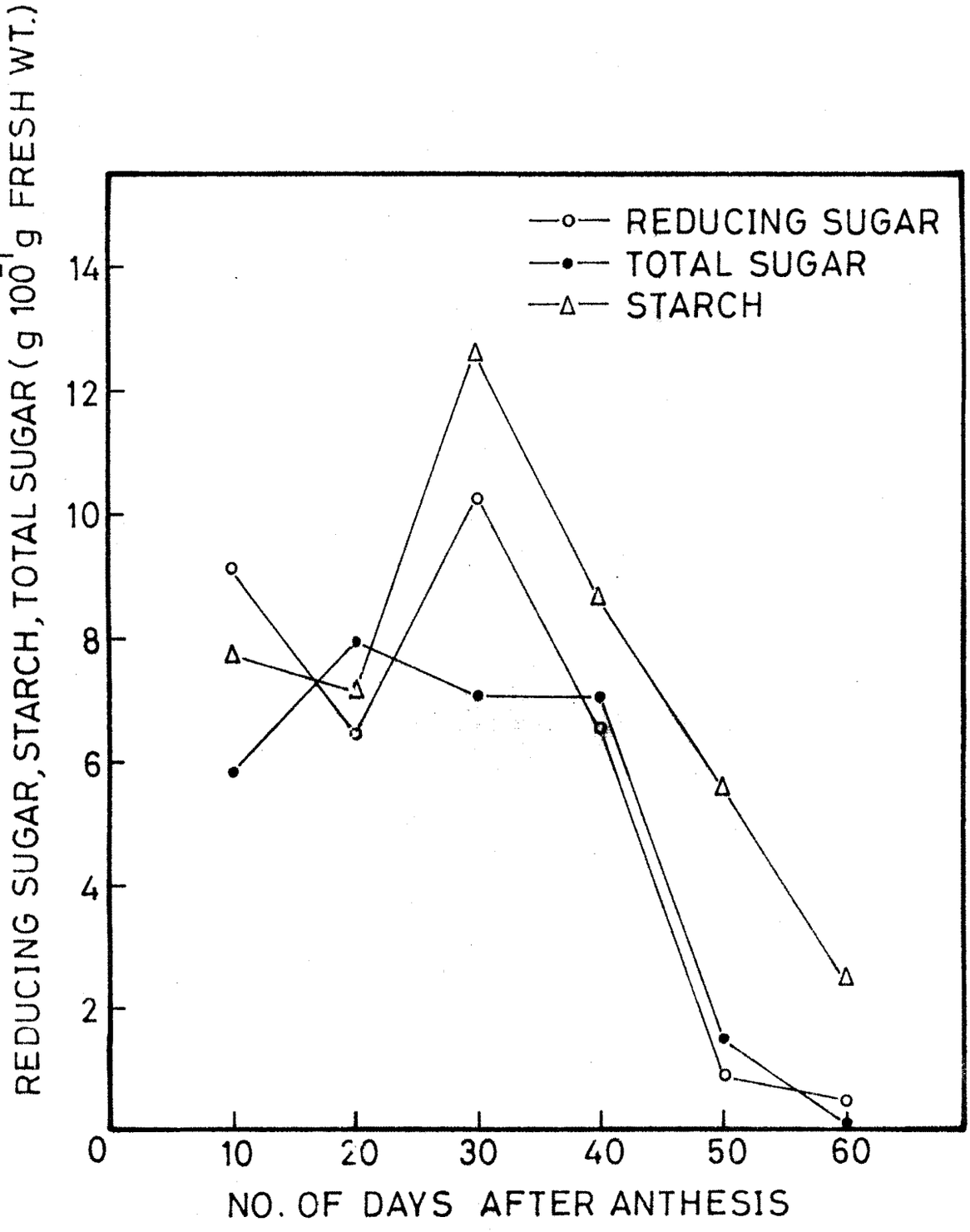

FIG. 2·15 REDUCING SUGARS, TOTAL SUGARS AND STARCH IN THE POD COVER OF *P. tetragonolobus* DURING POD DEVELOPMENT.

and total sugar till the end of maturity while reducing sugar declined at maturity of seed. Pod cover of P.tetragonolobus show steep decline in reducing sugars, total sugars and starch at maturity.

According to Table-2.10 our findings show starch contents are highest at 40 days after anthesis in C.cajan and P.tetragonolobus seeds. But according to Table-2.11 by Reddy et al. (1984) value of Insoluble sugar is more than our findings. Red gram contains highest percentage of insoluble sugar, while value of total sugars in pod cover of C.cajan and P.tetragonolobus and different legumes are nearly equal.

AMYLASE

Amylase is one of the important enzyme systems of the carbohydrate metabolism. It is hydrolytic type of enzyme which brings about hydrolysis of amylase, a component of starch to liberate sugar units of glucose. Germinating starchy seeds are rich in the enzyme system. During germination starch is broken down to sugars which are liberated during respiration and are used for further development of seedlings.

Among the main enzymes involved in starch breakdown in plants are α and β amylase and starch phosphorylase. Juliana and Vener (1969) investigated the role of these enzymes in starch metabolism of germination pea and concluded that α-amylase was major enzyme responsible for initiating the degradation of starch granules

and β-amylase and starch phosphorylase. Juliana and Verner (1969) investigated the role of these enzymes in starch metabolism of germinating pea and concluded that α-amylase was major enzyme responsible for initiating the degradation of starch granules and β-amylase and starch phosphorylase acted on the product of hydrolysis of the starch by α-amylase.

Davis and Bill (1978) found that occurrence of α-amylase was also present in the axis portion of ungerminated pea seeds. The occurrence of this enzyme was demonstrated with crude homogenate (also containing β-amylase) using 3 different methods. The increase in total amylase activity (primarily β-amylase) paralleled germination. The accumulation of α-amylase activity was not initiated for an additional day. The increased α-amylase activity was found in the etialated stem, the distribution being higher in growing than in non-growing portion.

Wilson et al. (1978) found that β-amylase from Barley was capable of hydrolysing isolated starch from soybean seed imbibed for 18 hr. Mature soybean seeds contain high levels of β-amylase (Adams et al., 1980; Birk and Waldman, 1965) but α-amylase and starch phosphorylase levels are very low (Peat et al., 1949; Vin and San (1948). Soybean seeds contain very little starch at maturity. They contain 10-15 per cent starch at earlier stages of development and germination. Smirnova and Konikova (1962) have showed that age of the seeds does not affect amylase content in starch grains

and legumes.

The variation in starch and soluble sugar content in phosphorylase and amylase activities in cotyledons of germinating seeds of C.arietinum are determined by Tarargo (1979). He found that α-amylases were chiefly responsible for amylase activities. Phosphorylase played an important role during the 1st and 2nd days of germination, but it was regarded to occupy a secondary position as the amylase activity increased. The increase in α-amylase activity during germination was due to de novo synthesis of the two isoenzymes of amylase. Since both are inhibited by cycoheximide and actinomyces D, this de novo synthesis depends on same embryo produced factor unreplaceable either by GA or by kinetin.

Amylase plays a major role during germination of seed and α-amylase activity is more during the process. As pod develops and matures, α-amylase activity is reduced. Rauf (1980a) observed a continuous decline in α-amylase activity in developing pods and seeds of groundnut. Sharma and Pant (1978) studied amylolytic enzymes in the germinating non-germinating seed. Activity of amylase increased slowly during the initial 6 days followed by a steep rise till 10th day; then there was a rapid fall. Enzyme activity in non-germinated seeds increased slowly in the 1st two days and reached the maximum by the 4th day. There was a slight decrease in the activity by 6th day followed by a rapid decline upto 12th day when its level was lower than that found in the non-germinated seeds. Thus the period of higher amylase activity in the developing seeds and pod

cover coincided with a decrease in the phosphorylase activity.

Hildbrand and Hynouitz (1981) studied the role of α-amylase in starch metabolism in soybean during seed development and germination. They observed that the total amylase activity peaked just prior to seed maturity and drafted off slowly. However total activity of the enzyme in chesnut and Atona was very low throughout seed development and germination.

From observation Table-2.12 it is evident that α-amylase activity is more in the pod cover than in the seed of C.cajan. The α-amylase activity is constant from 5 to 10 days after athesis. It increased from 15 to 25 days after athesis and then gradually declined till the maturation of pod.

OBSERVATION TABLE 2.12

Days after anthesis	α-amylase seed	Activity value pod cover
10	10	4
15	16	10
20	20	11
25	9.9	5
30	4	3.3
35	1.6	1.1
40	1.4	0.7

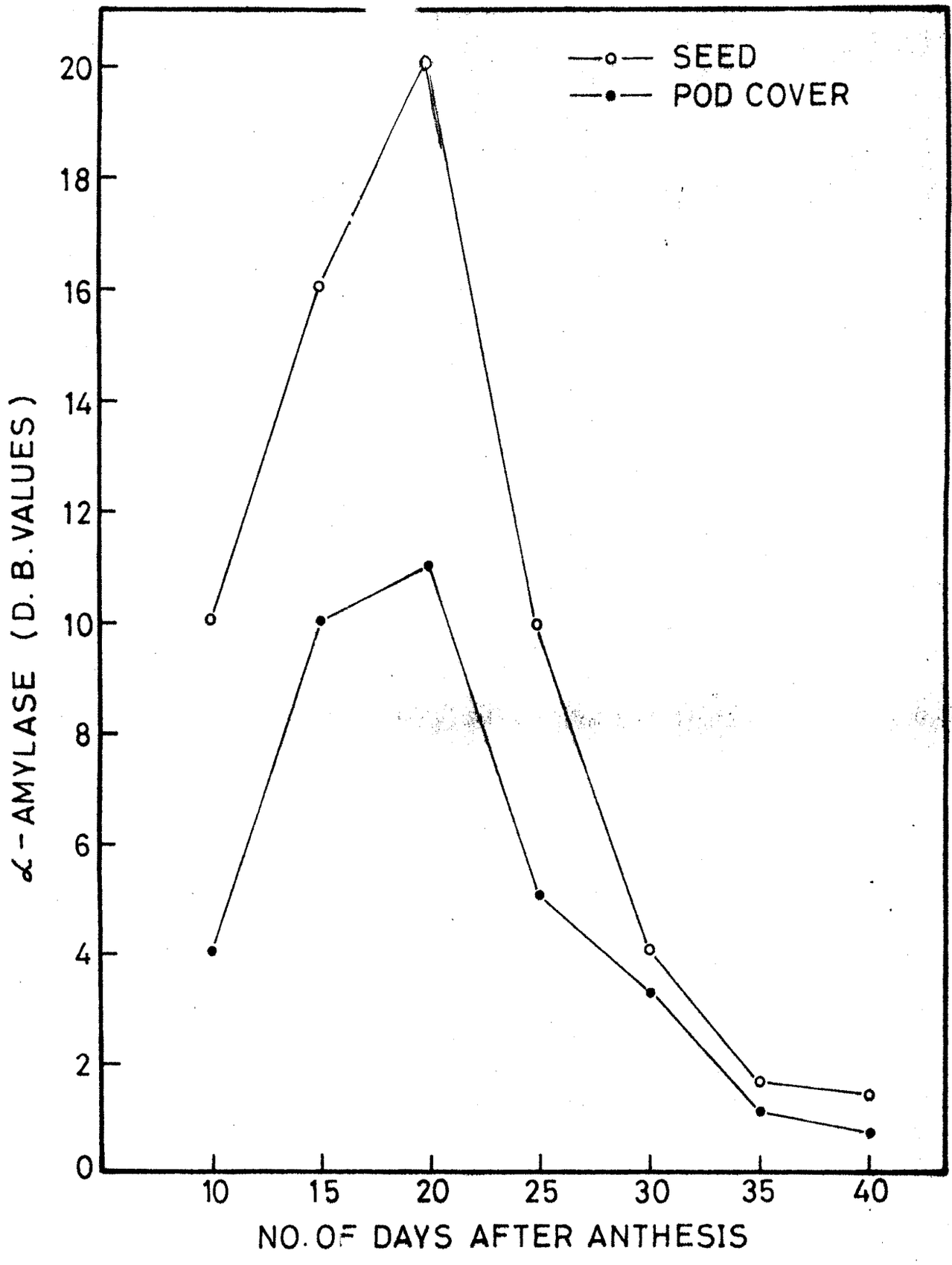

FIG. 2.16 α-AMYLASE IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

POLYPHENOLS

Results and Discussion:

Polyphenols are widely distributed in plants. They are commonly known as tannins. Recently considerable importance has been attached to this group of substances as some of the phenolic compounds act as phytohormones. The functions of most of these substances are obscure, and many appear, at present, simply the byproducts of metabolism.

Polyphenols have been shown to be antiphysiological substances in food legumes. These are known to reduce digestibility of protein and availability of minerals and vitamins (Salunkhe et al., 1981). Salunkhe et al. (1982) further observed that these are mainly located in the seed coats with low or negligible amounts in cotyledons. Polyphenols are known to interact with proteins, leading to either inactivation of enzymes such as trypsin or chymotrypsin or making protein insoluble. Polyphenols are known to inhibit several enzymes including pectin esterase, cellulases, amylases, α-galactosidases, lipase and several proteolytic enzymes. In addition to reduced protein digestibility, polyphenolic compounds inhibit other hydrolytic enzymes such as β-amylase and lipase. The intense colours and astringent flavours imparted by these compounds are not always desirable and may contribute towards an off-flavour or colour. This could lower consumer acceptance of this particular food.

There was significant reduction in the polyphenol content

of the seed during its maturation such a decrease during fruit maturation was reported by Goldstein and Swain (1963). Chavan et al. (1979a) & 1979b also observed a decrease in the plyphenol content in the developing seeds of Sorgham. Senguptá (1990) studied the changes of phenolic compounds in groundnut seed during their development and germination. It was found that the phenolic content declined slowly during peg penetration to initial seed development. There was a sudden increase in it towards the last stage of maturity in both the dormant and non-dormant cultivars. Phenol content at maturity was higher in seeds of non-dormant cultivars than in dormant ones. During seed germination, the rate of leaching of phenolic compounds from the seed was faster from the non-dormant than dormant seeds. Thus seed dormancy in groundnut does not seem to be related to high phenol content but the seed germination in non-dormant cultivars than in dormant ones. During seed germination, the rate of leaching of phenolic compounds from the seed was faster from the non-dormant than dormant seeds. Thus seed dormancy in groundnut does not seem to be related to high phenol content but the seed germination in non-dormant cultivars appeared to be due to faster release of phenols from the seeds by leaching.

Rao and Decsthale (1988) estimated polyphenoloxidase (PPO) activity in whole seeds and seed functions of germinating C.arientinum, C.cajan, V.radiata and V.mungo. Increase in PPO activity during germination was greatest in V.mungo and the least in C.arientinum.

Embryos were richest in PPO activity. Seed coat fractions lacked PPO activity except in V.mungo, C.cajan and C.arientinum seed coats contained a water-soluble and heat stable PPO inhibitor.

Griffiths (1960) studied the polyphenols of Theobroma and Herrania seeds. The major polyphenols of Theobromecacao were found in related SPP which are not used for cocoa manufacture. The polyphenols of Theobroma are sufficiently characterised to permit the identification of seeds of unknown SPP by paper chromatography of their phenolic constituents. It is clear that the polyphenol content in C.cajan is more in the seeds and pod covers developed for 10 to 20 days after anthesis (Table-2.13). Then it was decreased as the pods matured. The polyphenol content of the seeds of C.cajan is less as compared to that in pod cover.

The observed decrease in polyphenol content may be due to polymerization of the existing phenolic compounds producing high molecular weight insoluble polymers. The possibility of binding of polyphenols with other organic substances (Salunkhe et al., 1981) or alteration in the chemical structure of polyphenols that render them incapable of giving a chemical colour reaction measured by Folin-Denis reagent used in the present investigation cannot be ruled out.

TABLE-2.13 Polyphenol content of C.cajan seed and pod cover during their developmert and that of P.tetragonolobus during seed maturation.

Days after anthesis	Polyphenols (g 100^{-1} g fresh weight)				
	C.cajan		P.tetragonolobus (seed)*		
	Seed	Pod cover	Sri Lanka	Nigeria	Indonesia
10	1.45	1.85	-	-	-
15	1.25	1.50	-	-	-
20	1.00	1.25	-	-	-
25	0.75	1.00	-	-	-
30	0.50	0.75	-	-	-
35	0.25	0.50	-	-	-
40	0.01	0.25	3.2	2.6	2.5
50	-	-	1.7	2.1	2.0
60	-	-	1.5	1.6	1.8
70	-	-	1.4	1.6	1.6
80	-	-	1.5	1.4	1.4

*Salunkhe et al. (1981) ?

1982 ?

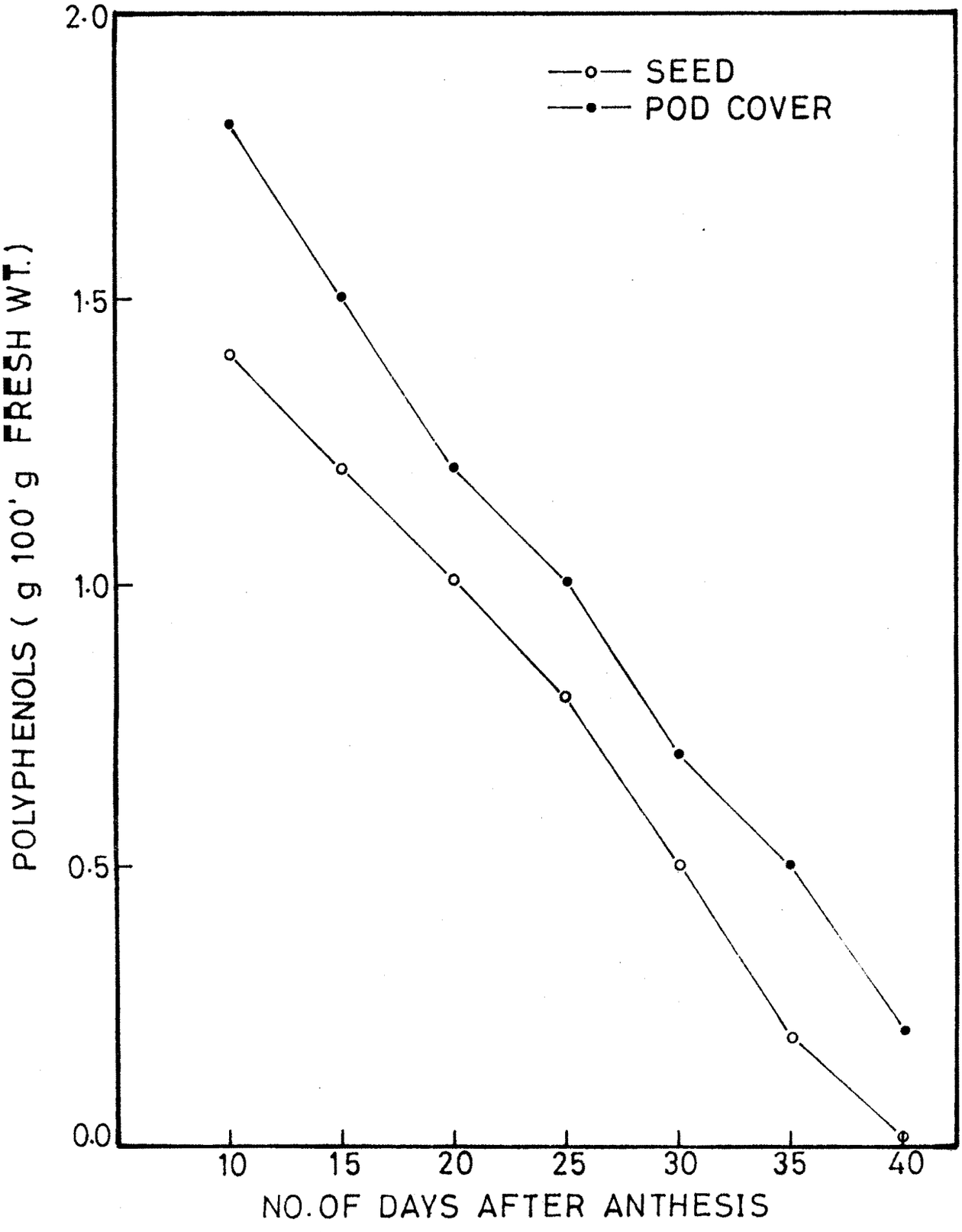

FIG. 2·17 POLYPHENOLS IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

MINERAL NUTRITION OF PLANTS

From long past, investigations about the nature and usefulness of some nutrient materials necessary for the growth of plants have been tried. A lot of experience is required for the preparation of a suitable soil substratum by adding manures, for the growth of crop plants. The practice of 'Crop-rotation' was known and used by Indian farmers since long past. It was known that by growing some leguminous crop every year in the same field, the fertility of its soil was not minimised. But the field soil loses fertility after few years of the cereal crops are grown in this soil every year because the crop gradually depletes the soil of its nutrient elements, year after year. This is why the 'Crop-rotation' was practised by growing different types of cereals and leguminous crops alternately on the same soil so that the fertility of the soil is maintained year after year.

The uptake of mineral nutrients from the soil by plants may be studied in different ways often with different results leading to differences in the conclusions and the recommendations made by (Brown (1979). The chief difficulty which arises is due to the fact that the actual amounts of various mineral nutrients taken up may not be needed by the plant. High uptake may be reflection of luxury consumption rather than real need. While there may well be difficulties in establishing the optional level of uptake for specific nutrients it is rather easier to establish the levels at which deficiency

of the element or elements occurs.

The needs of the plant for mineral nutrients is neither uniform nor constant throughout the life of the plant. This applies perhaps more so to the pulses than other crops. The mineral requirements of crop plants such as the pulses can be studied basically in two major ways. Plants may be grown in entirely experimental conditions and the changes in the pattern of mineral uptake were studied.

I: Composition of Plant-ash:

Plant body is made up of water and other solid material organic and inorganic compound. Water alone makes 10 to 95 per cent of the body weight of the plant. After decomposition of organic mater inorganic salts left over are generally silicates, phosphates, sulphates, carbohydrates and oxides of many mineral elements such as Sodium (Na), Potassium (K), Calcium (Ca), Iron (Fe), Aluminum (Al), Magnesium (Mg), Manganese (Mn), Zinc (Zn), Copper (Cu), Molybdenum (Mo) and in addition Boron (B), Titanium, Vanadium, Cobalt, Rubidium, Nickel.

Elements like C, H, O important nutrients in the production of carbohydrates which enter into the composition of cell wall and protoplasm and elements like S, P, N are required in the formation of protein which is important constituent of protoplasm; these elements are known as protoplasmic elements. Elements like Ca, Mg are important

constituents of Chloroplast.

II: Classification of Mineral Requirements of Plants:

Many mineral elements are necessary or 'Essential' for the plant but there are some which although absorbed by the plant do not play any role in plant metabolism. Based upon this kind of necessity and usefulness, elements can be placed into the following categories:

(A) Invariable - Primary Elements: These are the elements which are found essentially in all plants making between 1 to 60 per cent of the total weight of the plant and include such elements as H, C, O, N, P etc.

(B) Invariable - Secondary Elements: Present in all plants such as Ca, Na, Mg, K, Fe, S, Cl, etc.

(C) Invariable - Microconstituents: Present in all plants e.g., Cu, B, Si, Mn, F, I etc.

(D) Variable Secondary Elements: found in certain plants in extremely low concentrations include Li, Ls, Ag, Be, Sr, Cd, Ge, Sn, Pb, As, Cr, Co, Ni, Al, Mo, Ba etc.

(E) Variable Microconstituents: found in certain plants in extremely low concentrations, include Li, Ls, Ag, Be, Sr, Cd, Ge, Sn, Pb, As, Cr, Co, Ni, Al, Mo, Ba etc.

Thacher (1934) classified these elements on the basis of

general function they take part in, as follows:

(i) Elements taking part in energy transfer reactions, e.g., H, O, P.

(ii) Oxidation and reduction controllers, e.g., Mg, Fe, Co, Ni, Cu, Zn, Mn, Mo.

(iii) Storage elements e.g., C, N, S, P.

(iv) Structural elements, e.g., Ca, Mg, P, F, Si.

(v) Regulators of Osmotic concentration and electrolytic equilibrium, e.g., Na^+, K^+, Ca^{++}, Mg^{++}, Cl^-, PO_4 CO_3 Br, NO_3 etc.

(vi) Catalytic e.g., Fe, Cu, Mn, Zn, I, S, Mo, CO etc.

(vii) Enzyme activators e.g., Ca, Mg, K, CO, Zn, Cu, Mo.

Minerals play the following general roles in plants.

(1) Body Building Material: C, H, O are constituent of most organic compounds of cell body such as carbohydrates in cell wall protein in protoplasm fats etc.. S, P and N occur in proteins Ca^{++} and Mg^{++} are important constituents in cell walls and chloroplasts.

(2) Osmotic Ingredients: Mineral salts present in the cell sap are responsible for creating osmotic pressure.

(3) pH Effect: The reccgnizable mineral mineral salt present in in all sap effect the pH of the protoplasm.

(4) Permeability: Permeability of cytoplasmic membranes is influenced by presence of mineral ions near them and can

be increased or decreased depending upon the type of ion present.

(5) Toxic Effect: Elements like As, Hg, Cu, are toxic under certain conditions and damage the protoplasm.

(6) Catalytic Role: Cu, Zn, Fe, Mg, Ma act as catalytes in specific enzymatic reactions.

(7) BalancingFunction: Very often the presence of salt is beneficial in that it can balance the antagonistic effects between other salts.

There had been some controversy among scientists about the meaning or the form of essential elements, because it is often seen that a plant may absorb a certain element. But this element plays no essential role in the body-functions. Therefore, Arnon (1939) proposed the following criteria to determine the essentiality of an element.

a) The element is essential whose deficiency or absence causes certain abnormal features in the growth of the plant, presenting it to complete its life cycle.

b) The supply of that element alone can restore the normal condition of the plant.

c) That element alone is directly responsible for causing abnormal features to develop by its deficiency or absence.

The essentiality of an element depends on requirement by the plant.

(1) Macro Nutrients: These elements are required by the plant in a major amount, e.g., N, P, K, Ca, Mg, Na, Fe, C, H, O, S.

(2) Micro-nutrients (Trace Elements): These elements are used in very less amount but play a vital role in plant metabolism, e.g., Cu, Zn, B, Mn, Mo, Ca, Ni, Se.

Pod Development and Mineral Nutrition:

West Afri. Jour. Biol. App. Chem. (1967) chemical studies on pulses like V.unquiculata, groundnut, Lima bean, Pigeon pea and Soybean. In Soybean 'Ca' was 0.38 per cent while in others it was 0.05 to 0.10 per cent phosphorus was 0.40 per cent, while in others 0.31 to 0.54 per cent phosphorus ranged from 30 to 133 per cent in pigeon pea and 33 per cent in groundnut. According to Hocking and Pate (1928) mineral nutrition of fruiting plant i.e., Pisum sativum L. albus and L. angustifolias show specific change in mineral contents in leaves, pods, seed coat and embryo. He further stated that P, N and Zn increase rapidly in plant organ relative to dry matter accumulation; other elements more or less parallel with (K, Mn, Cu, Mg and Fe) or significantly behind Ca and Na. Dry matter increases N, P and K is lost from the leaf, pod and seed coat during senescence 20-60 per cent. Khokar and Warsi (1987) studied nutrient uptake in gram seeds and stated that the rate of nitrogen, phosphorus and potassium is increased with or without zinc. Application of K alone increases its uptake;

TABLE-2.14 : Mineral composition of pods at different developmental stages in C.cajan seed and pod cover.

Days after months	Seed (mg 100^{-1} g dry weight)					Seed (ppm dry weight)				
	Na	K	Ca	P	Mg	Mn	Fe	Cu	Ni	Zn
10	2.6	20	35.6	0.45	24.0	0.04	41.20	2.40	1.80	0.52
15	5.0	60	73.6	0.40	36.20	0.16	43.20	5.20	3.40	0.82
20	4.4	56	67.8	0.35	43.20	0.40	45.60	4.60	3.80	0.84
25	3.4	50	65.6	0.33	52.60	0.30	41.20	4.20	3.40	0.86
30	3.0	40	55.6	0.30	45.60	0.26	37.80	3.60	2.60	0.96
35	1.4	38	48.8	0.28	39.80	0.24	34.00	2.40	2.40	0.76
40	1.0	30	44.8	0.29	35.60	0.18	29.40	3.00	2.20	0.54
	Pod cover Mg 100^{-1} dry weight					Pod cover (ppm dry weight)				
10	2.6	10	59.8	0.31	35.0	0.18	34.40	1.4	0.6	0.26
15	4.2	50	79.8	0.29	58.8	0.40	35.8	3.2	1.0	0.28
20	3.0	40	75.6	0.27	62.6	0.76	57.2	3.2	1.4	0.48
25	2.4	30	72.8	0.25	67.8	0.54	49.6	2.8	1.8	0.54
30	1.8	28	71.6	0.22	64.8	0.50	46.4	2.4	4.4	0.44
35	1.6	22	69.4	0.20	44.0	0.40	40.4	2.2	4.0	0.38
40	1.2	20	64.0	0.15	38.6	0.40	30.8	1.8	1.0	0.28

TABLE-2.15 Mineral composition of pods at different stages of development in P.tetragonolobus.

Days after anthesis	Seed gm 100^{-1} dry weight				Pod cover gm 100^{-1} g dry weight			
	Na	K	Ca	P	Na	K	Ca	P
10	0.05	0.21	1.61	0.31	0.60	1.07	4.08	0.52
15	-	-	-	-	-	-	-	-
20	0.02	0.20	0.84	0.51	0.56	0.98	3.60	0.46
25	-	-	-	-	-	-	-	-
30	0.02	0.14	1.48	0.48	0.42	0.85	1.80	0.24
35	-	-	-	-	-	-	-	-
40	0.15	0.06	0.92	0.50	0.44	0.68	1.68	0.20
50	0.02	0.05	0.94	0.51	0.50	0.68	1.62	0.16
60	0.02	0.07	0,84	0.54	0.52	0.86	1.68	0.16

Note: Values are expressed as of 100^{-1} g dry tissue.

TABLE- 2.16 Mineral contents (mg/100 g) in food legumes.

Food Legume	Na	Ca	P	Mg	Fe	Cu
Horse gram	37.3	105	310	172	11.9	5.5
Moth bean	11.5	120	320	225	9.6	1.1
Chickpea	29.5	114	387	168	6.2	2.3
Soybean	27.0	226	546	236	8.5	2.4
Winged bean	40.0	290	277	170	11.0	1.5
Black gram	39.8	154	385	185	9.1	0.72
Cowpea	23.2	77	414	230	5.9	0.75
Green gram	28.0	124	326	171	7.3	0.97
Lathyrus	37.7	120	317	92	6.3	0.77
Lentil	40.1	69	293	94	4.8	0.66
Peas	20.4	75	298	124	5.1	0.85
French bean	15.0	260	410	195	5.8	0.95
Pigeon pea	28.4	124	304	133	5.8	1.25
Lima bean (baby)	3.76	76	397	164	6.79	0.64
Lima bean (large)	19.00	57	440	183	8.28	0.84

but had no effect on or slightly inhibited M and P uptake. Robson (1988) showed in Lupine transport of potassium, calcium and manganese is from Pod cover to seed. Wallace (1990) stated plant responses to some trace elements like Sn, Cr, Ni, Pb, V, Li. Present discussion is to record organize distribution of each element and its presence in pod cover and seeds at different developmental stages of C.cajan and P.tetragonolobus.

Sodium:

The amount of sodium present on earth is more than any other monovalent cation. However, its microquantity is sufficient for plants. Brownwell and Wood (1957) have shown that an Australian salt bush growing in arid region, Atriplex vesicaria requires sodium for its growth. It accumulates large amounts of sodium and chloride ions in its leaves. Kartz and Mayers (1955) have shown that both sodium and potassium required for better growth in many members of Ceanophyceae. Allen and Arnon (1965) have found that Na is essential for growth of blue green algae.

Work of Joham (1955 and 1957) and Whitenberg and Joham (1964) showed that sodium can partially substitute calcium in maintaining carbohydrate translocation. They showed that calcium deficient plants lived longer and had normal carbohydrate distribution when sodium was added in the nutrient medium.

Na^+ accumulation in pod cover is more than seeds. The

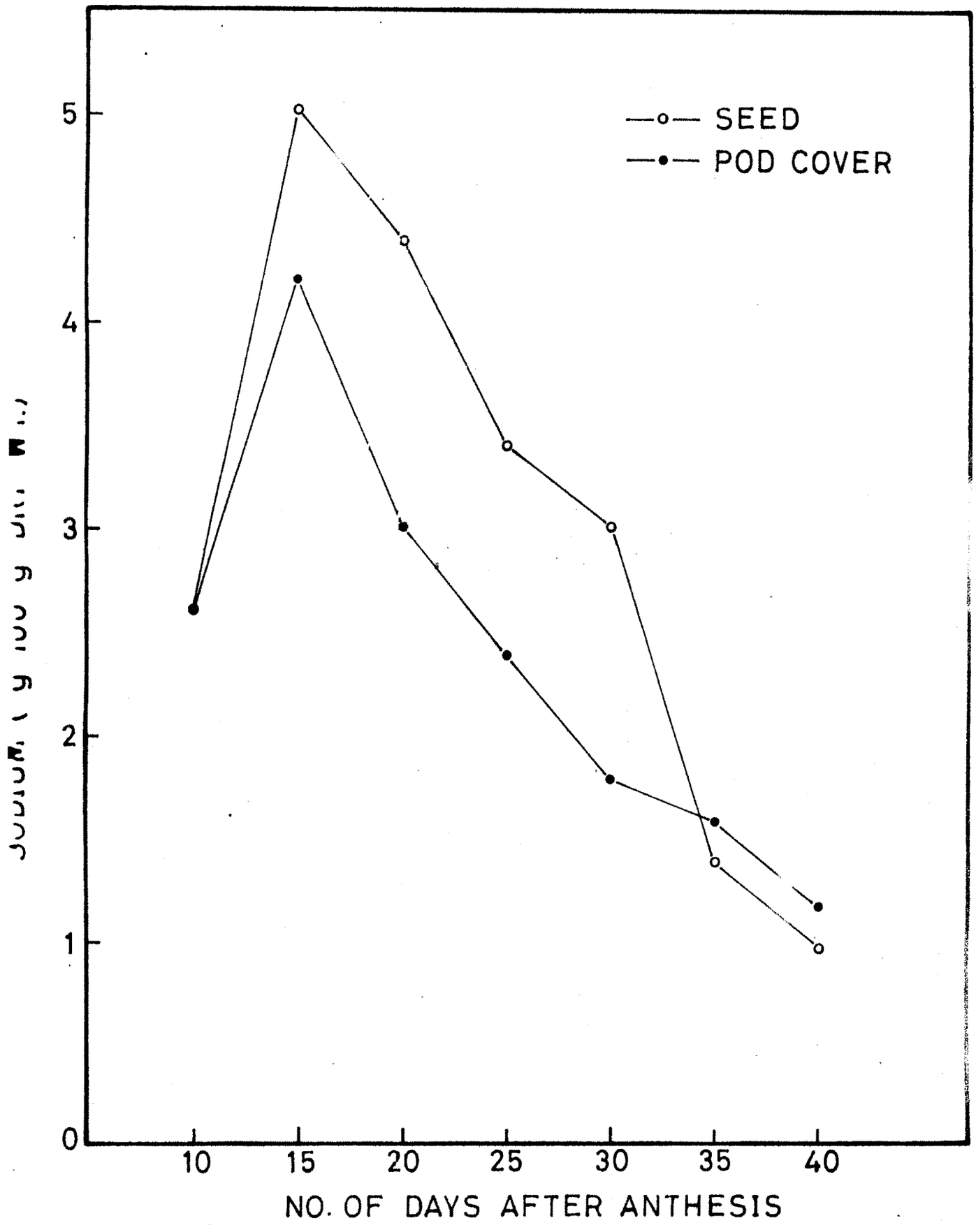

FIG. 2·18 SODIUM IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

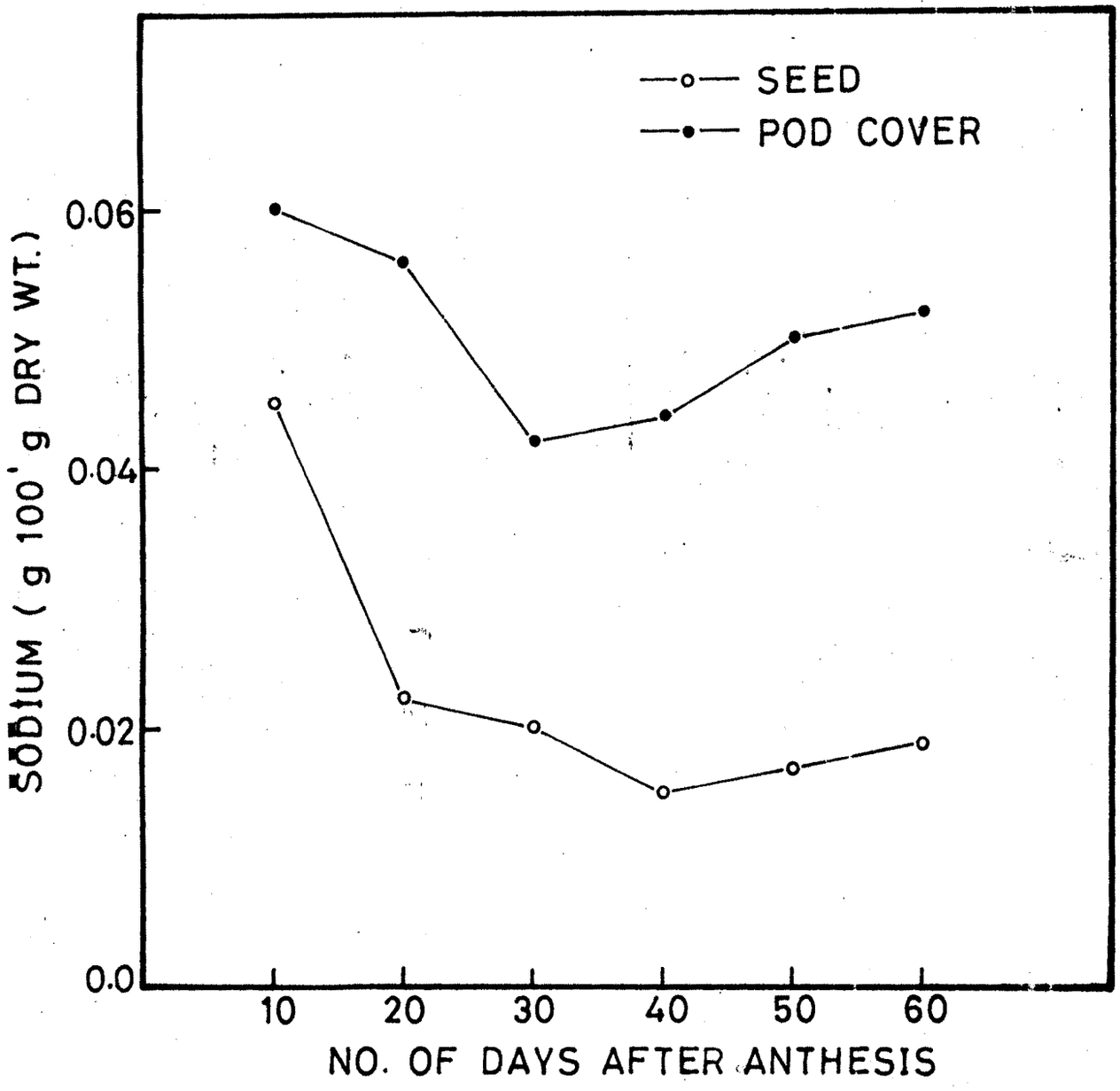

FIG. 2.28. SODIUM IN SEED AND POD COVER OF *P. tetragonolobus* DURING POD DEVELOPMENT.

accumulation of Na^+ in seed and pod cover during early stages of their development in C.cajan is less than that during maturation stage of pod cover and seed. Thus, it appears that more Na^+ is translocated to pod cover and seed during maturation.

The role of Na^+ in plants is uncertain. It is a micronutrient. Brownwell and Crossland (1972) found that plants having Cu, photosynthesis require Na^+ as a micronutrient. From the present results it appears that Na^+ has to play some role, may be a little in both C.cajan and P.tetragonolobus in pod development. In C. cajan accumulation of Na^+ was little more as compared to P.tetragonolobus.

Potassium:

There is evidence that among all essential mineral cation species K^+ is the only one which can be transported against an electrochemical gradient (active transport) into plant cells (Spanswick and Williams, 1964; Dunlop and Bowling, 1971; Ansari and Bowling, 1972). Potassium is relatively highly mobile. Its main transport direction is towards the meristematic tissues. Often K^+ from older plant parts is redistributed to younger ones. The bulk of K^+ is mainly taken up during the vegetative growth stage. On the other hand K^+ uptake and retention in plant cell are also competitively affected by H^+ Ca^{2+}, Mg^{2+} and Na^+ (Gartel, 1955). K^+ in the phloem sap is the main counter ion of malate which is transported from the shoot to the root and respired. In the respiration process the anion equivalents of malate are transferred to HCO_3 which are released from the roots into the nutrient medium.

It has been said that a plant is able to tolerate the harmful effects of disease, insects and cold conditions due to the presence of potassium in its cells. Potassium plays an important role in synthesis of starch, translocation and synthesis of sugars, growth of meristematic cells, development of young leaves and secondary roots and synthesis of proteins. Seeds of cereals are more swollen and their straw is stronger when adequte supply of potassium is obtained by these plants.

According to Broyer and Stout (1959) K^+ is linked with carbohydrate metabolism. Hartt (1929, 1934) observed that the depression in photosynthetic activity and translocation of sugars is caused by K^+ deficiency. Burr and Hartt (1960) reported that potassium deficiency reduces the rate of photosynthesis. Alten et al. (1938) found that under conditions of K^+ deficiency the photosynthesis activity of older leaves decreases rapidly, leading the leaf to senescence, withdrawal of K^+ and its translocation to newly developed parts.

Daniel (1956) reported that K^+ deficiency depressed the respiration in V.chlorella. Evans (1950) has shown that K^+ is necessary for the activation of Pyruvate kinase in Pisum sativum. Evans et al. 1950 ?

Baumeister and Schimidt (1962) have shown that K^+ plays a key role in protein synthesis. The average value of K^+ in the terrestrial plants ranged from 0.3 to 6 per cent (Ferry and Ward, 1959). Burridge et al. (1964) have noted in Theobroma cacao, K^+ is withdrawn from the leaves and supplied to the developing pods. Due to its

Burridge 1964 ?

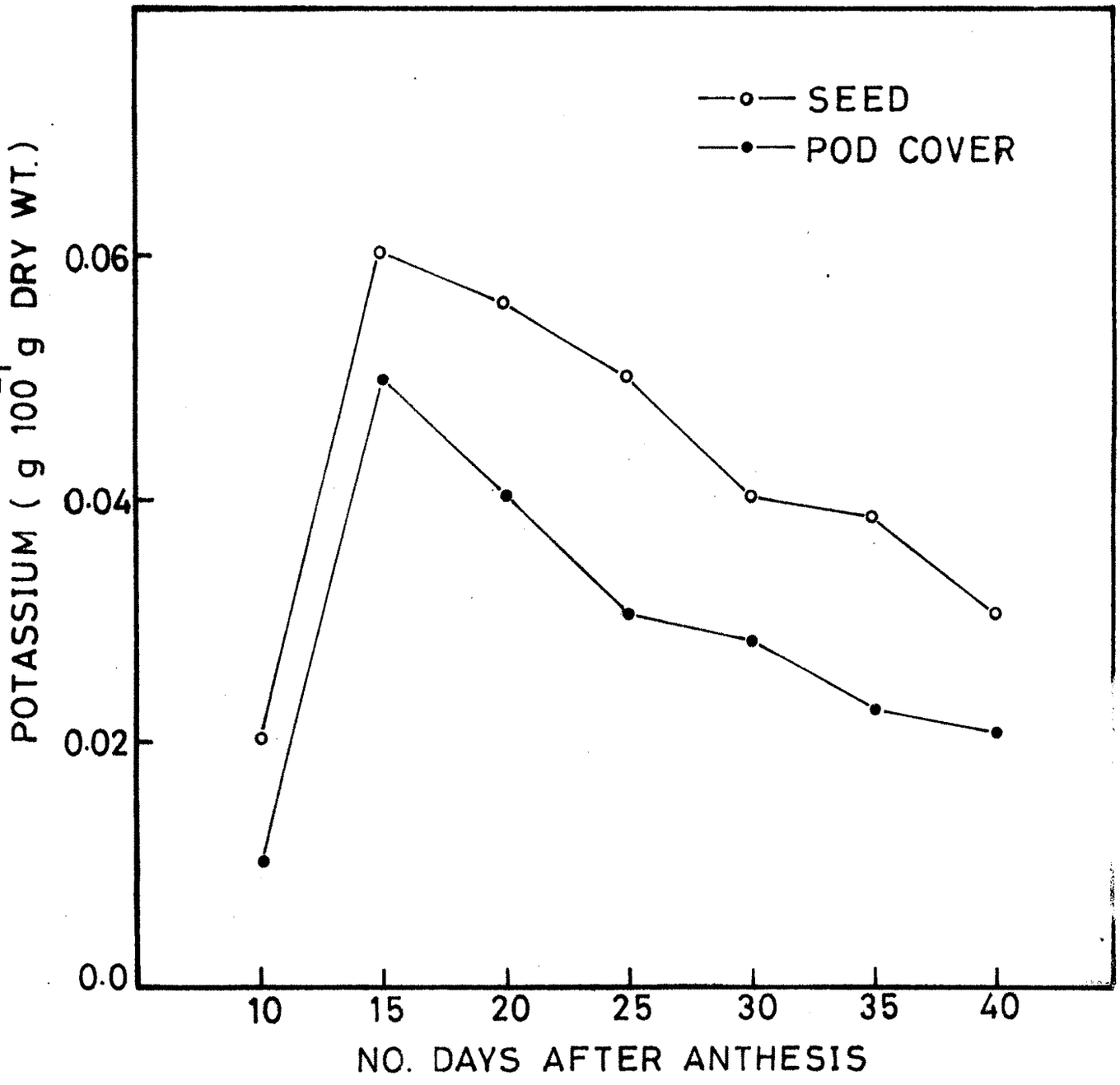

FIG. 2.19 POTASSIUM IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

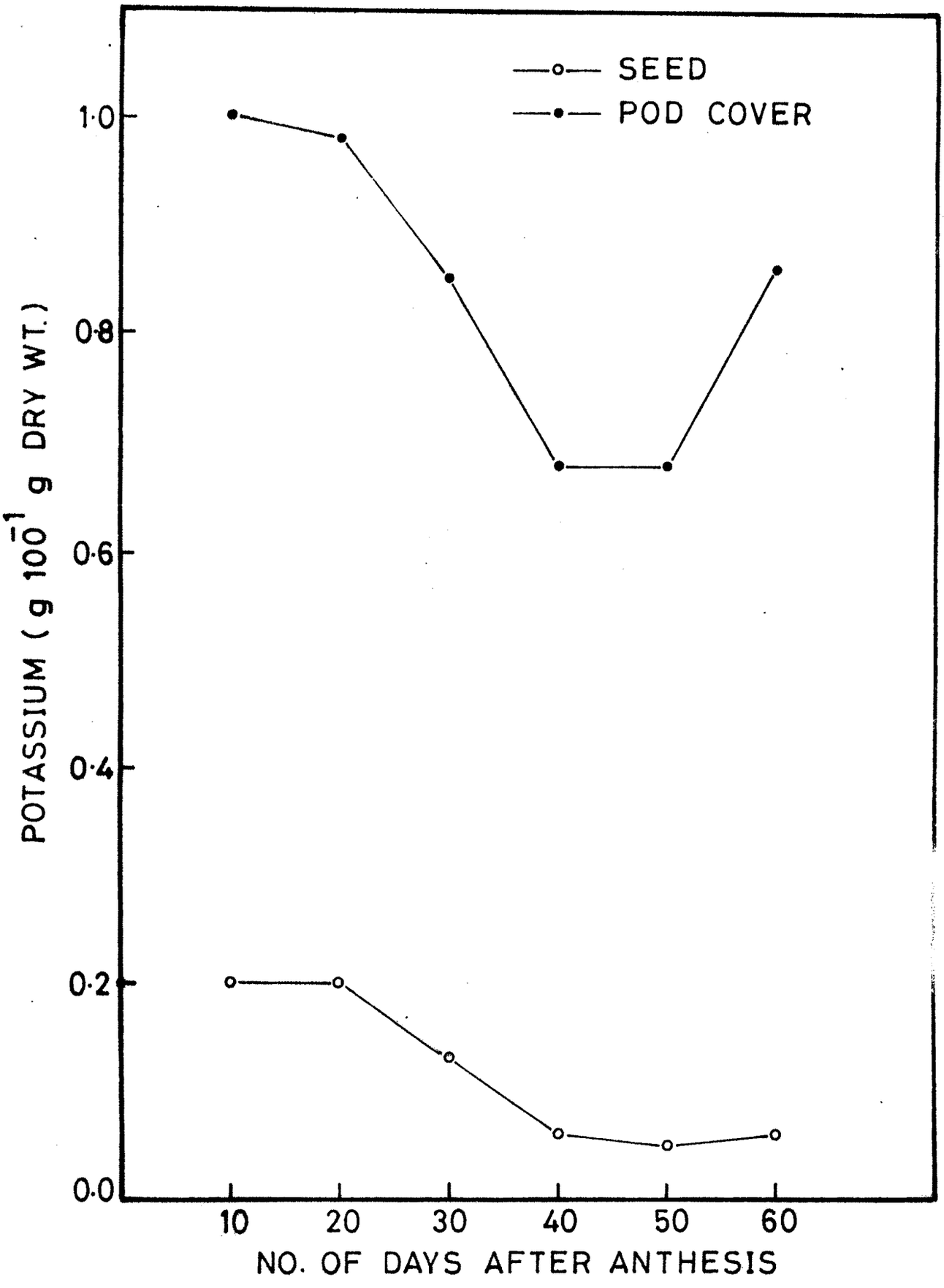

FIG. 2·29 POTASSIUM IN SEED AND POD COVER OF *P. tetragonolobus* DURING POD DEVELOPMENT.

mobility it is more in younger parts and in meristematic cells. Nason and McElory (1963) observed meristematic tissues are rich in K^+

Table-2.14, 2.15 records K^+ values in pod cover and seeds at different developmental stages in C.cajan and P.tetragonolobus. The highest values for K^+ have been recorded in the pod cover at juvenile stage as compared to seeds in both the plants. This means pod cover retains maximum amount of K^+. From the present results, therefore, it appears that the rate of translocation of this monovalent cation from pod cover to seed is low in P.tetragonolobus and C.cajan. However, this is not responsible for any deficiency symptoms particularly in seeds. Smith (1962) stated that mineral content of plant is dependent on age of plant or organ. Usually young plant and young plant parts have high concentration of N, P, K. According to Hocking and Pate (1978) some 60-90 per cent of K^+ is lost from the leaf pod and seed coat during senescence. K^+ absorbed is transferred to embryo for further growth and as seed matures the rate of absorption is low. Iskander (1987) studied the maturation and mineral contents of Soybean. He observed that the mineral content of mature, green mature and dry mature soybean differed and the concentration of K^+ at these 3 stages was 17.8, 21.3 and 19.7 mg 100^{-1} g.

Calcium:

Legumes are considerably richer in calcium than are most cereals. Soybean, horse-gram and a few other species contain over 200 mg of Ca 100^{-1} g dry weight, while groundnut has that well below the average. Ca^{2+} values vary widely within a species in

relation to factors such as variety, climate, cultural methods and mineral content of soil. Higher plant often contains Ca^{2+} in appreciable amounts and generally in the order of about 5-30 mg Ca 100^{-1} dry wt. The lower Ca^{2+} content is found in monocotyledons than in dicotyledons. Legumes and hurb have also shown to have a higher demand for Ca^{2+} than the grasses (Mangel and Kirkby, 1982). The reasons for this higher demand by dicotyledons for Ca^{2-1} is causally connected with the higher cation exchange capacity of the roots as well as in other plant parts of dicotyledons. In content to potassium and phosphate the transport of Ca (and Mg) is restricted to an area just behind the root tips. As roots age the endodermis becomes suberized. Larkson (1967) argues that as the radial movement of Ca^{2+} is prevented by the suberized endodermis; it is not transported effectively by the symplast. Ca uptake appears mainly to be a passive process. The same holds for the translocation of Ca^{2+} within the plants. Ca in the xylem sap is translocated in an upward direction with the transpiration stream. Thus to a large extent the intensity of transpiration controls the upward translocation rate of Ca^{2+} (Azaroff and Pitman, 1966).

Iljin (1938) has found that there is a positive correlation between Ca^{2+} content and organic acids of plants. Bharucha and Dabholkar (1958) have also shown that there is corresponding increase in organic acids with increase in Ca^{2+} content.

Ca^{2+} activates number of enzymes like arginine Kinase,

adenosine triphosphatase, adenyl kinase and succinyldehydrogenase (McElory and Nanson, 1954; Ellfolk, 1956) has shown that Ca^{2+} activates aspartase. Davidson and Lang (1958), Pirson (1955) and Einset and Clerk (1958) have shown that Ca^{2+} also activates phospholipases. Eystr (1964) has reported that it is essential element for nitrogen fixation in algae. Nodulation and successful symbiotic N_2 fixation requires relatively high concentration of calcium. The implement of nitrate reduction in Ca-deficient plants is apparently not attributable to a reduced carbohydrate level or decrease in translocation of reduced N_2 compounds (Banathetal, 1966).

The average values of Ca^{2+} in land plants vary between oil 0.1 to 3.5 g 100^{-1} g of dry weight of dry matter (Ferry and Ward, 1959). According to Epstein (1965) the optimum value for calcium in terrestrial plants is 0.5 per cent dry weight. Jones and Lunt (1967) stated that grasses and cereals have low Ca^{2+} content with values between 0.2 and 0.5 per cent while tobacco has 3.4 per cent Ca^{2+}

Jacobson et al. (1960, 1961), Epstein (1961), Rains et al (1964) and Waisel (1962) reported that Ca^{2+} was essential for integrity of the ion transported mechanism and was also involved in regulation of cell permeability for various ions. In the presence of calcium, potassium inhibits absorption of Na^{+}. It is an established fact that Na^{+} interferes with the Ca^{2+} uptake. Osmond (1966) working on Atriplex spp. has shown that relatively low calcium content of Atriplex leaves is due to the presence of high level of Na^{+} which depresses the

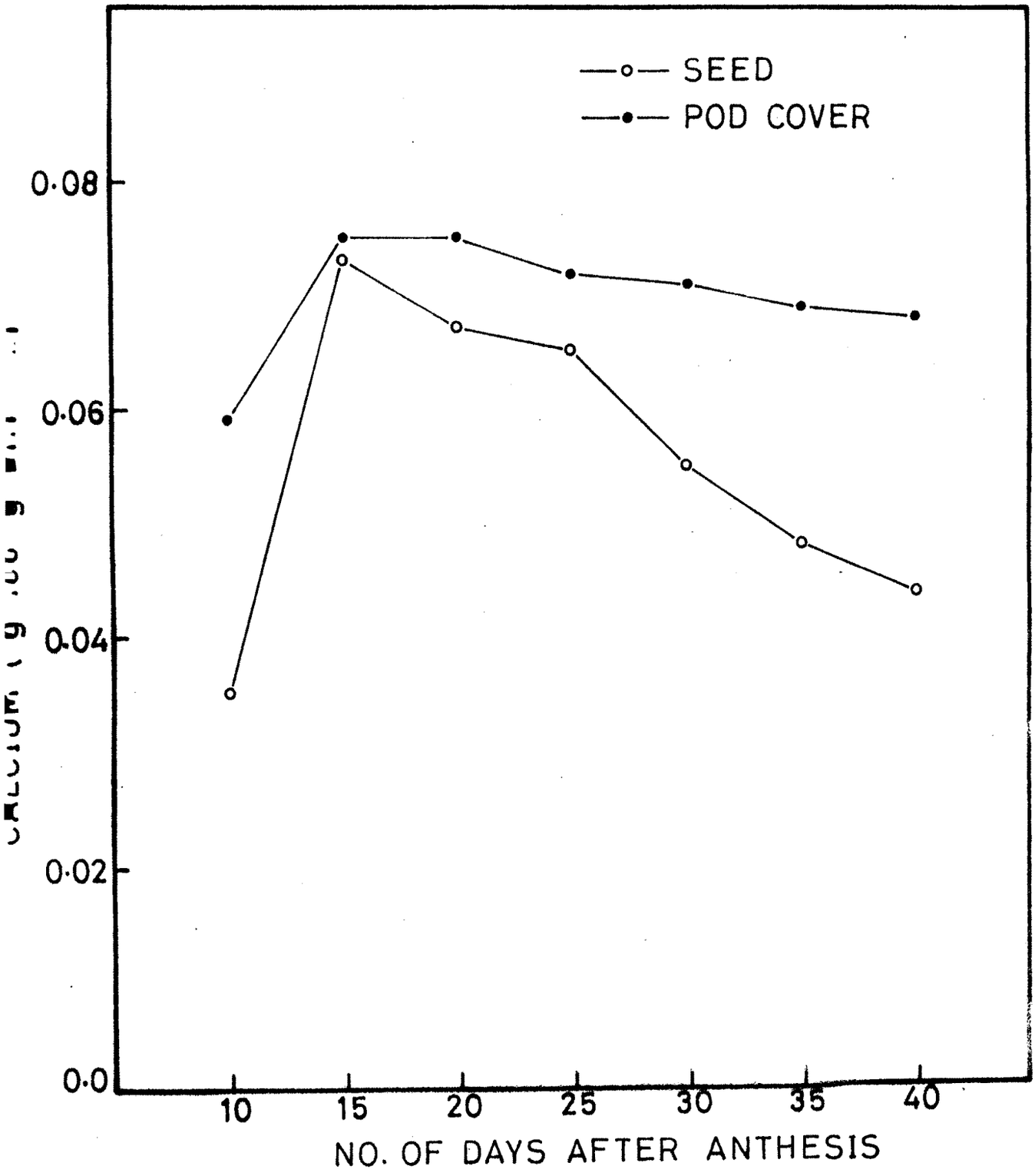

FIG. 2.20 CALCIUM IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

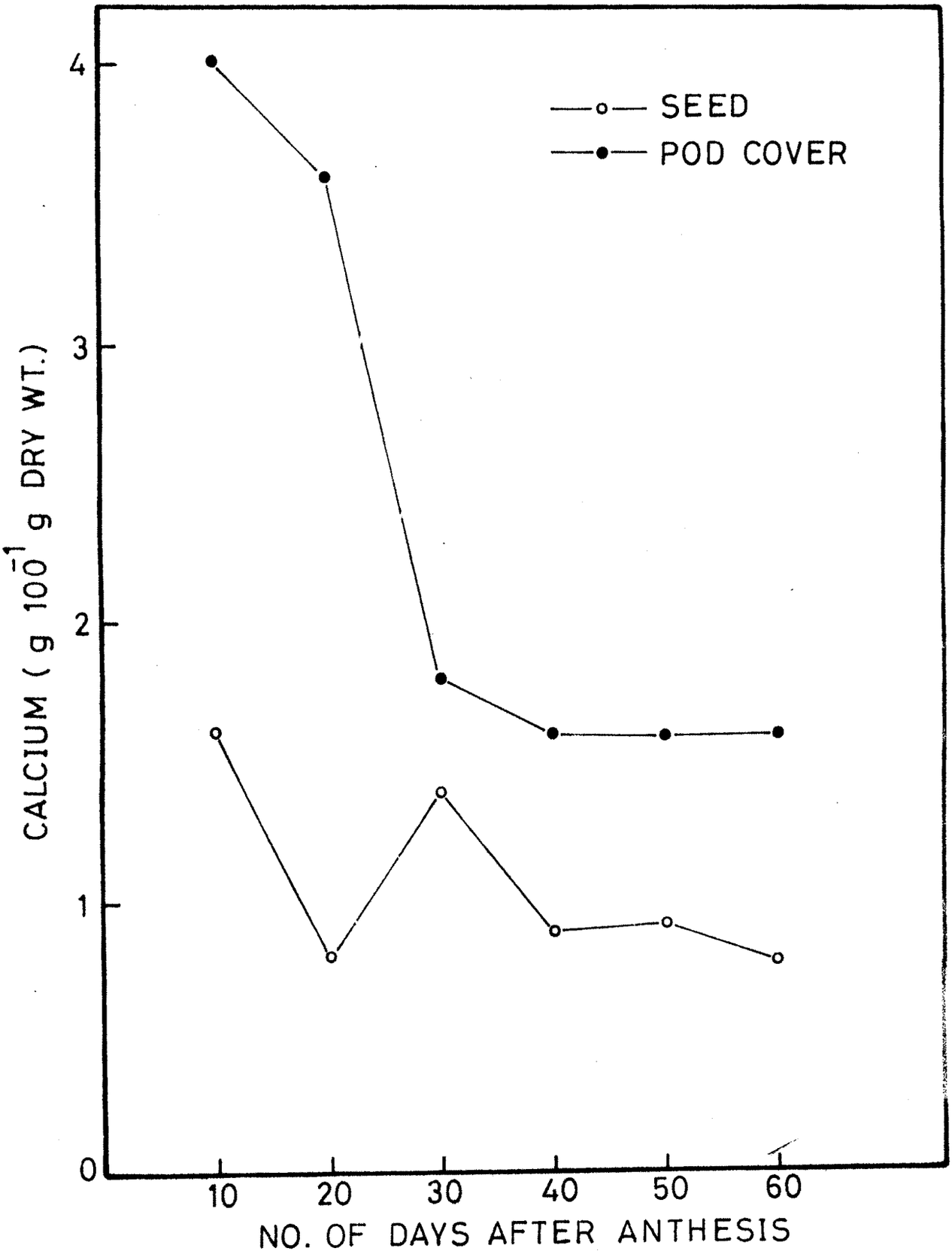

FIG. 2.30 CALCIUM IN SEED AND POD COVER OF *P. tetragonolobus* DURING POD DEVELOPMENT.

calcium uptake. Further he suggested that two processes of cation uptake were involved in a 'specific' process, possibly metabolically dependent, i.e., active transport operating at high concentrations.

Joshi et al (1976) have done considerablework on mineral nutrition of mangroves. They have reported that values of Ca^{2+} in Mangroves range from 0.42 to 1.25 per cent dry weight. These values are low as compared to the glycophytes. Epstein (1969) has proposed that Ca^{2+} is an integral part of the plasmalemma, governing its normal impermeability and transport of ions. They proposed that a deficiency of Ca leads to an impairment of the membrane structure, increasing cell permeability.

Calcium content of winged bean and C.cajan and other legumes is depicted in Table-2.14 and Fig. 2.20 . From the observation it is clear that the largest amount of Ca^{2+} was in the pod cover of C.cajan and P.tetragonolobus. (It is also evident that upto 30 days after anthesis the Ca content of both pod cover and seeds of C.cajan was increased slowly and was maintained. However, with development the Ca content of both these parts of pods in P.tetragonolobus was decreased continuously with a slight accumulation in pod cover after 30 days). As the pod became senescent and dry their calcium content decreased. This decline might be due to retranslocation of Ca^{2+} from the pod cover to seeds and to other plant parts also. Hocking and Pate (1977) observed that less than 20 per cent of sodium and calcium were lost during senescence of pods. Hocking (1980) further observed

Hocking & Pate 1977 ?

that Kennedia Prostrata pods lost about 9-15 per cent of calcium during senescence.

Iskander (1987) stated that during maturation stage in soybean the mineral content of the pods declined. The calcium content of immature, green mature and dry mature soybean was 3.44, 2.98 and 2.73 mg g^{-1} dry weight respectively.

Phosphorus P^{5+}:

Legumes contain a significant amount of P^{5+}. This is largely present as phytic acid, though its level can be reduced by different processes (Lolas and Markakis, 1975). Phosphorus plays an important role in cell metabolism concerned with formation of new cells, growth of roots, growth of leaves, emergence of ears of cereals, seed formation and maturation of fruits and seeds etc.

Plant root can absorb phosphate in very few amount 'PO'_4 uptake is usually associated with higher metabolic activity Respiratory metabolism derives the active 'PO'_4 uptake process. Phosphate uptake rate varies from species to species. Rate of 'PO_4^-' uptake is pH dependent. It declines rapidly with increasing pH. PO_4 absorbed by plant cells rapidly becomes involved in metabolic process. PO_4 is readily mobile in the plant and can be translocated easily in an upward and downward direction.

The most important compound in which phosphate groups are linked by pyrophosphate bonds is adenosine triphosphate (ATP).

The energy absorbed during photosynthesis or released during respiration or anaerobic carbohydrate breakdown is utilized in the synthesis of the phosphate bonds in ATP. In this form the energy can be conveyed to various enderogenic processes such as active ion uptake and the synthesis of various organic compounds.

Immediately after pollination there is an increase in 'P^{5+}' transport towards the young developing seeds. Phosphorus in phytin of seeds is regarded as a phosphorous reserve. During seed germination phytin phosphorus is mobilized and converted into other phosphate forms needed in the metabolism of young plants. (Reddy et al., 1978).

Peterburgski (1968) reported that P^{5+} contents in Phaseolus vulgaris decreased at night, compared with their nutrition during the daytime or throughout the 24-hour period. Kumar Singh and Narwal (1987) studied the effect of different sources and utilization of 'P' in soybean. They found that sulphur application increased the 'P' concentration in the leaves, stems, pods and seeds at 110 days after sowing. 'Zn' depressed the 'P' concentration in all part of the plant. In soybean 'P' was 0.40 per cent dry weight while it was in others 0.31 to 0.54 per cent. Total 'P^{5+}' in pigeon pea and groundnut was 13 and 33 per cent. At 110 days P^{5+} concentration in the plant parts was in the order seeds leaves pods stems.

Laver and Blevins (1990) studied the dry matter accumulation and phosphate distribution in soybean and observed different levels

of phosphate level in soybean. The plants were established in hydrophonic culture in a greenhouse at 0.4, 0.20, 0.10 or 0.05 m phosphorus and sampled 4 times during reproductive growth. The proportion of total plant phosphorus retained by nodules in low 'P' plants (8.9 per cent) was greater than that retained by nodules of highs P plants (4.2 per cent). At 0.05 m M P, proportioned a greater proportion of their 'P' to seeds (74 per cent). Mehta and Khatri (1962) have observed that in pigeon pea movement of Ca, Mg are greater at all stages of growth in Cajanus cajan, seeds are richer in leaves than other organs and seeds are rich in N.P.K. than other tissue 'P' 91 b (4.08 Kg).

It is evident from Table-2.14 and Fig. 2.3 that the phosphorus content of winged bean pods is relatively higher than that in C.cajan pods; average value of phosphorus in winged bean seeds is higher than that in pod cover. Hocking and Pate (1978) studied mobilization of minerals to developing seeds of a number of legumes and observed that endosperm minerals are of only minor significance in embryo nutrition. Comparison of the mineral balance of plant parts of Lupinus spp. with that of stem xylem sap and fruit tip phloem sap support the view that leaves and pods are principal recipients of xylem-borne minerals and that export from these organs via phloem is major source of minerals to the seeds. Endosperms and embryo are often observed between species in mineral composition of plant organ and in the effectiveness of transfer of specific minerals to the seeds.

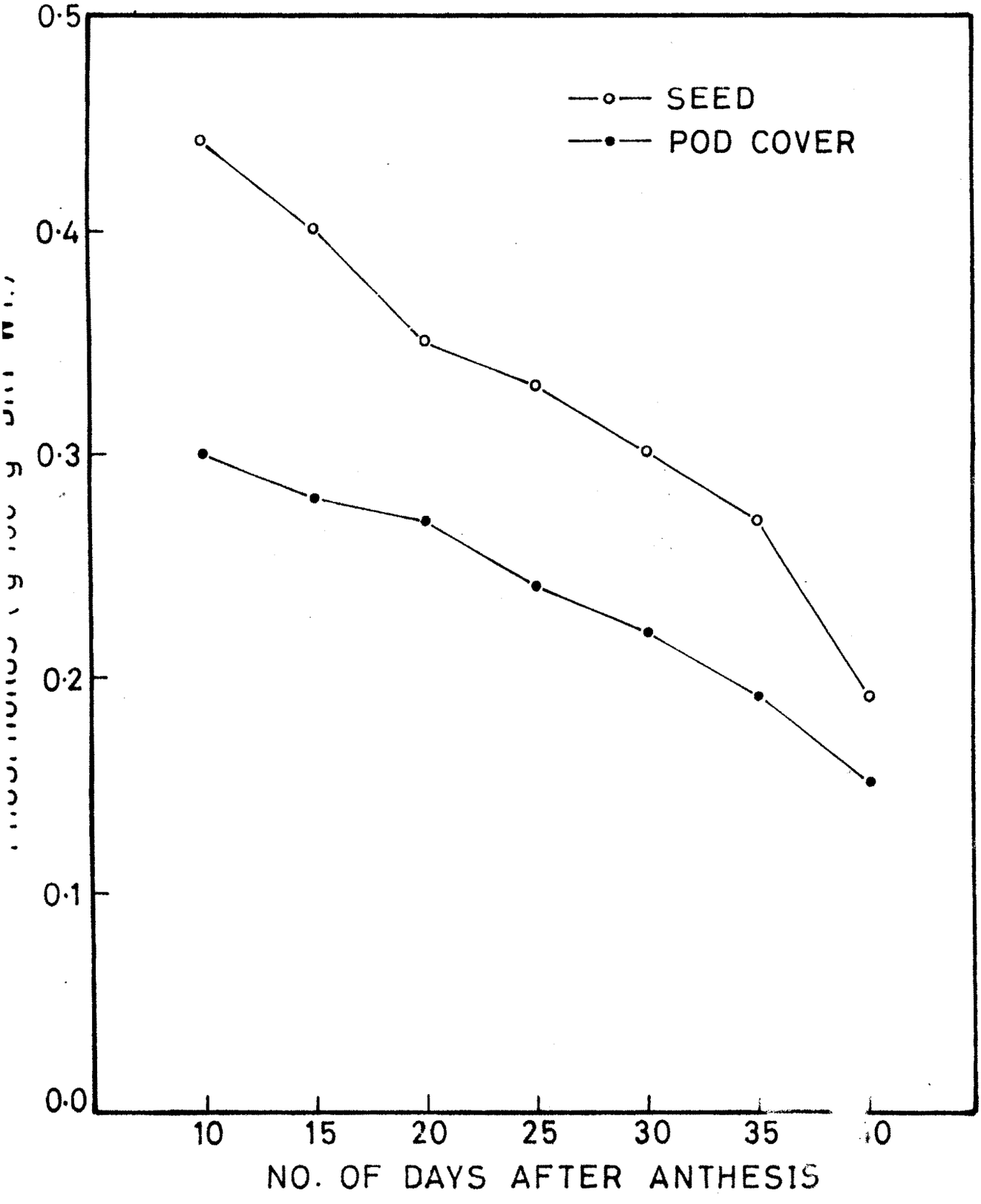

FIG. 2.21 PHOSPHORUS IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

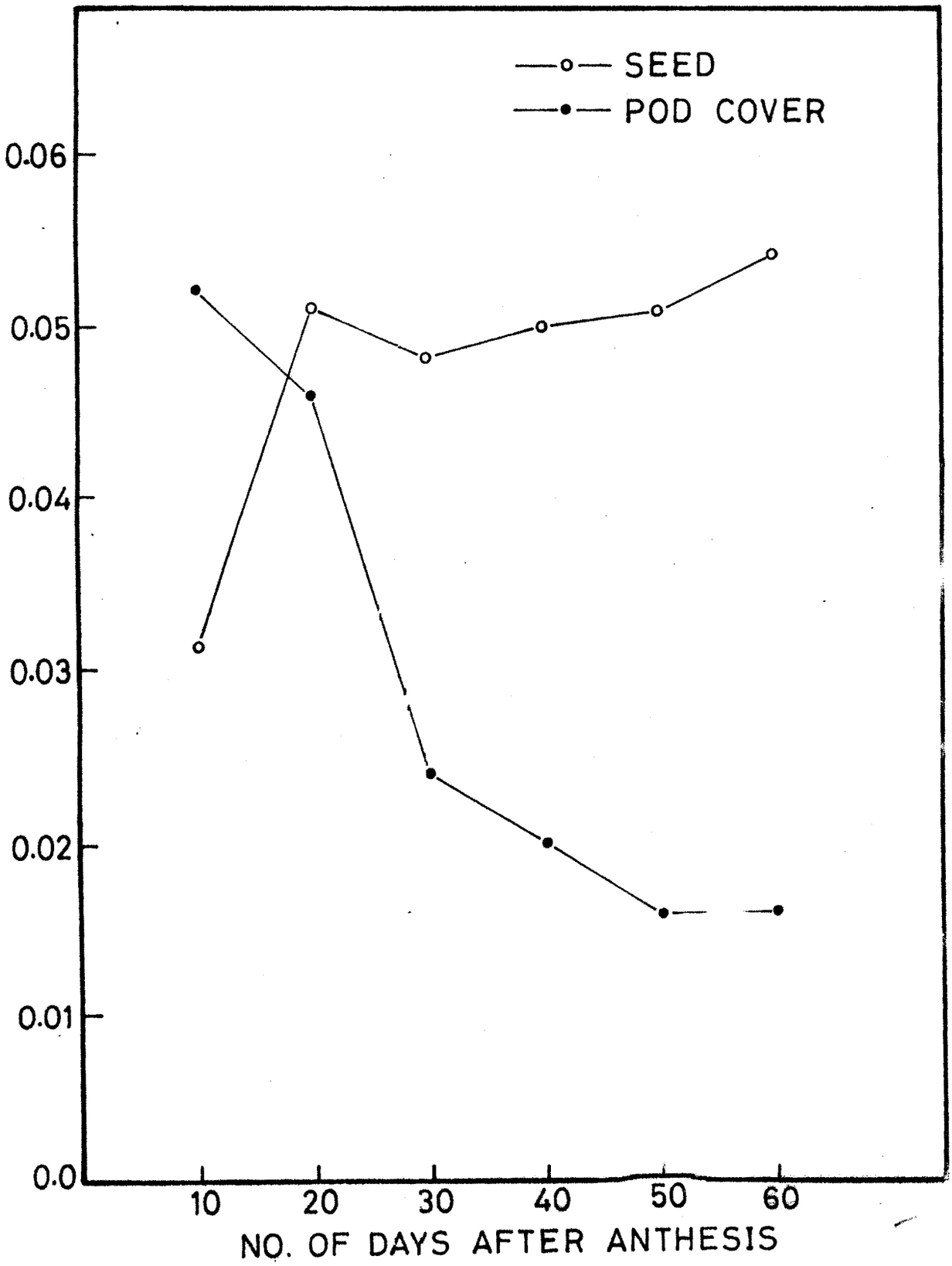

FIG. 2·31 PHOSPHORUS IN SEED AND POD COVER OF *P. tetragonolobus* DURING POD DEVELOPMENT.

In C.cajan phosphorus value was more in juvenile stage of development of pod anc it was observed that the 'P' value in seed was more than that in pod cover. This is because 'P' plays major role in metabolic activity, i.e., seed formation, maturation of fruit and seeds. The accumulation of 'P' was greater in the early stages of pod development as 'P' is needed for growth of seeds and as pod became dry the percentage of 'P' declined due to retranslocation of 'P' from pod. The work of Hocking and Pate (1978) also supports this statement as they found that there was some 60-90 per cent of the N, P, and K lost from the leaf, pods and seed coat during senescence.

Magnesium:

Mg^{2+} is one of the most important plant nutrients. It is the major constituent of all green plants. It is a part of chlorophyll a and b molecules and hence present in all autotrophic plants. Without chlorophyll formation, photosynthesis is prevented. The organization of proteins, RNA and DNA is dependent upon the activity of Magnesium. Magnesium takes part in the phosphorylation of many compounds by ATP, under many types of enzymes of kinase category. Magnesium is found both in the combined form as well as in the form of inorganic salts in the cell. Mazelis and Stumpf (1955) have found that Mg is involved along with adenine nucleotide and Krebs cycle intermediates, in the esterification of 'P' in ATP. Calvin (1954) has suggested that coenzymes such as ATP and ADP get linked to the enzyme surface

through Magnesium.

Arnon (1958) has shown that Mg plays a role in photosynthetic phosphorylation. Shibko and Pinchat (1967) found that in cell free particles from the bacteria Olcaligenes fecalis Mg is necessary for phosphorylation. Arnon et al (1955) have shown that CO_2 fixation in dark and light cannot take place if Mg^{++} is present in broken chloroplast preparation. Weissbach et al. (1956) have observed that RuDP-case which catalyses the reaction of CO_2 fixation, requires magnesium as a co-factor.

At flowering deficiency symptoms are associated with Mg^{++} concentrations in the leaf of 0.24 per cent or less (Webb et al., 1954). Normal plants contain about 0.37 per cent Mg. In the maturation phase Mg contents in field grown samples of soybeans showed a range if 0.53-0.79 per cent Mg (Austin, 1930). Magnesium levels may be maintained in the tops until the last three weeks before maturity in which period the concentration may fall to as low as half the original level from 0.8 to 0.4 per cent.

In germination the cotyledons serve as magnesium reserves; initially, magnesium content may in fact increase during the later development (McAlister and Krober, 1951).

The pod covers of C.cajan are relatively richer in Mg as compared with seeds. (Table 2.4, Fig. 2.22). Probably pod cover plays a major part in photosynthetic activities of developing pods.

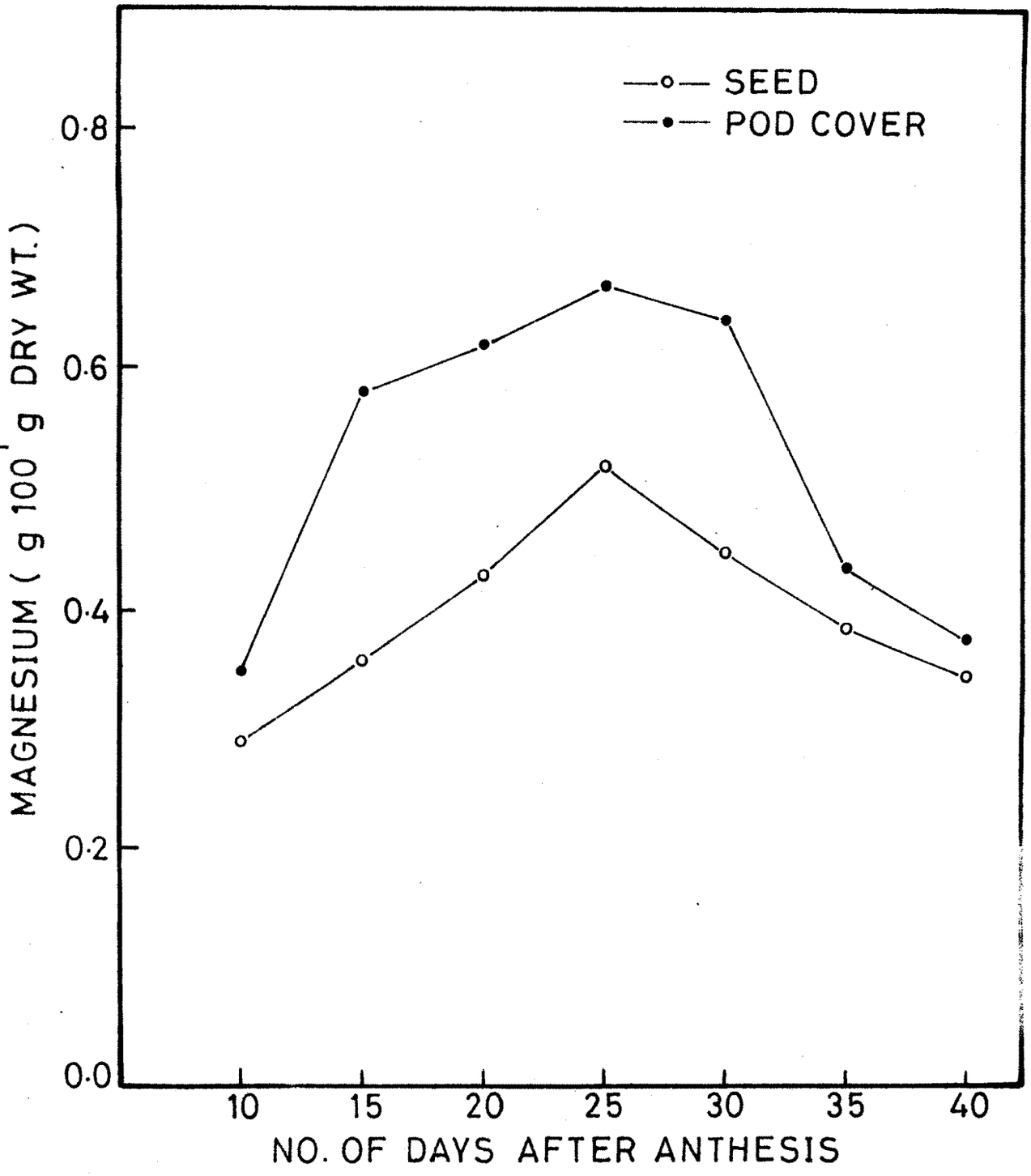

FIG. 2.22 MAGNESIUM IN SEED AND POD COVER OF *C. cajan* DURING POD DEVELOPMENT.

Values of Mg^{++} in C.cajan during early development of seeds were steady but were slightly increased during maturity period and declined during senescence. Bear et al. (1951) have shown that rapidly growing tissues and active mitotic cells have more magnesium than in older parts. As we know, during senescence the breakdown of chlorophyll and dissolution membrane may proceed to completion and Mg^{++} in the major constituent of chlorophyll.

Iron:

Iron uptake is inhibited in many plants by high pH. High concentrations of calcium and phosphate ions immobilize iron in the plant and may be responsible for a physiological iron deficiency. It is absorbed by the plants in the form of ferous ions. It has been reported that Fe content of some fruits and vegetables ranged from less than 4 to more than 10 ppm (10 mg/lit). Some plants contain more Fe than this but not enough to be considered a macro-nutrient.

Its main role in the formation of iron prophyrin, a precursor of chlorophyll, has been well established although chlorophyll does not contain iron; synthesis of chlorophyll is dependent when Fe remains. Thus iron is essential to the normal development of chlorophyll and when deficient the symptom of espression in the plants of chlorosis.

Fe is believed to have a significant role in the terminal respiration of plants, which involves transfer of electrons to molecular

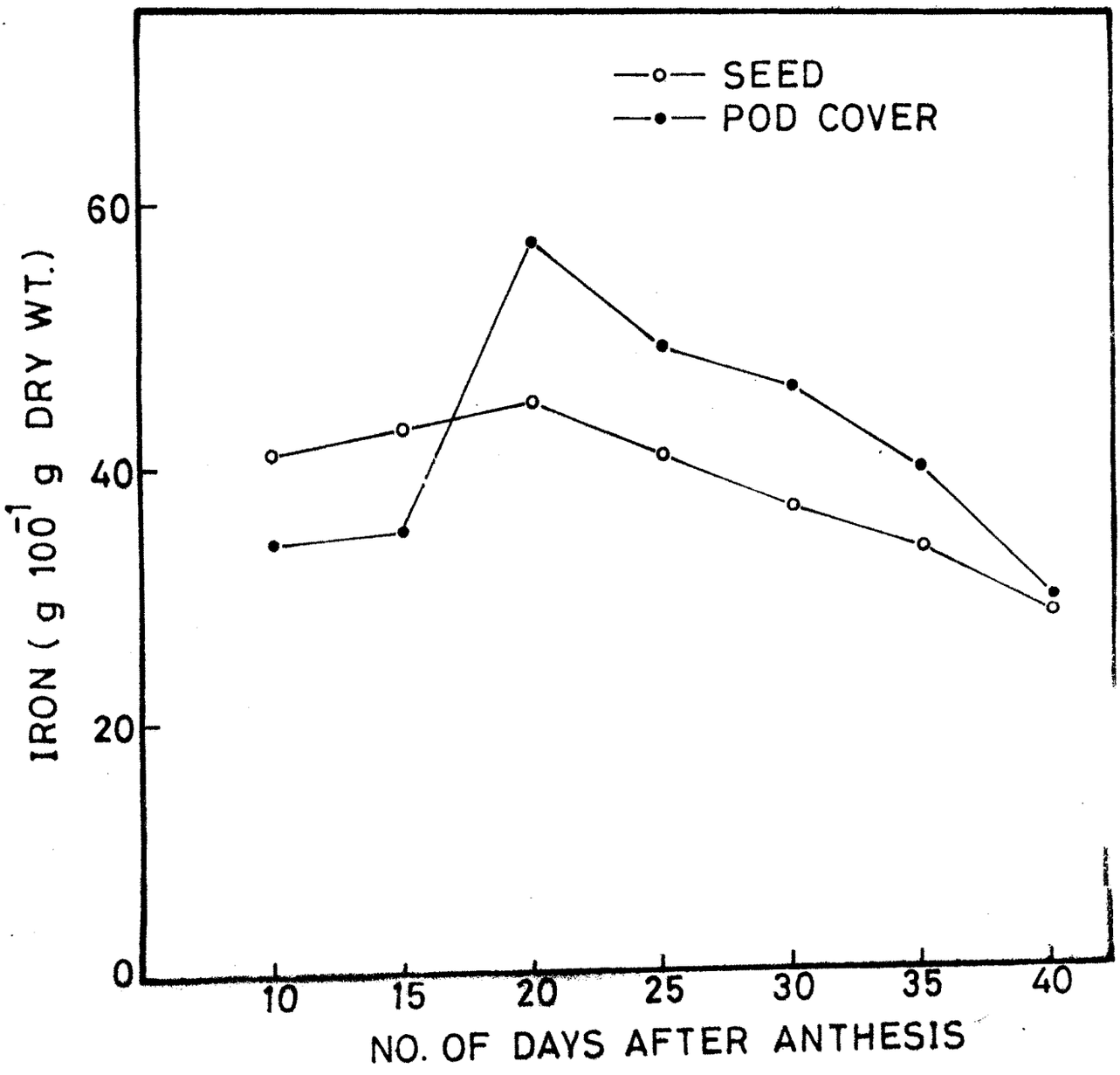

FIG. 2.24 IRON IN SEED AND POD COVER OF *C. cajan* DURING POD DEVELOPMENT.

oxygen. It is mediated exclusively by the iron porphyrin containing series of cytochromes like a, a3, b, b3, c and e etc. During the electron transport ion of cytochrome continuously undergoes oxidation and reduction between Ferric and Ferous forms. The energy required for salt ion uptake and accumulation is mediated by the home containing system. Its role in the nitrogen fixation is due to iron-porphyrin.

From Table-2.14 Fig. 2.24 it is evident that the values of Fe in the pod cover and seeds of C.cajan were highest during maturation stage of development (20 days after anthesis). It decreased towards senesconce or crying of pods. The Fe content of pod cover and seeds was more or less similar. Probably Fe may be translocated to the other plant parts during later stages of development of pods after maturation.

Manganese:

Manganese is essential in rather smaller or trace amounts in plants and its primary role seems to be in the growth and development of radicle and plemule. High amounts of Mn in plants are toxic producing irregular arrangement of tissues. It takes part in promoting the catalitic activity of certain enzymes which catalyse oxydation and reduction reactions. Mn also plays a role in photosynthesis during photolysis or oxidation of water to oxygen and hydrogen (Homonn, 1967). High quantity of Mn in soil prevents the absorption of Fe by plants and the plant shows Fe-deficiency symptoms. Mn effects the absorption of K and Ca by plants from soil.

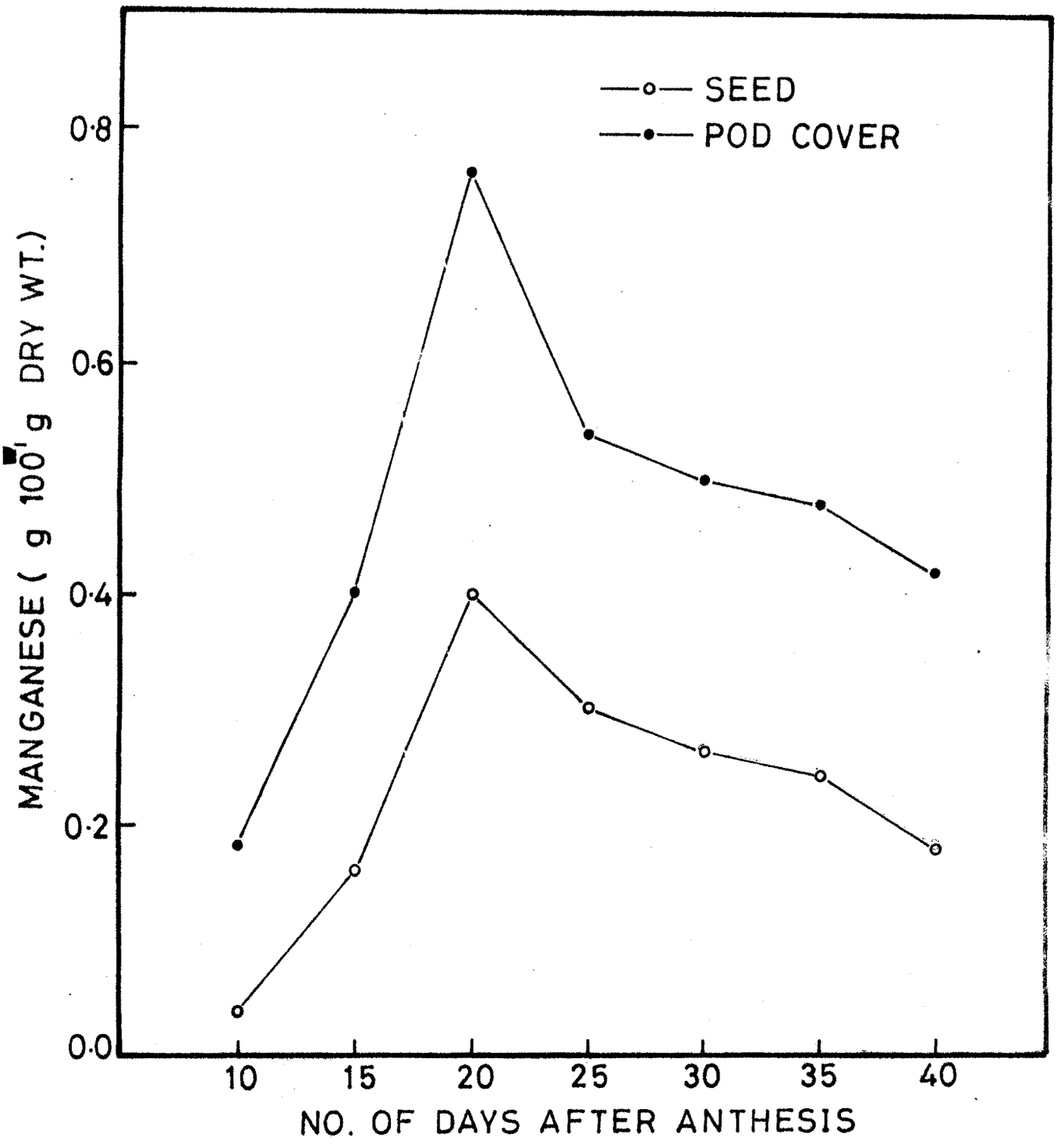

FIG. 2·23 MANGANESE IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

From Table 2.14 and Fig. 2.23 it is evident that Mn were more in the pod covers than in seeds in C.cajan. The Mn content of the pod during its juvenile stage of development (10 days after anthesis) was less but as seeds and pod cover grow the value of Mn also increased and then dropped down when pods were dried. According to Laszlo (1990) Cu and Mn contents of seed coats initially increased and then dropped accompanied by an increase of these minerals in the embryo. This finding suggests that cationic metals are not passively assimilated in conjunction with dry matter accumulation but rather are subject to ion-specific seed coat unloading transport and cotyledonary uptake process. Iskander (1987) reported that the Mn content in soybean seed at different developmental stages such as immature, green mature and dry mature stages was 28.6, 29.7 and 28.3 mg/g respectively.

Copper (Cu):

Copper is essential for the growth and reproduction of green plants in minute quantity but its presence in higher quantity is poisonous (Sommer, 1930), Lipman and Mackinnery (1931) found that barley plants were unable to develop seeds when Cu was omitted. Green (1938) reported that plant tissues on an average contain from 2 to 4 ppm of Cu depending on species, soil and amount of fertilizer used.

From physiological point of view 'Cu' is a constituent of some enzymes. It is claimed to be an active agent for some oxidases

and tyrosinase. Tyrosinase is helpful in chlorophyll formation. Patnaik (1955) has found that when no 'Cu' was given in culture solution plants were yellowish green in colour and there was a marked retardation of chlorophyll synthesis. Cu is also an essential component of a number of different plant enzymes, such as polyphenol oxidase, monopolyphenol-oxidase, lactase, ascorbic and oxidase and cytochrome oxidase (Nansonand McElory, 1963).

Singh (1966) has shown that nitrogen content of plant leaves bears a direct relation to the role of copper application. Copper accumulation is directly related to the copper supply of the soil while independent of N_2 supply. Neish (1939) has suggested that Cu functions in photosynthesis. Absence of 'Cu' reduces photosynthesis and assimilation of CO_2 (Loustalot et al., 1945; Brenchly, 1914). Lal and Subba Rao (1953) have discussed the role of Cu in crop production. 'Cu' concentration in plants varies from plant part to part. Govindrajan (1954) reported that the yield increased by 15 to 20 per cent by application of five pounds of copper sulphate per acre.

The deficiency of Cu causes leaf tip chlorosis, interveinal chlorosis, rosetting, curving of camina and withering and drying. The plants exhibit poor tillering and growth and detopped. These studies indicate that 'Cu' plays important role in plant growth and apparency development. Hallworth et al. (1960) suggest that there is a specific requirement for Cu in symbiotic nitrogen fixation where

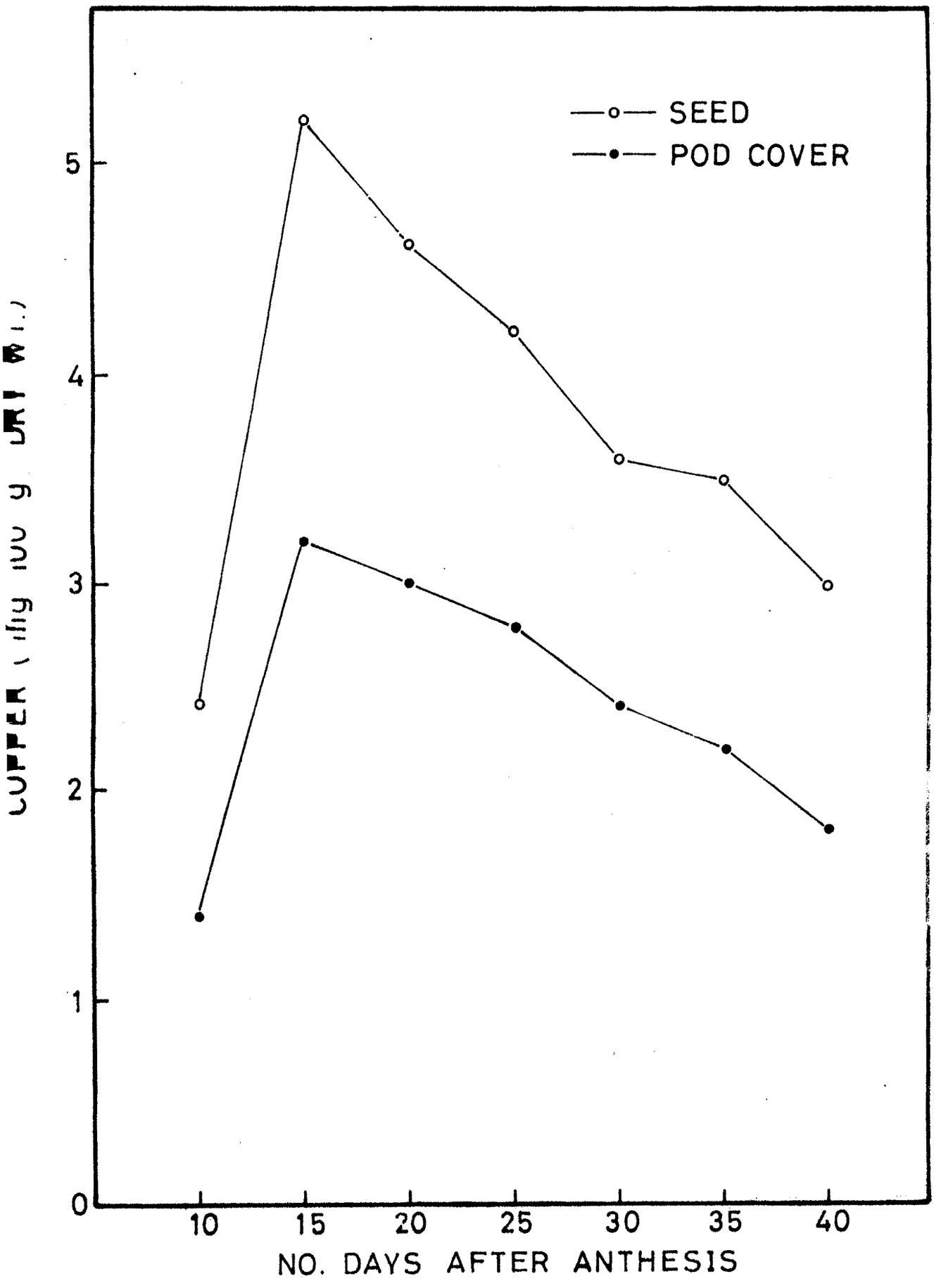

FIG. 2.25 COPPER IN SEED AND POD COVER OF *C. cajan* DURING POD DEVELOPMENT.

Cu is involved in leghaemoglobin synthesis.

From Table- 2.14 and Fig. 2.25 it can be seen that green mature seeds of C.cajan contained maximum amount of Cu. This high amount of Cu probably adds to the activity of photosynthetic electron transfer in this plant. The Cu content of both pod cover as well as seeds was decreased during later stages of development. The decrease was more rapid in the pod cover than in the seeds. Probably little amount of Cu may be transferred to seeds during later stages of development.

Zinc (Zn):

Zinc is one of the important micronutrients in plants. It is believed that Zn is important for the activity of many dehydrogenases which take part in respiratory metabolism. Zinc also plays a role in protein synthesis because, without it aminoacids accumulate as they are not condensed to form proteins. Zn plays a role in the biosynthesis of chlorophyll precursors and in plants synthesis as opposed to Mn, Cu and Fe.

Lipman et al. (1926) and Sommer (1928) established that Zn is absolutely essential for the growth and development of green plants. Neish (1939) for the first time demonstrated its presence in protoplasm and chlorcplast of wild red clover Infolium pratense.

Moghe (1965) reported that zinc content of cestain crops

grown in India is as follows: Vegetables 28.2 ppm, pulses 34.5 ppm, cereals 27.8 ppm, cotton and sugarcane 36 ppm and grasses 18.5 ppm. Zinc catalysed the process of oxidation in plant cells and is vital for the transformation of carbohydrates.

Thatcher (1934) reported that zinc and copper act like a pair of co-ordinated catalysts in oxidation-reduction phenomenon and care particularly concerned in reactions involving the transfer of hydrogen.

Zn forms a part of dehydroeptidase and glycoglycine dipeptidase enzymes functioning in protein metabolism (Nanson and McElory, 1963).

The most common Zn deficiency symptom is yellowing or chlorosis of leaves usually in fruit and nut plants. In several cases the plants become stunted and may die prematurely. It also shows adverse effects on seed production in beans and peas. Riceman and Jones (1958) found that seed and flower production was raised to normal level. Zinc improves the nodulation and nitrogen fixation by enhanced root growth (Malcuwar et al., 1982) and by activation of several enzyme systems and auxius (Smith, 1982). Zinc in soil application supported higher pod yield of groundnut which may be due to better nodulation and nitrogen fixation as also observed by Shrinivasan and Velu (1982).

Seeds of C.cajan are rich in zinc while its concentration

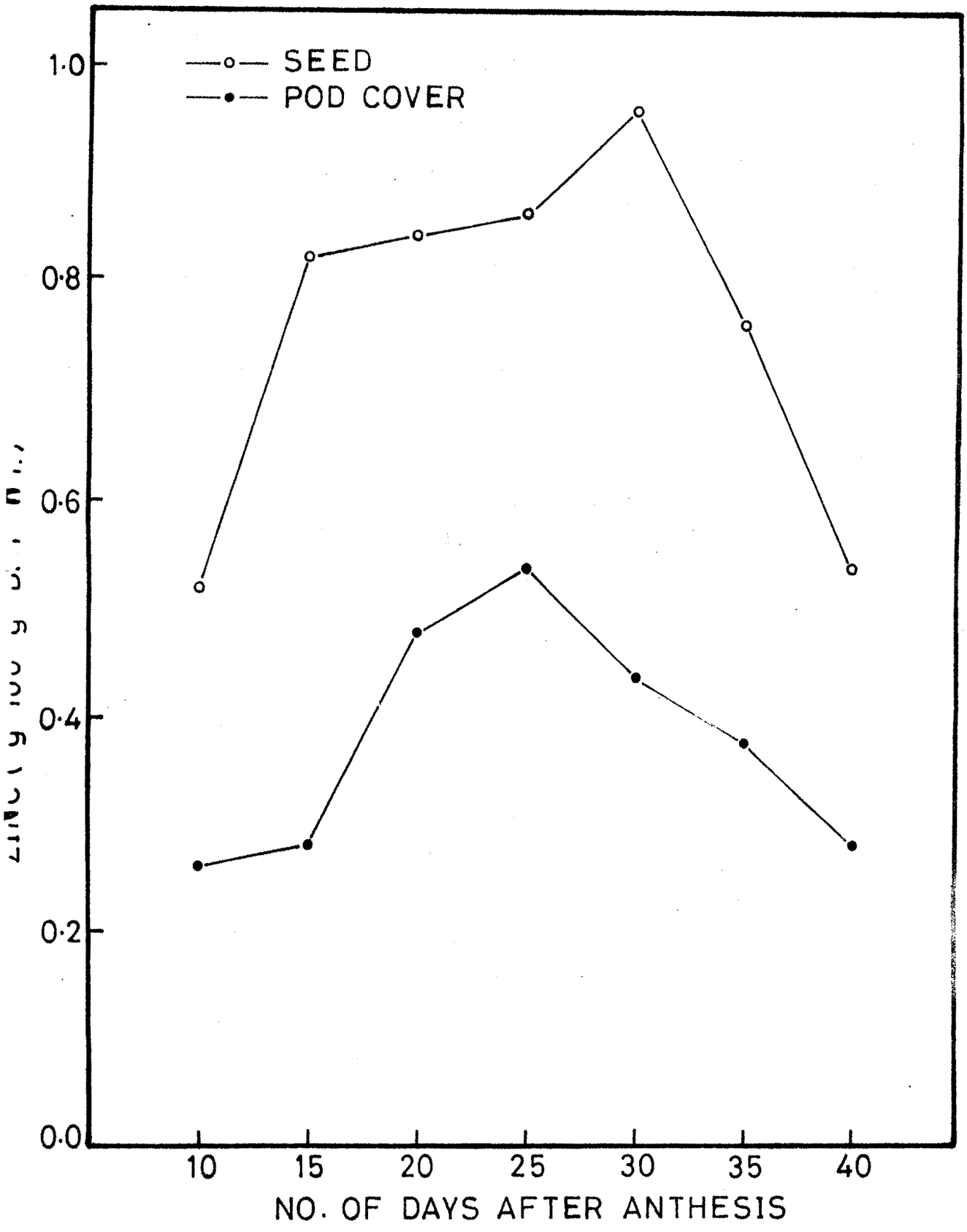

FIG. 2·27 ZINC IN SEED AND POD COVER OF C. cajan DURING POD DEVELOPMENT.

in pod covers is relatively less (Table-2.14 and Fig. 2.27). It was found that Zn was accumulated to a considerable extent in seeds upto 30-35 days stage. Similar trend was also shown by pod covers upto 25 days. The Zn content of pod cover was rapidly decreased during the later stages of pod development while the extent of decrease of Zn in seeds was not so significant. It appears that some amount of Zn was translocated to the developing seeds of this plant.

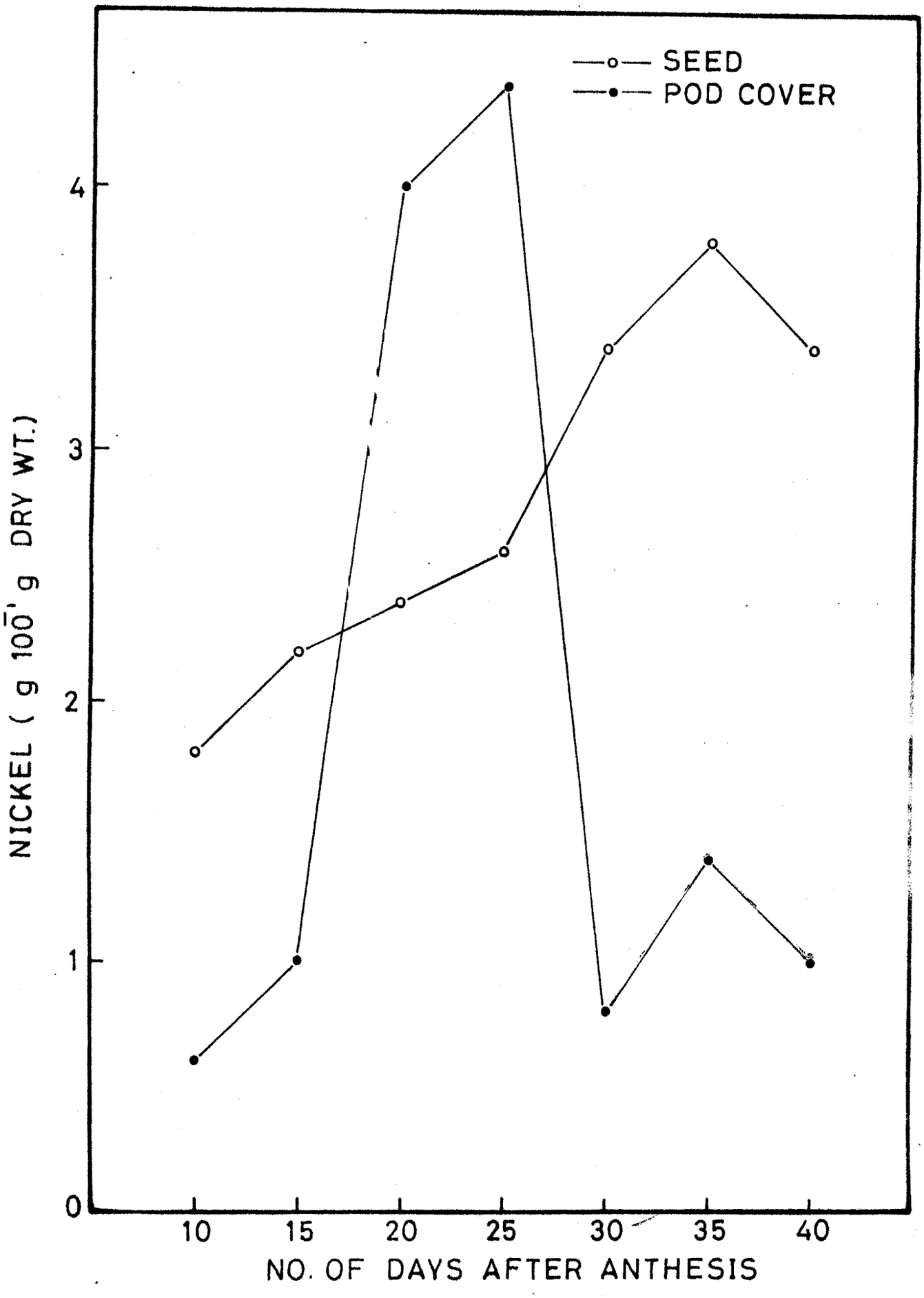

FIG. 2·26 NICKEL IN SEED AND POD COVER OF *C. cajan* DURING POD DEVELOPMENT.