Effect of heat treatment and germination on alpha amylase inhibitor activity in chick peas (Cicer arietinum L.)

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Received 28 June 1992; accepted in revised form 10 May 1993

Key words: Alpha amylase inhibitor, Chick pea, Germination, Heat treatment, Pressure cooking, UV irradiation

Abstract. Chick pea seeds of twenty eight varieties were analysed for alpha amylase inhibitor activity (AIA) using salivary amylase. The effects of heat treatment and germination on the activity of the antinutritional factor was investigated. Heat treatment and germination decreased the activity of amylase inhibitor. Chick pea meal was also subjected to UV irradiation and pressure cooking. These treatments decreased alpha amylase inhibitor activity. The amylase inhibitor activity decreased as the days of germination increased and negligible inhibitor activity was observed on the 6th day of germination.

Introduction

Among food legumes chick pea (Cicer arietinum L.) is a valuable source of protein, minerals and vitamins. It occupies an important place in human nutrition in many developing countries [1]. Chickpea has many antinutritional factors in it and alpha amylase inhibitor is one among them [2, 15]. The antinutritional factors present in raw pulses are partly removed during domestic and industrial processing resulting in improved nutritive quality [3, 4]. Dry and moist heating at normal or increased pressure are major steps in processing procedures. Effects of heat and germination on antinutritional factors in pulses have been reported [5, 6]. However, the major emphasis so far has been on soybean and Phaseolus species [7]. Simple processing methods like heat treatment and germination have been shown to reduce the level of amylase inhibitor activity in cereals like wheat [8] and legumes; such information on chick pea is, however, lacking. The purpose of present investigation is to study the effects of heat treatment, germination and UV irradiation on alpha amylase inhibitor activity in chick peas.

Materials and methods

Chick pea seeds were procured from the Agriculture Research Station, Gulbarga and local market. Seeds were passed on the screen to remove debries and broken
seeds and stored in a polythene bags. Human salivary amylase was partially purified according to the method of Meyer et al. [9]. All the chemicals used were of analytical grade, purchased from Loba Chemicals, Bombay.

Heat treatment. The chick pea extract was subjected to heating in a boiling water bath at 100 °C. The heating was done at different time intervals and cooled.

Pressure cooking. About 2 grams of seeds were soaked in distilled water at 4 °C. The soaked seeds were then subjected to pressure cooking in a cooker for different time intervals. After cooking the seeds were grounded in a pestle and mortor.

UV irradiation. Two grams of defatted chick pea meal was subjected to UV light in an UV chamber. The UV light exposure was at different time intervals. Then the exposed sample was used for assay.

Germination. The chick pea seeds were sterilized by treating with 0.1% mercuric chloride. The sterilized seeds were washed and soaked overnight at 4 °C. The soaked seeds were placed on moist filter papers in petridishes and incubated at 30 °C. The seeds were moistened with distilled water at regular intervals of 12 hours. The inhibitor activity was determined at different days of germination.

Alpha amylase assay. The assay of alpha amylase was carried out by the method of Bernfeld [10]. The reaction mixture contained; 0.5 ml enzyme solution and 0.5 ml of substrate were incubated at room temperature (37 °C) for 3 minutes and the reaction was terminated by adding 1 ml of DNS reagent. Then all the tubes were kept in boiling water bath for 5 minutes and cooled. The amount of maltose liberated by enzyme was measured at 540 nm, after adding 10 ml of distilled water. The activity is defined as the amount of enzyme that liberates one μ mole of maltose under assay conditions per minute.

Alpha amylase inhibitor assay. Two grams of fine powdered and dried sample was stirred with 16 ml of 0.02 M phosphate buffer of pH 6.9. The homogenate was stirred for about 5–6 hours in cold at 4 °C and centrifuged in 1 Remi cooling centrifuge at 10,000 × g for 20 minutes at 0 °C. The supernatent solution was subjected to dialysis against 0.1 M phosphate buffer overnight. The clear dialysed solution was used for assay of amylase inhibitor. One unit of inhibitor activity is defined as the amount that reduced amylase activity by one unit.

Results and discussion

Effects of heat treatment. Effects of heat treatment on amylase inhibitor in chick pea (variety, Jawari small) are presented in Table 1. The process of