

## The Changes of $\alpha$ -galactosides during Germination and High Pressure Treatment of Chick-Pea Seeds

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**Abstract:** The  $\alpha$ -galactosides negatively affect of digestibility of grain legumes. The most effective way of  $\alpha$ -galactosides decreasing is germination. The content of  $\alpha$ -galactosides in chick-pea was decreased by 4 days germination up to 24% of original value in dry seeds. The 2 days germinated chick-pea seeds were treated by pressure 500 MPa for 10 minutes. The  $\alpha$ -galactosides content was decreased by this treatment up to 31% of value before pressurisation and during 21 days storing decreased up to 7% of value before pressurisation.

**Keywords:** chick-pea;  $\alpha$ -galactosides; germination; high-pressure treatment; storage

### INTRODUCTION

Chick-pea (*Cicer arietinum* L.) is important grain legume with high nutritive value (high content of protein with biological value coming up animal protein biological value, high content of some vitamins, minerals and dietary fibre) [1], but the chick-pea consumption is negligible in Czech Republic. Main reasons very low chick-pea consumption, estimated by our sociological research [2], are unfamiliarity with chick-pea and health problems after consumption e.g. flatulence. Flatulence is caused, most of all, by presence of  $\alpha$ -galactosides.  $\alpha$ -Galactosides – oligosaccharides of the raffinose family – RFO – (such as raffinose, stachyose, verbascose etc.) – contain  $\alpha$ -1,6-galactose linkage indigestible to mammalian enzymes [3]. They are transported no changed to colon, where are fermented by anaerobic intestinal microflora to short chain fatty acids and various flatus gasses (hydrogen, methane, etc.), so that produces discomfort to the consumer. Germination has been documented to be an effective treatment to remove antinutritional factors (e.g. phytates and raffinose oligosaccharides) in legumes [4]. During this process  $\alpha$ -galactosides are degraded producing available sugars. The content  $\alpha$ -galactosides decreases and number of microbes increases during germination of grain

legume seeds. The microbial contamination of germinated seeds is the main reason of their short shelf life and suitability for food or dishes preparation. The suitable technology for destroying of microbial contamination (with exception of spores) of germinated grain legume seeds is high pressure processing [5]. This technology ensures the high quality of food products (flavour, colour, vitamins content, biological active components, etc.) similar to that of the fresh raw materials [6]. The goal of this paper was the determination of  $\alpha$ -galactosides changes in chick-pea seeds during germination, high pressure treatment of germinated seeds and during storage germinated seeds treated by high pressure.

### EXPERIMENTAL

#### Material

**Plant material.** The chick-pea seeds (*Cicer arietinum* L.), year of harvest: 2002, origin: Turkey.

**Packaging material for high pressure treated germinated chick-pea seeds storage.** PA/PE 80 bags (VAC STAR).

**Standards.** Raffinose and Stachyose: Sigma Aldrich, Deutschland, Verbascose: Megazyme International, Ireland.

## Equipments

**High pressure equipment.** Press CYX 6/0103 (ZDAS Joint Stock Co., Czech Republic). Working pressure up to 600 MPa, chamber volume – 2 l, power input – 7.5 kW, pressure medium – tap water.

**Drying equipment.** Drying balances Precisia HA60, Switzerland.

**HPLC equipment.** System Dionex (Sunnyvale, USA) for HPAEC with PAD, pump GS50 (Dionex Corporation, Sunnyvale, USA), electrochemical detector ED50 (Dionex Corporation, Sunnyvale, USA), thermostat STH 585 (Dionex Corporation, Sunnyvale, USA), autosampler model 234 (Gilson, France), inlet (Rheodyne with loop 100 µl), column CarboPac PA1 (Dionex), 2 × 250 mm, precolumn PA1 (Dionex) 2 × 50 mm.

## Methods

**Germination.** Chick-pea seeds were germinated 1–5 days in aerated water medium at 20°C. Seeds were incubated in 500 ml aeration bottles (80 g of seeds and 200 ml of tap water in each bottle), water was changed after 24 hours.

**Pressurisation.** Two days germinated chick-pea seeds were treated by pressure 500 MPa for 10 min in solution of citric acid in water (pH = 2.5), ratio chick-pea: citric acid solution = 2:3. The acid medium is necessary for spores germination prevention.

**Storage of treated material.** High pressure treated samples of germinated chick-pea seeds in the solution of citric acid were stored in refrigerator at temperature 5–8°C. Time of storage 1–21 days.

Samples were analysed for of  $\alpha$ -galactosides content after 7, 14 and 21 days of storage.

**Dry matter determination.** Dry matter was determined by drying to constant weight with help of drying balances.

**$\alpha$ -galactosides determination.**  $\alpha$ -Galactosides were extracted from grounded chick-pea seeds by boiling with ethanol for 60 min. Extracted  $\alpha$ -galactosides were determined on above described equipment under following conditions. Mobile phase: NaOH solution in deionised and degassed water (gradient: 16mM to 191mM, 4.6mM/min), flow rate: 0.25 ml/min, temperature: 25°C. Contents of individual  $\alpha$ -galactosides were quantified by inner standard method (standards of raffinose –raf, stachyose –sta, and verbascose –ver were used).

## RESULTS AND DISCUSSION

The results of  $\alpha$ -galactosides content determination in chick-pea seeds during germination are demonstrated on Figures 1 and 2. The  $\alpha$ -galactosides content is continually decreasing up to 4<sup>th</sup> day of germination (up to 24% of original value in dry seeds). The fall is made by hydrolysis on  $\alpha$ -1,6-galactose linkages and by extraction to water medium. The increasing is probably caused by unblocking of  $\alpha$ -galactosides from bounded forms. The ratio of individual  $\alpha$ -galactosides is changed during germination due to these processes.

Two days germinated chick-pea seeds were chosen for high pressure treatment. The  $\alpha$ -galactosides content in this seeds was not minimal, but microbial contamination was not too high. In

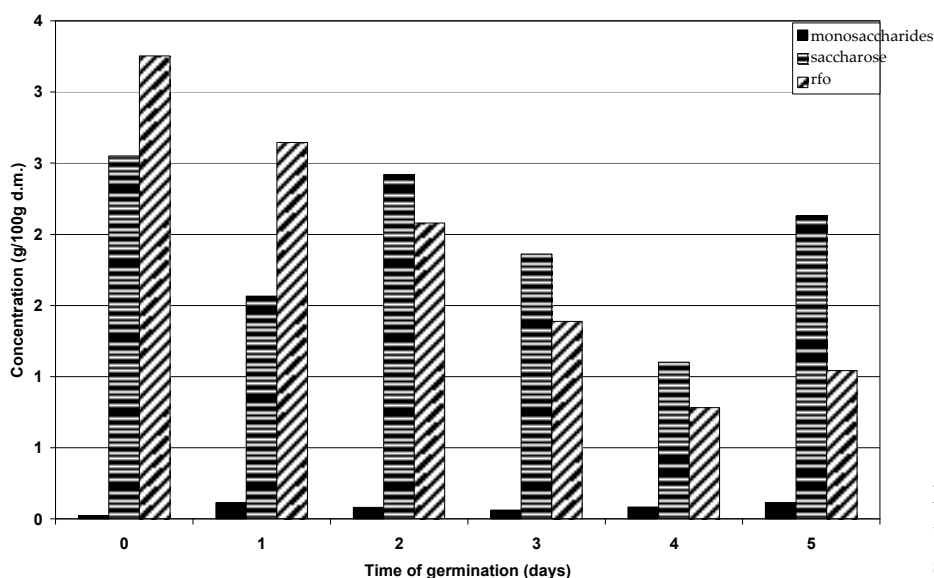


Figure 1. Changes of  $\alpha$ -galactosides content in chick-pea seeds during germination

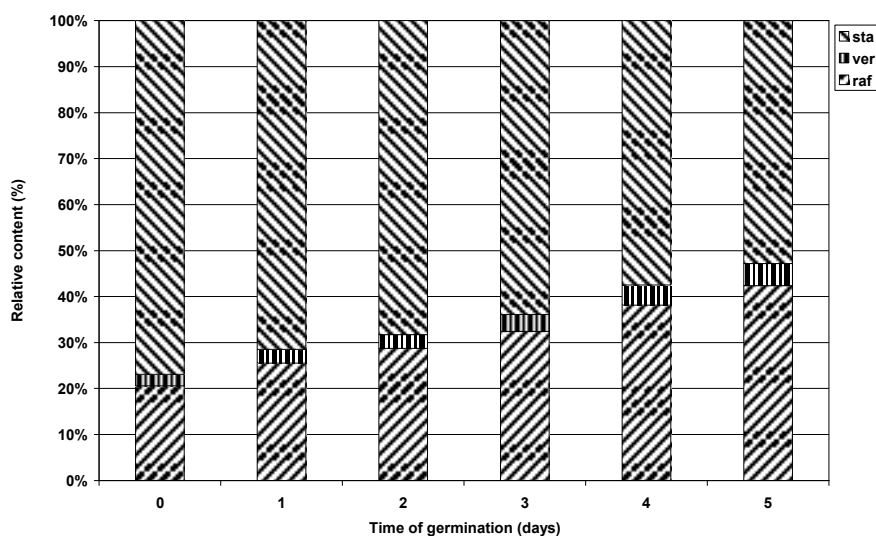


Figure 2. Changes of ratio of individual  $\alpha$ -galactosides in chick-pea seeds during germination

addition, the small quantity of  $\alpha$ -galactosides is, from nutritional point of view, desirable, because they act as prebiotics. The results of  $\alpha$ -galactosides content determination in 2 days germinated high pressure treated chick-pea seeds during storage are demonstrated on Figure 3. The  $\alpha$ -galactosides content is decreased by high pressure processing up to 31% of original value before pressuring and it is continually decreasing during storage up to 7% of original value after 21 days of storing. This decrease is probably caused by hydrolysis with high pressure activated  $\alpha$ -galactosidases and acid hydrolysis.

## CONCLUSIONS

The germination is suitable technological processing for  $\alpha$ -galactosides content in chick-pea seeds decreasing and consequently for their digestibility increasing. The content of  $\alpha$ -galactosides in chick-pea was decreased by 4 days germination up to 24% of original value in dry seeds. The high pressure treatment and following refrigerate storage are suitable technologies not only for microbes destroying but for further  $\alpha$ -galactosides content decreasing as well. The  $\alpha$ -galactosides content of 2 days germinated chick-pea seeds treated by

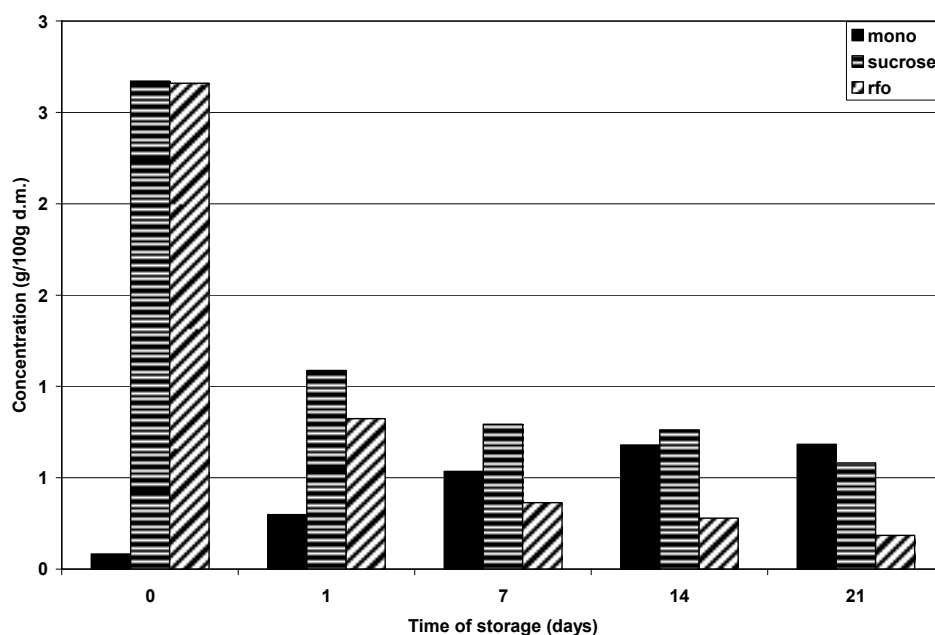


Figure 3. Changes of  $\alpha$ -galactosides content in 2 days germinated high pressure treated chick-pea seeds during storage

pressure 500 MPa for 10 minutes was decreased up to 31% of value before pressurisation and during 21 days storing decreased up to 7% of value before pressurisation.

**Acknowledgement:** The work was realised with financial support of projects QF3287 of the Ministry of Agriculture of the Czech Republic and 223300004 and 223300005 of the Ministry of Education, Youth and Sports of the Czech Republic.

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