In-vitro digestibility and Functional properties of Chickpea Resistant starch

Publication History
Received: 11 November 2015
Accepted: 19 December 2015
Published: 1 January 2016

Citation
In-vitro digestibility and Functional properties of Chickpea Resistant starch
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Abstract
Starch is the most abundant carbohydrate in chickpea seed and is considered to be competitive in the food industry. The study was aimed to investigate the in-vitro digestibility and functional properties of chickpea resistant starch through repeated (five cycle) retrogradation process. Chickpea starch was subjected to retrogradation (121°C with 30 minutes) and different storage time intervals of 24, 48 and 72 h at 4°C. The in-vitro digestibility and functional properties of native chickpea starch (NCPS) and treated starches were determined according to the standard procedures. Water binding capacity (WBC) and solubility of treated starches were significantly (P<0.05) increased whereas swelling power was (P<0.05) decreased than native chickpea starch. No significant difference was observed in bulk density (BD), moisture and dry matter among the starches. Compared to native chickpea starch, the RS content of treated starches were significantly (P<0.05) increased with time of storage whereas RDS content was decreased except in T2. The SDS content of treated starches were significantly (P<0.05) decreased than native chickpea starch (NCPS). Hence, repeated (five cycle) retrogradation was one of the methods for resistant starch formation and it can be used in the preparation of bakery products.

Key words: Retrogradation, Invitro digestibility, chickpea starch, functional properties.

Introduction
Chickpea (Cicer arietinum L.) is the largest made food legume in South Asia and therefore the third largest made food legume globally. India graded first in terms of chickpea production and consumption within the world. Concerning 65% of world space with 68% of world production of chickpea is contributed by India (Reddy and Mishra, 2010). Current opinion on healthy intake habits could result in an increase the proportion of legume starches as associate alternate to cereals (Tharanathan and Mahadevamma, 2003). Starch is that the rich saccharide in chickpea seed and is taken into account to be competitive within the food industry (Goni and Valentin-Gamazo, 2003).

The digestion rate powerfully depends on processing and therefore the state of the starch (Lehmann and Robin, 2007) sources of starch with a variety of substance of rapidly digestible starch (RDS), slowly digestible starch (SDS) and resistant starch (RS) are of nice interest (Pongjant et al., 2009). Resistant starch (RS) has recently gained consideration as a purposeful food ingredient as result of its potential health benefits and purposeful properties in foods (Sajilata et al., 2006). In step with Englyst, et al., (1992), RS is classified into four major groups; RS1, RS2, RS3, and RS4.Starch retrogradation, further as short retrogradation and long term retrogradation, is an inevitable development that gelatinized starch changes from an amorphous state to a crystalline space (Tian et al., 2009a). The long term retrogradation primarily related to amylopectin might reduce the digestibility of the starch by amylase (Tian et al., 2009b). Park et al., (2009) revealed that temperature-cycled retrogradation provided a wonderful amount of resistant starch from waxy maize starch. Sources of cereal grains, roots, tubers and legumes manufacture resistant starch through the process of cyclic heating, autoclaving and extrusion ways.
In general, several ways involving physical, chemical and enzymatic transformations are used to change the properties of starch that enhance health attributes and/or minimize defects in structure. Researchers have tried to enhance the RS yields by: (1) heat-moisture treatment and annealing (Brumovsky and Thompson, 2001) (2) enzyme method (Vatanasuchart et al., 2010; Zhang and Jin, 2011), (3) combined heat/enzyme treatment (Mutungi et al., 2009) and (4) chemical treatment (Haynes et al., 2000). Since customers are progressively inquisitive about natural and organic foods for healthiness and ecological reasons, using physical treatment seems more attractive. However, there was no work had been reportable on preparation of resistant starch from chickpea starch by retrogradation, thus the current aim of our work is to know the impact of repeated (five cycle) retrogradation on chemical, functional, pasting, structural characteristics and in vitro digestibility of chickpea starch.

**Materials and Methods**

**Materials**

Chickpea was purchased from Tamil Nadu Agricultural University, Coimbatore. Starch was isolated from chickpea by the method of Vasanthan (2001). This starch was used for preparation of Repeated (five cycle) retrograded starch. All other chemicals and reagents were of reagent grade.

**Preparation of repeated (five cycles) retrograded chickpea starch**

Repeated (five cycle) retrograded chickpea starch was prepared according to the method of Xie et al., (2014) with slight modifications. Repeated (five cycles) retrogradation treatments can be described as follows: chickpea starch (25.0 g) and distilled water (50 ml) were placed in a 150 ml glass container, sealed, and heated in boiling water at 121°C for 30 min. The resultant gel was hermetically sealed and stored at 4°C for 24h, 48h and 72h, respectively, to perform single cycle of retrogradation treatment. The retrograded samples were heated in boiling water for 30 min again and subjected to 5 cycles of retrogradation treatment with time intervals of 24h, 48h and 72h. Under these conditions above, chickpea starch was treated successively by triple retrogradation to perform 5 cycles. After treatment, each of resulting gel was cut into pieces (less than 5 cm in length and thickness) then oven dried at 45°C temperature for 4-6h. All samples were fine grained to submit a 100-mesh sieve to obtain the starch products.

**Measurement of in vitro digestibility of starch**

Invitro digestibility of the starch samples were estimated by previously described methods (Englyst et al., 1992; Englyst et al., 1999 and Zhang et al., 2011) with some modification.

**Functional properties of native and treated starches**

Water binding capacity of the samples was determined by the method of Yamazaki (1954). Swelling power and solubility of the starches were determined by the strategy of Gani et al., (2010). Bulk density was estimated by the method of Adeleke and Odedeji (2010). Moisture content and dry matter were estimated by the method of Adebayo et al., (2010).

**Statistical Analysis**

The data were analyzed using an SPSS version 16.0. The mean and standard deviation of means of the triplicate analysis of the samples were calculated. Analysis of Variance (ANOVA) was performed to determine significant differences between the means, while the means were separated with the least significant difference (LSD).
Results and Discussion

In vitro digestibility profiles of native chickpea starch and treated starches

RDS, SDS and RS contents of the native chickpea starch and treated starches (T1, T2 and T3) are presented in fig.1. The results showed that RS content of repeated (five cycle) retrograded starches, T1 (74.57%), T2 (76.92%) and T3 (83.54%), were significantly (P<0.05) increased than native chickpea starch (40.16%). Hughes et al., (2009) also reported that RS content of native chickpea starch was in the range of 24.14-40.57%. The RS content of repeated (five cycle) retrograded starches increased with increased retrogradation time from 24h to 72h. The RDS content of treated starches, T1 (6.46%), T2 (14.99%) and T3 (1.32%), were significantly (P<0.05) decreased than native chickpea starch (7.78%) except for T2. The SDS content of treated starches-T1 (18.96%), T2 (8.08%) and T3 (15.13%) were significantly (P<0.05) decreased than native chickpea starch (52.05%). Dundar and Gocmen (2013) reported that the RS yield of starch increased from 24.94% (24h-storage time) to 30.41% (72h-storage time) at an autoclaving temperature of 145ºC. Erlangen et al., (1993) reported that yields of resistant starch mostly depended on storage time and on temperature, and that storage temperature influenced the type of resistant starch crystals (A or B, X-ray diffraction pattern) formed. Vasanthan and Bhatti (1998) reported that the effect of retrogradation and annealing on the resistant starch content of amylomaize, barley, field pea, and lentil. In their study, they indicated that the RS content of retrograded starch gels (6–9%, w/w) were on top of the native starches (3–5%, w/w).

![Fig. 1 Measurement of in vitro digestibility of native chickpea starch (NCPS) and treated starches (T1, T2 and T3). All values are means of triplicates ± standard deviation. Bars assigned with different superscripts are significantly different (P<0.05). RDS, SDS, RS indicate rapidly digestible starch, slowly digestible starch and resistant starch respectively. NCPS=Native Chickpea Starch, T1, T2 and T3 represent repeated (5 cycles) retrograded starches stored at 4ºC for 24h, 48h and 72h.](image-url)
Functional properties of native chickpea starch and treated starches

Functional properties of native chickpea starch and treated starches (T1, T2 and T3) are presented in Table 1. Water binding capacity (WBC) and solubility of treated starches were significantly (P<0.05) increased whereas swelling power was (P<0.05) decreased than native chickpea starch as the storing time increased. Similar result like increase water binding capacity, solubility and decreased swelling power is observed by Reddy et al., (2013) in kidney bean starch. Similar result like increase water binding capacity, solubility and decreased swelling power is observed by Reddy et al., (2013) in kidney bean starch. Similar result like increase water binding capacity is observed by Dundar and Gocmen (2013). Water binding capacity of all samples was compared to other studies of RS from chickpea starch (Polesi and Sarmento 2011). An increased water activity in retrograded starch is the result of a change in molecular structure or any other mechanisms leading to an easier mobility of the starch, where leaching of starch was also observed (Govindasamy et al., 1996). The increase in water binding value was primarily because of the gelatinization caused by heating and autoclaving at higher temperature (Dundar and Gocmen, 2013). Water solubility is often used as a degradation indicator of molecular components. The increase in solubility might occur as a result of changes within the molecular structure or as an independent mechanism that results in the mobility of the starch elements, leading to the in leach of carbohydrates from molecules involved (Govindasamy et al., 1996).

Table 1. Functional properties of native chickpea starch and treated starches

<table>
<thead>
<tr>
<th>Functional properties</th>
<th>NCPS</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
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<tbody>
<tr>
<td>WBC</td>
<td>93.59±3.12&lt;sup&gt;a&lt;/sup&gt;</td>
<td>166.66±1.51&lt;sup&gt;b&lt;/sup&gt;</td>
<td>169.18±2.31&lt;sup&gt;b&lt;/sup&gt;</td>
<td>175.24±2.31&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Swelling power</td>
<td>9.54±0.23&lt;sup&gt;a&lt;/sup&gt;</td>
<td>6.06±0.11&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5.8±0.1&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5.63±0.30&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Solubility</td>
<td>2.33±0.57&lt;sup&gt;a&lt;/sup&gt;</td>
<td>12.4±0.43&lt;sup&gt;b&lt;/sup&gt;</td>
<td>13.06±0.15&lt;sup&gt;b&lt;/sup&gt;</td>
<td>13.43±0.35&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>BD</td>
<td>0.94±0.09&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.93±0.02&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.91±0.02&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.93±0.00&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Moisture</td>
<td>10.93±0.62&lt;sup&gt;a&lt;/sup&gt;</td>
<td>12.1±4.60&lt;sup&gt;a&lt;/sup&gt;</td>
<td>12.07±1.81&lt;sup&gt;a&lt;/sup&gt;</td>
<td>12.78±0.61&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Dry matter</td>
<td>89.12±0.63&lt;sup&gt;a&lt;/sup&gt;</td>
<td>87.89±4.60&lt;sup&gt;a&lt;/sup&gt;</td>
<td>87.91±1.81&lt;sup&gt;a&lt;/sup&gt;</td>
<td>83.16±6.42&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

All values are means of triplicates ± standard deviation. Means in the same row with different letters are significantly different (P<0.05). WBC and BD indicate Water binding capacity and Bulk density.

Zhou et al., (2004) proposed that lower swelling power of wrinkled pea starch might be attributed to its lower amylopectin content and strong interaction between amylose chains. Consequently, the treatment of chickpea starch with retrogradation may lead to the depolymerization of amylopectin, making the starch to lose the ability to hold absorbed water and swell (Wang and Wang, 2003). Low swelling power of treated starches may additionally as a result of existence of huge number of crystallites fashioned by the association between amylpectin chains throught storage. Starch granular stability is increased as result of crystallite formation and swelling power decreases (Sing et al., 2004). No significant difference was discovered in bulk density (BD), moisture and dry matter among the starches.

Conclusion

The results showed that the repeated (5 cycle) retrogradation of chickpea starch with longer storing time (72h), showed beneficial impact on RS formation. When chickpea starch was subjected to repeated (5 cycle) retrogradation with longer storing time (72h), the highest RS yield was obtained. Water binding capacity (WBC) and solubility of treated starches were significantly (P<0.05) increased whereas swelling power was (P<0.05) decreased than native chickpea starch. Compared to native chickpea starch, the RS content of treated starches were significantly (P<0.05) increased with time of storage whereas RDS content and SDS was
significantly (P<0.05) decreased. Compared to the native starch, the RS preparations obtained in the present study was useful for the preparation of food product that need comparatively higher water binding capacity, solubility and less swelling power.

Acknowledgement
The authors gratefully acknowledge the Department of Food Science and Nutrition for providing University Research Fellow (URF) and Tamil Nadu Agricultural University, Coimbatore for providing desi chickpea seeds.

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