Indian J. Plant Physiol., Vol. XXXV, No. 2, pp. 191-194 (June, 1992)

SHORT COMMUNICATION

SOME BIOCHEMICAL CHANGES IN DEVELOPING SEEDS OF BAMBARRA GROUNDNUT (*VOANDZEIA SUBTERRANEA* THOUARS)

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Received on 6 Sept., 1991

Fresh weight, dry matter, total phosphorus, soluble N, protein N, starch and sugar contents were analysed at 10, 20, 30, 40 and 50 days of developing seeds of bambarra groundnut (*Voandzeia subterranea* Thouars). The period between 30 and 40 days of seed development was found to be the most active phase. Comparison of data on growth with biochemical changes shows that the increase in starch and protein N contents was correlated with dry matter while sugar, soluble N and phosphorus contents were related to the changes in seed water content.

Bombarra groundnut is an indigenous African leguminous crop. Like peanut (*Arachis hypogea* L.), it produces geotropic fruits. Chemical composition of bambarranut seeds has been worked out by several investigators (International Grain Legume Information Centre, 1978). Though the production and utilization of bambarranut seed is increasing in Africa due to its high carbohydrate (45-50%) and protein (18-20%) contents. Survey of literature indicates that work related to seed development is lacking. Hence, physiological studies on developing seeds in bombarra groundnut was undertaken.

Bombarranut seeds (cv. BA/2, cream coloured without 'eye') supplied by Agricultural Research Station, Ukiriguru, Tanzania were grown in the departmental garden. Flowering took 40-45 days from the date of sowing. After fertilization, the peduncles were labelled when they touched the soil surface. Pods were removed at 10, 20, 30, 40 and 50 days (mature) after labelling. Studies were made on entire seeds during development. Seeds were mature when the parenchymatous lining of the shell had disappeared and brown patches had appeared on the sides of the shell.

Total nitrogen was determined by the standard microkjeldahl method. Soluble

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N was extracted as per the method employed by Singh, *et al*, (1980). The seed material (150 mg) was homogenized with 15 ml 10% (W/v) trichloroacetic acid (TCA) and then shaken for 30 min. in a mechanical shaker. The material was centrifuged (12000 g for 15 min) and the residue washed twice with 10% TCA. The supernatent and the washings were pooled and made up to 25 ml. Aliquots were used for soluble N estimation. Protein N was calculated by subtracting the levels of soluble N from the total N. Starch content was determined as per AOAC method (AOAC, 1965) and soluble sugar content of the ethanol extract was determined by the anthrone colour reaction (Yemm and Willis, 1954) using glucose as the standard. Total phosphorus was extracted as per the method employed by Hall and Hodges (1966). The material was thoroughly homogenized in deionised water, aliquots of the homogenate wet washed in 10 N sulphuric acid, and final clearing was accomplished with hydrogen peroxide. The digested samples were diluted and boiled for 10 min. to break pyrophosphate bonds and then assayed for total-P according to the method of Fiske and Subbarow (1925).

Sigmoid growth curve was obtained when the fresh and dry weights of the developing bambarranuts were plotted against time. The rapid growing phase corresponded with time between 30 and 40 days of seed development (Fig. 1A). Thus, this period appears to be the most active stage for accumulation of reserve material. Water content in the seeds increased up to 40 days followed by a decline till maturity. As a result, slight increase in fresh weights was observed between 40 and 50 days although the dry matter continued to accumulate.

The decrease in water content of the seeds from 40 to 50 days might indicate utilisation of water in the synthetic processess as also of the withdrawal of excess water. As the bambarranut pods develop below the soil surface, there is less possibility for evaporation of seed water. Total P content increased up to 40 days of development followed by a decline till maturity (Fig. 1B) correlating with changes in water content. This loss of P from the seed cannot be explained by the evaporation of water from the seed. Instead, it suggests the migration of an aqueous solution. In developing pea seeds also, loss in P content was correlated with desiccation of seeds (Rauf, 1978).

Comparison of data on growth with changes in starch and soluble sugar content (Fig. 1C) show that the increase in starch content is correlated with dry weight while the sugar content, which showed a rapid rise up to 40 days of development followed by a decline, is closely related to the changes in seed water content. A fall in sugar content linked with a fall in seed moisture was also observed in the developing seeds of *Pisum sativum* (Bain and Mercer, 1966; Rauf, 1978) and *Cajanus cajan* (Singh, *et al*, (1980).

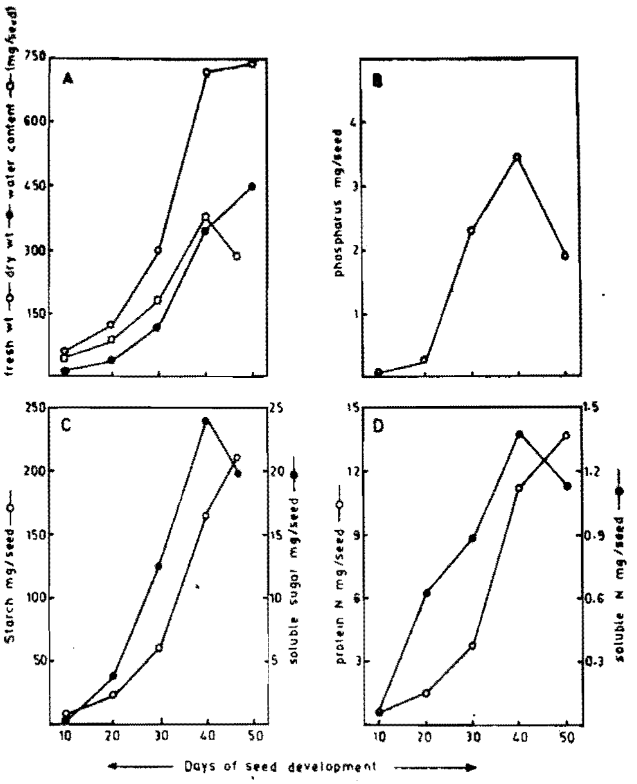
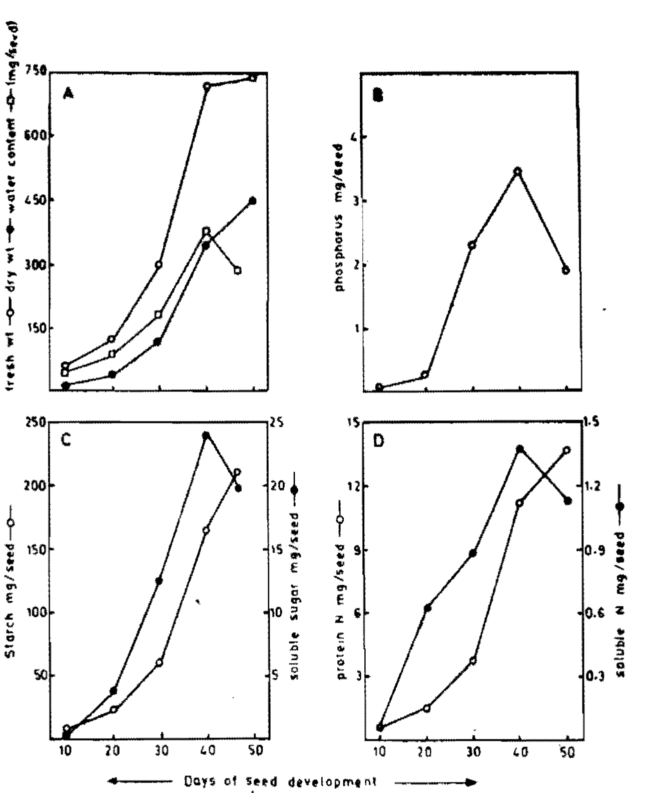


Fig. 1. Growth pattern and biochemical changes in developing seeds of bambarra nut.

In developing pea seeds (Bain and Mercer, 1966), correlation between sugar and water contents of the cotyledons was found closely associated with the differentiation and enlargement of the vacuolar system and expansion of cells. It is possible therefore, that the sugars entering the cell during this time are largely accumulated into the vacuoles. Such a situation, by controlling the osmotic pressure of the vacuoles, could provide the positive turgor necessary for the expansion of the cells (Bain and Mercer, 1966). A fall in sugar content accompanied by increase in starch in developing bambarra nut seeds (from 40 to 50 days-Fig. 1C) probably indicates the utilization of vacuolar sugars in starch synthesis, thereby decreasing the osmotic pressure of the vacuoles leading to the loss of water in the cells and consequently seed dehydration and cessation of expansion of cells in the seeds.

The protein nitrogen per seed increased throughout the development (Fig. 1D). Soluble N calculated as proportion of the protein N (calculated from Fig. 1D) increased rapidly from 10 days up to 20 days (1/10 to 1/2.4) and then progressively



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In developing pea seeds (Bain and Mercer, 1966), correlation between sugar and water contents of the cotyledons was found closely associated with the differentiation and enlargement of the vacuolar system and expansion of cells. It is possible therefore, that the sugars entering the cell during this time are largely accumulated into the vacuoles. Such a situation, by controlling the osmotic pressure of the vacuoles, could provide the positive turgor necessary for the expansion of the cells (Bain and Mercer, 1966). A fall in sugar content accompanied by increase in starch in developing bambarra nut seeds (from 40 to 50 days-Fig. 1C) probably indicates the utilization of vacuolar sugars in starch synthesis, thereby decreasing the osmotic pressure of the vacuoles leading to the loss of water in the cells and consequently seed dehydration and cessation of expansion of cells in the seeds.

The protein nitrogen per seed increased throughout the development (Fig. 1D). Soluble N calculated as proportion of the protein N (calculated from Fig. 1D) increased rapidly from 10 days up to 20 days (1/10 to 1/2.4) and then progressively decreased as the seeds developed indicating the possibilities of higher rate of utilization of amino acids for protein synthesis starting from 20 days of seed development. However, the continuous increase in soluble N per seed up to 40 days shows that the rate of synthesis of amino acids was higher than the utilization and vice versa from 40 to 50 days where a decline in soluble N occurred.

In mature seeds, the dry matter, starch and protein N were higher and the seed water content, soluble sugars and soluble N were lower than in immature seeds (at 40 days of development).

REFERENCES

AOAC (1965). Official Methods of Analysis, 10th ed., Association of Offi.ial Analytical Chemists. Washington, D.C.

Bain, J.M. and Mercer, F.V. (1966). Subcellular organization of the developing cotyledons of *Pisum sativum* L., *Aust. J. Plant Physiol.* **6** : 225-263.

Fiske, C.H. and Subbarow, Y. (1925). The colorimetric determination of Phosphorus. *J. Biol., Chem.*, **66** : 375-400.

Hall R.J., and Hodges T.K. (1966). Phosphorus metabolism of germinating oat seeds. *Plant Physiol.* **41** : 1459-1464.

International Grain Legume Information Centre (1978). Bambarra Groundnut *Voandzeia subterranea* Thouars) : Abstracts of World Literature 1900-78. International Institute of Tropical Agriculture, Ibadan, Nigeria.

Rauf, A. (1978). Comparative growth and biochemical studies on seed development. 1. Biochemical changes in developing seeds and pods of certain legumes. *The Plant Biochem. J.*, **5** : 157-166.

Singh, U., Jambunathan, R. and Narayanan, A. (1980). Biochemical changes in developing seeds of pigeon pea (*Cajanus cajan*). *Phytochem.* **19** : 1291-1295.

Yemm, E.W. and Willis A.J. (1954). The estimation of carbohydrates in plant extracts by anthrone. *Biochem. J.* **57** : 508-514.