



# In vitro starch hydrolysis and estimated glycemic index of tef porridge and injera



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## ABSTRACT

The aim of this study was to investigate the in vitro starch digestibility of injera and porridge from seven tef varieties and to estimate their glycemic index. The total starch, free glucose, apparent amylose, resistant, slowly digestible and rapidly digestible starches of the varieties ranged between 66 and 76, 1.8 and 2.4 g/100 g flour dry matter (DM), 29 and 31%, 17 and 68, 19 and 53, 12 and 30 g/100 g starch DM, respectively. After processing into injera and porridge, the rapidly digestible starch content increased by 60–85% and 3–69%, respectively. The estimated glycemic index of porridge and injera of the varieties ranged 79–99 and 94–137 when estimated based on model of Goni et al. (1997) whereas from 69 to 100 and 94 to 161, respectively based on Granfeldt et al. (1992). Tef porridge and injera samples studied here can be classified as medium–high GI foods, not to be considered as a proper food ingredient for diabetic people and patients in weight gain control.

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## 1. Introduction

Frequent consumption of high glycemic index (GI) carbohydrate foods is increasingly associated with higher risk of obesity, coronary heart disease, type 2 diabetes, cancer and other chronic syndromes. Glycemic index of a particular meal determines the rate of blood glucose rise (Sasaki, Okunishi, Sotome, & Hiroshi, 2016). Type 2 diabetes prevalence of Ethiopia adjusted to its national population was 4.4% (about 4.14 million) in 2013 and is projected to be 5.1% (about 7.75 million) by 2035 (Guariguata et al., 2014).

There is no single solution to suppress the increase of postprandial blood glucose level and the associated health disorders, however, incorporating organic acids in a meal, slow and low heat cooking process, replacing portions of carbohydrates by proteins, use of whole flour breads and fruits and vegetables has been recommended. Adherence to low glycemic index (GI) food and/or limited amount intake of high GI foods has been also reported as a major mitigation strategy to control the increase of blood glucose level in people with diabetes type 2 and to those of in body weight management (Karl et al., 2015).

Based on an in vitro measure of rate and extent of starch digestion, Englyst, Kingman, and Cummings (1992) categorized starches of different sources as rapidly digestible starch (RDS)-starches hydrolyzed within the first 20 min of digestion, slowly digestible starch (SDS)-starches digested within the following 100 min after RDS, and resistant starch (RS)-starches not digested within the 120 min of in vitro digestion. RDS causes a rapid increase in blood glucose level after ingestion, whereas SDS releases glucose slowly and consistently over an extended time. RS which resists enzymatic hydrolysis, is fermented in the large intestine releasing short chain fatty acids which are considered as beneficial (Lehmann & Robin, 2007). The rate of digestion of a typical starchy food is influenced by its botanical origin, which consequently determines the structure and shape of starch granules and the amylose content (Frei, Siddhuraju, & Becker, 2003), physicochemical structure of the starch such as crystallinity, chain length and chain distribution, molecular weight and weight distribution (Tian et al., 2016), thermal processing and moisture content, which determine the extent of starch gelatinization (Bjorck, Granfeldt, Asp, Liljeberg, & Tovar, 1994; Sasaki et al., 2016) and the presence of dietary fiber that changes the microstructure of foods and limits its water availability, and thus restricting starch gelatinization (Cleary & Brennan, 2006).

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Tef [*Eragrostis tef* (Zucc.) Trotter] is a cereal crop that has virtually been cultivated as human food only in Ethiopia for more than two thousand years (D'Andrea, 2008). Currently, it is acquiring popularity due to its high protein content and preferred amino acid profile, gluten free, high iron and fiber contents. So far, there are reports on the amylose content of tef, that ranged from 20 to 32% (Bultosa, Hall, & Taylor, 2002; Hager, Wolter, Jacob, Zannini, & Arendt, 2012) depicting a wide variability of normal to high amylose starch. High amylose starches require temperatures of up to 150 °C in the presence of water to fully gelatinize, which is not indeed attainable under normal cooking and baking circumstances and thus could result in foods with a lower GI (Van Amelsvoort & Weststrate, 1992). Soil & Crop Improvement (SCBV, 2007-01)-tef information map version and stated that the total starch content of tef is 60 g/100 g, of which the content of RDS, SDS and RS accounted for 20, 50 and 30 g/100 g DM starch, respectively and Abebe, Collar, and Ronda (2015) also showed RS, SDS and RDS fractions in the range of 7–11 g/100 g, 31–41 g/100 g and 29–33 g/100 g DM tef flour. Estimated glycemic index (eGI) of 74 and 45 for bread and egg pasta respectively from unknown tef varieties were reported by Wolter, Hager, Zannini, and Arendt (2013) and Hager, Czerny, Bez, Zannini, and Arendt (2013), respectively.

There are about 33 improved tef varieties (Derbew, 2013) and hundreds of farmers' local varieties in Ethiopia, differing in seed size and color from milky-white to almost dark-brown. However, there is no study on the properties of GI of the common tef food products such as injera (pancake) and porridge. Injera and porridge are among the major food products of tef and are staples of millions mainly in Ethiopia and frequently used in Ethiopian restaurants in major cities around the world. Therefore, the aim of this study was to investigate the in vitro starch digestibility and estimate the GI of fresh injera and porridge prepared from seven tef varieties which vary in color from brown to white.

## 2. Materials and methods

### 2.1. Sample and preparation

Tef varieties, Boset (DZ-Cr-409), Dega (DZ-01-2675), Quncho (DZ-Cr-387), Simada (DZ-Cr-285), Tsedey (DZ-Cr-37), Zagurey (local) and Zezew (local) were used in this study. The first five are white whereas the last two are brown seed color varieties. These tef varieties were obtained from Axum Agricultural Research Center (Tigray, Ethiopia). They were dried before harvest on the field and milled by disc attrition milling at a local tef miller, in the same way as tef is normally milled for the preparation of injera and porridge in Ethiopia. Some portions (about 1 kg) of each variety was pre-milled prior to each variety and discarded to prevent cross-contamination among the varieties. The flour moisture contents ranged from 7.9 to 8.4 g/100 g flour, with an average of 8.1 g/100 g flour and were not significantly different among varieties ( $p > 0.05$ ). The distribution of the flour particle size of the tef varieties was measured using a test sieve shaker (Endecott, LTD, London SW, England). This was in the range of  $100\% < 850 \mu\text{m}$ ,  $99\text{--}100\% < 425 \mu\text{m}$ ,  $96\text{--}99\% < 300 \mu\text{m}$ ,  $78\text{--}85\% < 212 \mu\text{m}$ ,  $66\text{--}77\% < 150 \mu\text{m}$ .

Fermented tef injeras were prepared following the procedure described by Urga and Narasimha (1997). Each of the varieties were spontaneously fermented for 42 h at 25 °C and subsequently baked at 180 °C for about 3 min. Stiff tef porridge was prepared following the traditional method. Briefly, tef flour and water were mixed in a ratio of 2:5 and cooked for 8 min at about 180 °C. The process of both injera and porridge making was based on the traditional practices in Ethiopia. The food products were sampled as eaten (fresh, when the temperature of the food was about 40 °C).

Three independent preparations were made for each product from each variety.

### 2.2. Free glucose (FG)

The FG content was measured according to Englyst et al. (1992) using an assay kit GOPOD-format K-GLUC 09/14 (Megazyme International Ireland Ltd) and was calculated as:

$$\% \text{glucose} = \frac{A_t \times V_t \times C \times D}{A_s \times W_t} \times 100$$

where

$A_t$ : absorbance of test solutions,  $V_t$ : total volume of test solutions ( $V_t = 25.2$  plus 1 mL per gram wet weight of samples used),  $C$ : concentration ( $C = 0.394$  mg glucose/mL) of standard, which may be corrected for moisture content,  $D$ : dilution factor = 18.

### 2.3. Total starch and starch digestibility fractions

#### 2.3.1. Total starch (TS)

The TS content was measured according to Englyst et al. (1992) using an assay kit GOPOD-format K-GLUC 09/14 (Megazyme International Ireland Ltd), calculated and expressed as:

$$TS = (TG - FG) \times 0.9 (\text{g starch}) / 100 \text{g flour DM.}$$

TG: total glucose

0.9: glucose to starch conversion factor

#### 2.3.2. Rapidly digestible starch (RDS)

The RDS content was measured based on an in vitro starch digestibility procedure (Englyst et al., 1992) using an assay kit GOPOD-format K-GLUC 09/14 (Megazyme International Ireland Ltd), calculated and expressed as:  $RDS = (G_{20} - FG) \times 0.9$  (g RDS)/100 g starch DM. where  $G_{20}$ : glucose content after 20 min of digestion, 0.9: glucose to starch conversion factor

#### 2.3.3. Slowly digestible starch (SDS)

The SDS content was measured based on an in vitro starch digestibility procedure (Englyst et al., 1992) using an assay kit GOPOD-format K-GLUC 09/14 (Megazyme International Ireland Ltd), calculated and expressed as:  $SDS = (G_{120} - G_{20}) \times 0.9$  (g SDS)/100 g starch DM. where  $G_{120}$ : glucose content after 120 minutes of digestion

$G_{20}$ : glucose content after 20 min of digestion

0.9: glucose to starch conversion factor

#### 2.3.4. Resistant starch (RS)

The RS content was determined based on an in vitro starch digestibility procedure (Englyst et al., 1992), calculated and expressed as:  $RS = TS - (SDS + RDS)$  (g RS)/100g starch DM.

### 2.4. Apparent amylose content

The amylose content of the starch of each tef flour was determined by the Megazyme kit K-AMYL (Megazyme International Ireland Ltd). The amylose content was calculated as:

$$\text{Amylose}(\%) \left( \frac{W}{W} \right) = \frac{\text{Absorbance}(\text{Con A supernatant})}{\text{Absorbance}(\text{total starch aliquot})} \times \frac{6.15}{9.2} \times 100/1$$

where 6.15 and 9.2 are dilution factors for the Con A and total starch extracts, respectively.

Con A: Concanavalin A

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