

BRIEF COMMUNICATION

## The level of proteins and leakage of solutes in germinating fresh and stored seeds of *Cicer arietinum* L.

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### Abstract

One-year-old seeds of chickpea (*Cicer arietinum* L. cv. C-235) lost about 23 % germinability and leaked larger quantities of N, P, K, saccharides and proteins into the soaking medium in the first 48 h, as compared with fresh seeds. The protein content in stored seeds decreased more than in fresh seeds, as the soaking progressed.

*Additional key words:* chickpea, nitrogen, phosphorus, potassium, saccharides.

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The required quantity of water, for the initiation of the process of germination of the seeds, is imbibed from the soaking medium within 48 h. During this process certain organic and inorganic substances are released by the seeds into its surroundings. However, the degree of loss is determined by a number of factors including the seed age (McKersie and Stinson 1980) and species (Lott *et al.* 1991). MacKay (1972) and Abdul Baki (1980) showed that the seeds releasing larger quantities of the solutes exhibited poorer germinability. With the aim to find this, fresh and one year old seeds of chickpea were germinated under controlled conditions and the release of important substances was estimated.

The seeds of chickpea (*Cicer arietinum* L. cv. C-235) were procured from the *National Seed Corporation*, New Delhi. Twenty five uniform seeds, both fresh (FS) and stored for one year in the dark, at 50 - 55 % relative humidity and a temperature of 30 - 32 °C (SS), were surface sterilized with 0.1 % mercuric chloride followed with repeated washings with sterilized double distilled water (DDW). These seeds were separately incubated, in the dark, at 25 ± 2 °C in 50 cm<sup>3</sup> of DDW in sterilized Petri plates. Each sample was replicated five times. These seeds were sampled at 6, 12, 24, 36 and 48 h after soaking and per cent germination was noted. The soaking medium was decanted into 50 cm<sup>3</sup> flask and volume made upto the mark with DDW. The leachates in the soaking medium at each sampling were analysed for their nitrogen, phosphorus and saccharide contents - following the method of Lindner

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(1944), Fiske and Subba Row (1925) and Dubois *et al.* (1956), respectively. Potassium content was determined by flame photometer. The protein content, both in the leachates and the seeds was estimated according to Lowry *et al.* (1951).

Germinability (seed coat rupture and the emergence of radicle) of 22.8 % chickpea seeds was lost on their storage (Table 1). A similar loss of seed viability after dry storage has also been reported in other crops (*e.g.* Mackay 1972, Kalpana and Rao 1991). This loss of vigour and germinability is ascribed to a number of factors, among them most important one is the loss of water soluble solutes into the soaking medium (Madhusudanan and Johnson 1990). It is further strengthened by our studies (Table 2) where one year old (SS) chickpea seeds leaked significantly larger quantities of N, P, K, saccharides and proteins and for a longer duration than fresh seeds (FS). In general, the rate of leakage was inversely proportional to the degree of hydration of the seeds.

The protein level in FS and SS exhibited a descending trend with a maximum decline between 6 and 24 h of germination (Table 2). These results slightly deviate

Table 1. Germination percentage in fresh (FS) and one year old (SS) chickpea seeds.

| Imbibition [h] | FS    | SS    |
|----------------|-------|-------|
| 24             | 90.66 | 67.00 |
| 36             | 96.33 | 73.33 |
| 48             | 96.33 | 74.66 |

C.D. at 5 %: seed = 0.99; treatment = 1.58; interaction = 2.24

Table 2. Contents of proteins in seeds and in leachate and contents of saccharides, nitrogen, phosphorus and potassium in leachate

| Seeds | Imbibition [h] | Proteins seed [mg g <sup>-1</sup> (d.m.)] | leachate [%] | Saccharides [%] | Nitrogen [%] | Phosphorus [%] | Potassium [%] |
|-------|----------------|---|--------------|-----------------|--------------|----------------|---------------|
| FS    | 0              | 243.75                                    |              |                 |              |                |               |
|       | 6              | 234.37                                    | 1.13         | 1.93            | 0.181        | 0.031          | 0.188         |
|       | 12             | 193.75                                    | 1.27         | 2.22            | 0.203        | 0.034          | 0.224         |
|       | 24             | 153.12                                    | 1.48         | 2.38            | 0.237        | 0.036          | 0.268         |
|       | 36             | 142.18                                    | 1.52         | 2.68            | 0.243        | 0.037          | 0.292         |
|       | 48             | 135.93                                    | 1.54         | 2.75            | 0.247        | 0.037          | 0.296         |
| SS    | 0              | 240.62                                    |              |                 |              |                |               |
|       | 6              | 237.50                                    | 1.51         | 2.00            | 0.242        | 0.036          | 0.244         |
|       | 12             | 218.75                                    | 1.68         | 2.52            | 0.269        | 0.038          | 0.260         |
|       | 24             | 187.50                                    | 1.88         | 3.00            | 0.301        | 0.039          | 0.320         |
|       | 36             | 181.25                                    | 1.97         | 3.20            | 0.316        | 0.041          | 0.348         |
|       | 48             | 169.68                                    | 2.02         | 3.38            | 0.324        | 0.042          | 0.360         |

C.D. at 5 %: proteins (seed) - 22.43; protein (leachate) - 0.14; saccharides - 0.16; nitrogen - 0.02; phosphorus - 0.001; potassium - 0.040.

from previous observations (Nagar *et al.* 1993) where the maximum decrease in protein content in other legumes was noted from 24 to 48 h. The rate of loss of protein was higher in FS than SS, although, at 0 h the seed protein level in both these samples was similar. Therefore, during the peak hours of hydrolysis fresh seeds lost 20 % more proteins than stored seeds. At the same time, FS leaked 32 % less proteins into leachates than SS (Table 2). The fresh seeds, therefore, synthesized proteins more efficiently than the stored seeds where it not only favoured the breakdown of reserve proteins, but the synthesis of specific proteins helped in the re-establishment of the integrity of the semipermeable membranes (tonoplast and plasma membrane), which acts as a barrier against the loss of water soluble solutes (Simon and Raja Harun 1972, Powell and Mathews 1977).

Membrane damage during seed ageing is also proved by ultrastructural observations on maize (Berjak and Villiers 1972) and phospholipid analyses in cucumber (Koostra and Harrington 1969). Therefore, quicker repairability in FS checked the loss of nitrogen, phosphorus, potassium, saccharides and proteins into the leachates.

## References

- Abdul Baki, A.A.: Biochemical aspects of seed vigour. - Hort. Sci. 72: 370-373, 1980.
- Berjak, P., Villiers, T.A.: Ageing in plant embryos. III. Acceleration of senescence. - New Phytol. 71: 513-518, 1972.
- Dubois, M., Gilles, K.A., Hamilton, J.K., Robers, P.A., Smith, F.: Colorimetric method for determination of sugars related substances. - Ann. Chem. 28: 350-356, 1956.
- Fiske, C.H., Subba Row, L.: Colorimetric determination of phosphorus. - J. biol. Chem. 66: 375-400, 1925.
- Kalpana, R., Rao, K.V.M.: Physiological and biochemical changes in stored seeds of pigeonpea. - J. indian bot. Soc. 70: 67-70, 1991.
- Koostra, P.T., Harrington, J.F.: Biochemical effects of age on membranal lipids of *Cucumis sativus* L. seeds. - Proc. int. Seed Test. Assoc. 34: 329-340, 1969.
- Lindner, R.C.: Rapid analytical methods for some of the more common inorganic constituents of plant tissues. - Plant Physiol. 19: 76-89, 1944.
- Lott, J.N.A., Cavdek, V., Carson, J.: Leakage of K, Mg, Cl, Ca and Mn from imbibing seeds, grains and isolated seed parts. - Seed Sci. Res. 1: 229-233, 1991.
- Lowry, O.H., Rosebrough, N.J., Farr, A.L., Randall, R.J.: Protein measurement with Folin phenol reagent. - J. biol. Chem. 193: 265-275, 1951.
- Mackay, D.B.: The measurement of viability. - In: Roberts, E.H. (ed.): Viability of Seeds. Pp. 172-208. Chapman and Hall, London 1972.
- Madhusudanan, K.N., Johnson, K.M.: Solute leakage from winged bean (*Phosphocarpus tetragonolobus*) seeds. - Seed Sci. Technol. 18: 157-162, 1990.
- McKersie, B.D., Stinson, R.H.: Effect of dehydration on leakage and membrane structure in *Lotus corniculatus* L. seeds. - Plant Physiol. 66: 316-320, 1980.
- Nagar, R., Bhargava, M., Nagar, P.: Relationship between peroxidase activity, protein and phenolics at different levels of germinated legumes. - J. indian bot. Soc. 72: 63-65, 1993.
- Powell, A.A., Mathews, S.: Deteriorative changes in pea seeds (*Pisum sativum* L.) stored in humid and dry conditions. - J. exp. Bot. 28: 225-234, 1977.
- Simon, E.W., Raja Harun, R.M.: Leakage during seed imbibition - J. exp. Bot. 23: 1076-1085, 1972.