

### **3. RESEARCH**

#### **3.1. Improving the quality of sorghum *injera* by decortication and compositing with tef**

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## Abstract

*Injera* is an Ethiopian fermented leavened pancake-like bread made from cereals, with tef being preferred. Decortication and compositing with tef were evaluated as methods to improve the *injera* making quality of red tannin-free and tannin-containing sorghums. Both decortication and compositing improved sorghum *injera* quality. Concerning decortication, mechanical abrasion was found to be more effective than hand pounding because acceptable *injera* was obtained with lower milling loss. Good quality *injera* was produced at an extraction rate of 54% for tannin-containing and 83% for tannin-free sorghum. With compositing, good quality *injera* was produced with a 50:50 composite of whole tannin-containing sorghum and tef. Both processes reduced the tannin content of the flours, which appeared to relieve the inhibiting effects of tannins on the fermentation. Decortication also seemed to improve sorghum flour *injera* making quality by improving flour pasting, probably as a result of reducing the level of interfering substances such as lipids and proteins. In contrast, the improvement brought about by compositing with tef seemed to be due to inherent differences between tef and sorghum starch granules and an increase in the water solubility index of the flour. Compositing seems to be a more useful method of improving sorghum *injera* quality than decortication as it avoids the grain loss associated with decortication.

### **3.1.1. Introduction**

*Injera*, a staple food in Ethiopia, is a fermented, leavened pancake-like bread prepared from cereals such as tef and sorghum. It is characterized by having “eyes” (honeycomb-like holes) in its top surface. These are produced due to the production and escape of carbon dioxide during fermentation and baking, respectively. *Injera* prepared from flour of tef, a tiny millet-like grain, is preferred because it is soft and can be rolled. Notwithstanding this, nearly 80% of the sorghum produced in Ethiopia is used for the production of *injera* (Gebrekidan and GebreHiwot 1982). Both white and colored tannin-free sorghums are used. However, the white tannin-free sorghums are the most preferred because of the light *injera* color (Gebrekidan and GebreHiwot 1982). Tannin-containing sorghums are predominantly used for local beer production.

Since colored tannin-free and tannin-containing sorghums are lower priced than white sorghum and tef, it would be most desirable to improve their *injera*-making potential through the use of simple, practical technologies. Such technologies include decortication (often incorrectly referred to as dehulling, since the sorghum grain does not have a hull) and the use of composite flours. In fact, compositing tef flour with sorghum flour has been found to improve *injera* texture (Gebrekidan and GebreHiwot 1982, Yetneberk and Haile 1992). However, these studies were very limited in scope. The objectives of the work reported here were to evaluate grain decortication and compositing with tef flour as methods to improve the quality of *injera* made from tannin-containing sorghum and decortication for tannin-free red colored sorghum.

### **3.1.2. Materials and Methods**

#### **3.1.2.1. Materials**

A red pericarp tannin-containing cultivar (Seredo) and a red tannin-free cultivar (IS 2284) were obtained from the Ethiopian Sorghum Improvement Program (ESIP) at Melkassa Agricultural Research Center. White tef was purchased from a local market.

### *3.1.2.2. Grain characterization*

Thousand kernel weight (TKW) was measured by weighing 1000 randomly selected unbroken kernels. Glume color was determined by examining the inside of the glume after removing the kernel (Rooney and Miller 1982). The presence of a pigmented testa was determined by the Chlorox Bleach Test method (Waniska et al 1992) The vanillin-HCl method of Burns (1971) with sample blank subtraction was used to determine the level of condensed tannins. Pericarp thickness was determined by viewing longitudinally sectioned grains using a scanning electron microscope. Endosperm texture was viewed using a stereomicroscope and images were captured with a camera. Total endosperm and floury endosperm areas and pericarp thickness were measured using the UTHSCSA, Image Tool Software, version 2.0 (University of Texas Health Science Center, San Antonio, USA). The vitreous area was expressed as a percentage of the total endosperm area. Grain color was measured in L, a b units using a Hunter Lab Color Quest 45/0 (Hunter Associates, Reston, USA).

### *3.1.2.3. Decortication and milling*

Decortication was performed by hand pounding and mechanical abrasion. For hand pounding, one kg of sorghum kernels was washed and placed in a wooden mortar and pounded with a wooden pestle. Samples were removed after 3, 6 and 10 min of pounding. The number of pounding strokes was recorded for each time interval. Decorticated kernels were spread on a clean cloth and dried in the sun. The bran and decorticated grains were separated by winnowing.

Mechanical abrasion was performed using a Tangential Abrasive Dehulling Device (TADD) according to Reichert et al (1982). One hundred g of sorghum grain was decorticated for 1, 2, 3, 4, and 5 min using a TADD fitted with sand paper of 60 grit (Norton type R284 metalite) (Norton Abrasives, Worcester, USA). Extraction rate in terms of percentage decorticated grain recovered was calculated for both methods. Samples were milled to flours using an Udy cyclone mill (Seedburo, Chicago, USA) fitted with a 200  $\mu$ m opening screen size. Tef was not decorticated prior to milling.

#### 3.1.2.4. Composite flours

Whole grain flour of the tannin-containing sorghum (Seredo) was composited with tef flour at five levels: 83.3%, 66.7%, 50%, 33.3% and 16.7%. 100% Seredo flour and 100% tef flour were also included.

#### 3.1.2.5. Flour characterization

The effects of compositing sorghum flour with tef flour on flour water absorption index (WAI) and water solubility index (WSI) were determined according to Anderson et al (1969). Pasting properties of flours from the sequentially decorticated tannin-containing sorghum (Seredo) and composite flours of tef and Seredo were determined using a Rapid Visco Analyser (RVA) Model 3D (Newport Scientific, Warriewood, Australia). Flour (4 g, 14% moisture basis) was mixed with 25 ml distilled water in the RVA sample canister. A programmed heating and cooling cycle was used, where the suspension was held at 50°C for 1 min, heated at 93°C for 8 min at a rate of 6 °C/min and then held at 93°C for 5 min before cooling to 50°C within 8 min, and finally held at 50°C for 1 min. The RVA parameters measured were: peak viscosity (PV), the maximum hot paste viscosity at 93°C; hot paste viscosity (HPV), the trough at minimum hot paste viscosity; and cold paste viscosity (CPV), viscosity after cooling to 50°C and holding at this temperature.

#### 3.1.2.6. Preparation of injera

*Injera* was prepared from flours of hand pounded and mechanically abraded sorghum and the composite flours of Seredo and tef using the method described in Chapter 3.2.

#### 3.1.2.7. Sensory evaluation of injera

A semi-trained panel (a panel briefed about scoring of *injera* sensory attributes) of 6 people, who were *injera* consumers, evaluated 3 replicate samples of *injera*. The attributes used, which were generated by the author, were *injera* top and bottom surface color, description of the eyes (honeycomb structure of the top surface of the *injera*),

texture, taste, aftertaste and overall rating. Rolled pieces of *injera* (3 mm wide) were presented to the panelists on a tray at ambient temperature (about 25°C) within 2 h after baking.

#### 3.1.2.8. Statistical analysis

Analysis of variance was performed on the data to establish significant ( $p < 0.05$ ) differences between the samples. Linear regression analysis was done to establish relationships between extraction rate and flour pasting properties.

### 3.1.3. Results and discussion

#### 3.1.3.1. Grain characterization

Seredo had a lower TKW (21.5 g) compared to IS 2284 (35.9 g) (Table 3.1.1). TKW of tef was only 0.3 g. Seredo and IS 2284 had purple glume color, while tef had tan glume color. The tef grain was much lighter (L value 64.8) compared to the sorghums, Seredo (L value 36.7) and IS 2284 (L value 33.2). Seredo contained a pigmented testa and had a tannin content of 7.3% catechin equivalents, while IS 2284 and tef were tannin-free. Seredo had a much thicker and loosely packed pericarp with starch granules in the mesocarp, while IS 2284 had a thin, tightly packed pericarp with practically no mesocarp (Fig 3.1.1). Seredo had a more floury endosperm compared to IS 2284.

#### 3.1.3.2. Decortication

Two methods of decortication were evaluated: traditional hand pounding with a pestle and mortar and mechanical abrasion (simulating industrial mechanical decortication). The objectives of decorticating the sorghum grain were to remove the bran, germ and the testa (which contained tannins in Seredo), in order to improve the color, taste and appearance of the *injera*. With hand pounding an increase in the number of pounding strokes was directly related to a decrease in extraction rate for both sorghum cultivars (Fig 3.1.2A). The extraction rate of Seredo was significantly ( $p < 0.05$ ) lower (64.3%) after 400 pounding strokes compared to IS 2284 (69%). The softer endosperm and thick pericarp

(Reichert et al 1982, Shepherd 1979, Lawton and Faubion 1989) explains the reduced milling yields from Seredo.

Similar results were obtained using mechanical abrasion (Fig 3.1.2B). After 4 min of abrasion, the extraction rate for Seredo was 66.8% compared to 71.2% for IS 2284. As stated, extraction rate depends on the hardness of the grain. For Seredo, as the time of abrasion was increased to 5 min decorticated grain recovered declined to only 54%. This is because soft floury grains have a tendency to disintegrate during decortication (Rooney et al 1997). Notwithstanding this, abrasion of Seredo for 5 min reduced the tannin content of the grain from 7.3% to 1.3% catechin equivalents (Fig 3.1.2B), a reduction of 82%. Beta et al (2000b) obtained a similar reduction in tannin content (71-81%) with abrasion of Zimbabwean tannin-containing sorghums, but with very much lower decortication losses (decorticated grain recovered 81-84%), due to the grains being harder.

Sensory responses for *injera* made from hand pounded Seredo and IS 2284 flours are presented in Table 3.1.2. *Injera* made from whole Seredo flour had brown top and bottom surfaces, without eyes. It had a sticky texture and a bitter taste. Hence it was rated as poor. The tannins in Seredo were responsible for the brown color and the bitter taste of the *injera*. Tannins are known to cause dark color and astringency or bitterness in foods prepared from whole sorghum (Bacon and Rhodes 2000). Carbon dioxide produced during fermentation is known to play a fundamental role in the formation of cellular structure of leavened breads (Bloksma 1990). Thus the absence of eyes in whole flour Seredo *injera* is indicative of very little carbon dioxide being produced during fermentation. Since condensed tannins are associated with anti-microbial (Ebi et al 1999) and enzyme-inhibitory activity (Scalbert 1991) it can be assumed that the tannins inhibited the fermentation process. *Injera* prepared from Seredo flour of the lowest extraction rate (64.3%) produced by hand pounding, had a slightly bitter taste and was sticky with small, scattered eyes. This was presumably because the flour still contained significant levels of tannins. The tannin-free cultivar IS 2284 made good *injera* from hand-pounded flour of higher extraction rate (76.7%). At 68.3% extraction, a similar rate to the lowest examined for the tannin-containing Seredo, the *injera* from IS 2284 was rated very good. The much better *injera* making quality of IS 2284 at similar extraction

**Table 3.1.1.** Grain characteristics of Seredo (tannin-containing) and IS 2284 (tannin-free) sorghum cultivars and tef

Cultivar	TKW <sup>a</sup> (g)	Glume color	Pigmented <sup>b</sup> testa	Tannins (mg/100g catechin equivalents)	Pericarp thickness ( $\mu$ m)	Endosperm vitreousness (% of total endosperm area)	Grain color		
							L	a	b
Seredo	21.5 $\pm$ 0.3 <sup>b</sup> <sup>c</sup>	Purple	Yes	7.30 $\pm$ 0.27 <sup>c</sup>	126.8 $\pm$ 26.5 <sup>b</sup>	36.3 $\pm$ 3.8 <sup>a</sup>	36.7 $\pm$ 0.4 <sup>b</sup>	9.5 $\pm$ 0.2 <sup>b</sup>	12.7 $\pm$ 0.4 <sup>a</sup>
IS 2284	35.9 $\pm$ 1.7 <sup>c</sup>	Purple	No	0.47 $\pm$ 0.08 <sup>b</sup>	0.6 $\pm$ 0.1 <sup>a</sup>	48.0 $\pm$ 5.7 <sup>b</sup>	33.2 $\pm$ 0.5 <sup>a</sup>	16.4 $\pm$ 0.4 <sup>c</sup>	13.0 $\pm$ 0.2 <sup>a</sup>
Tef	0.3 $\pm$ 0.0 <sup>a</sup>	Tan	No	0.05 $\pm$ 0.00 <sup>a</sup>	ND <sup>d</sup>	ND	64.8 $\pm$ 0.7 <sup>c</sup>	3.2 $\pm$ 0.1 <sup>a</sup>	14.4 $\pm$ 0.1 <sup>b</sup>

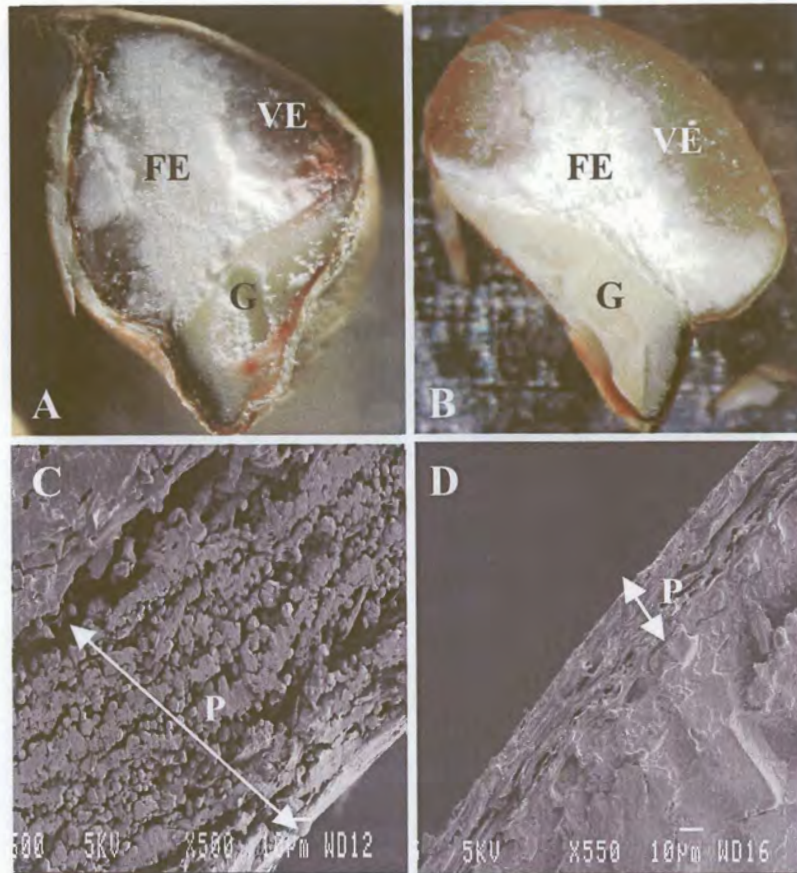
<sup>a</sup>TKW = Thousand kernel weight.

<sup>b</sup>Yes = pigmented testa present, No = pigmented testa absent.

<sup>c</sup>Values followed by the same letter in the same column are not significantly different ( $p > 0.05$ ).

<sup>d</sup>ND = Not determined.

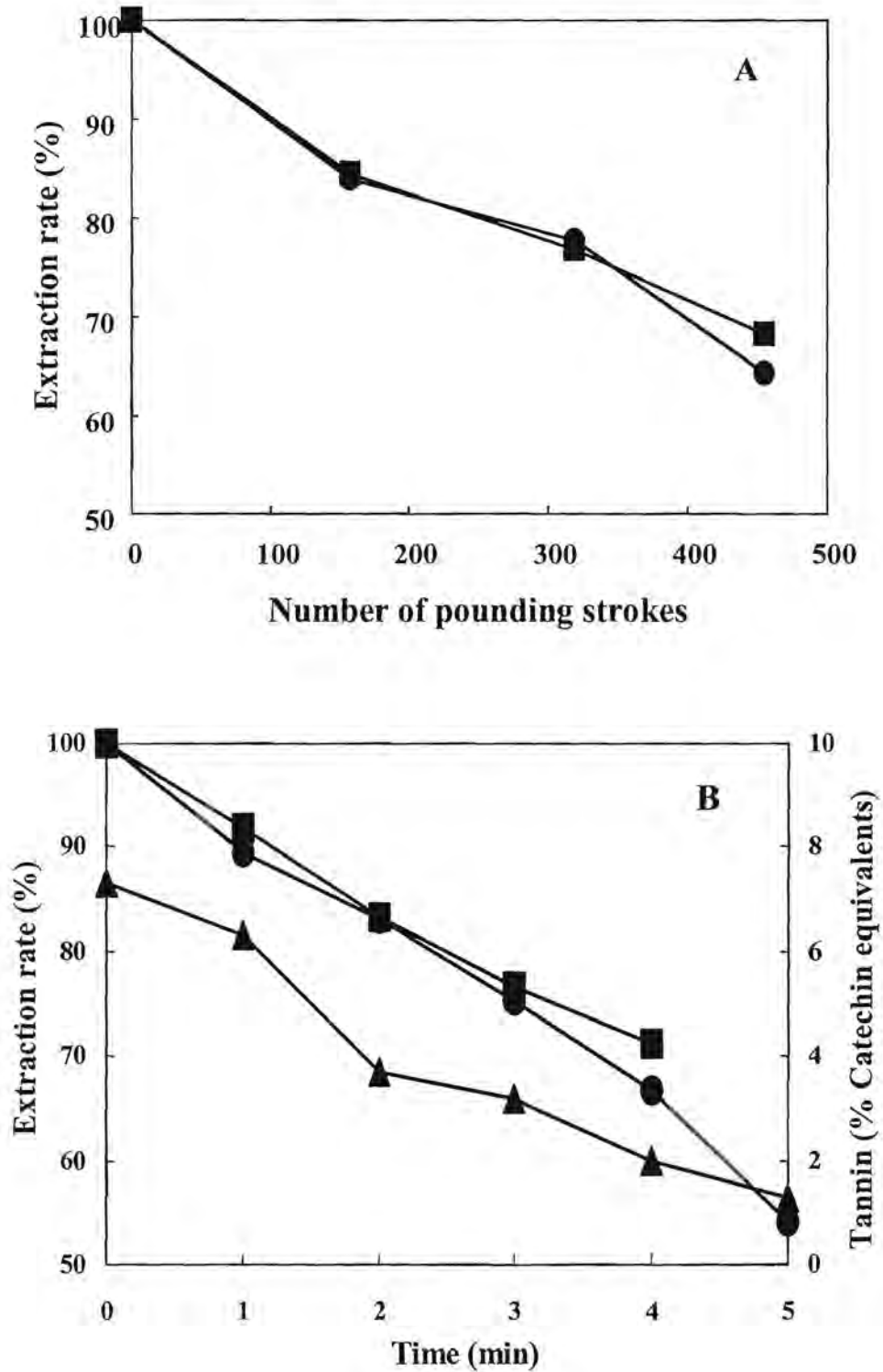




**Figure 3.1.1.** Micrographs of Seredo (tannin-containing) and IS 2284 (tannin-free) sorghum grains. **A**, Seredo endosperm; **B**, IS 2284 endosperm; **C**, Seredo pericarp; **D**, IS 2284 pericarp. P = pericarp; VE = vitreous endosperm; FE = floury endosperm; G = germ.

rates to Seredo was presumably due to the better milling characteristics of IS 2284 as a result of it being harder and the fact that it was tannin-free.

In contrast to hand pounding, mechanical abrasion of Seredo resulted in *injera* of good quality at an extraction rate of 66.8% (Table 3.1.2). This is presumably because at this extraction rate the tannin content of the flour had been reduced to only 2% (Fig 3.1.2B). Thus, mechanical abrasion, which works by polishing the grains against the abrasive surface and each other (Munck et al 1982), removed the tannin-containing testa layer more effectively compared to hand pounding. In hand pounding the pestle causes a



**Figure 3.1.2.** Effect of decortication on sorghum extraction rate. **A**, Hand pounding; **B**, Mechanical abrasion with TADD. Circles = Seredo (tannin-containing); Squares = IS2284 (tannin-free); Triangles = Tannin content of Seredo.

mechanical shock, generating strong interactive forces between the grains and between the grain and the equipment, which is said to result in the breaking off of large pieces of “hull” (Munck et al 1982). However, in the case of soft, tannin-containing sorghum grains such as Seredo considerable grain breakage appears to occur, as a result of these strong interactive forces. For the tannin-free red sorghum IS 2284, good quality *injera* was obtained by mechanical abrasion at an extraction rate of 83.3%, which was very much higher than for the tannin-containing Seredo and also higher than for hand pounded IS 2284.

To help explain the improvement in *injera* making quality with decortication, flours from sequentially TADD abraded tannin-containing sorghum (Seredo) were tested for their pasting properties. Whole Seredo flour had the lowest PV, HPV and CPV (Fig 3.1.3). With successive abrasion, PV, HPV and CPV increased markedly. Linear regression analyses of the relationships between extraction rate and PV, HPV and CPV gave r-values of  $-0.94$ ,  $-0.98$  and  $-0.98$ , respectively, indicating that the extent of decortication and these pasting parameters were closely related in an inverse manner. PV in particular indicates the water-holding capacity of starch and can be used as a measure of the resistance of starch granules to swelling (Fortuna et al 2000, Li and Yeh 2001). The relationship between PV and extent of decortication suggests that one or more of the grain components capable of inhibiting starch swelling were progressively removed with the bran.

Proteins and lipids are involved with resistance to starch swelling (Whistler and BeMiller 1999). Reductions in starch swelling and gelatinization rates in the presence of protein in sorghum have been associated with the tendency of endosperm protein to act as a physical barrier to starch swelling (Chandrashekar and Kirleis 1988). It is probably of significance that the highest concentration of endosperm protein in sorghum is in the peripheral endosperm (Shull et al 1990), and much of the lipid is in the germ (Wang et al 1997), both of which would be removed by decortication. Decortication would also enrich the starch content of the decorticated grain through the removal of grain components such as non-starch polysaccharides, proteins, and lipids, which are concentrated predominantly in the outer endosperm and the germ (Wang et al 1997). It is therefore additionally possible that the successive removal of those grain components in the bran may have led to a

**Table 3.1.2.** Sensory panel responses of *injera* prepared from sorghum flours of Seredo and IS 2284 decorticated by hand pounding and with TADD at different extraction rates

Cultivar	Extraction rate (%)	<i>Injera</i> quality attributes						Overall rating
		Top surface color	Bottom surface color	Description of the eyes <sup>a</sup>	Texture	Taste	Aftertaste	
<b>Hand pounding</b>								
Seredo (tannin-containing)	100.0 <sup>b</sup>	Brown	Brown	None (flat <i>injera</i> )	Sticky	Bitter	Bitter	Poor
	84.0	Brown	Brown	None (flat <i>injera</i> )	Sticky	Bitter	Bitter	Poor
	77.6	Brown	Brown	Few, small, scattered	Sticky	Bitter	Bitter	Poor
	64.3	Light brown	Light brown	Few, small, scattered	Sticky	Slightly bitter	Slightly bitter	Poor
IS 2284 (tannin-free)	100.0 <sup>b</sup>	Red	Light brown	Few, large, scattered	Crumbly	Slightly sweet	Slightly sweet	Poor
	84.5	Red	Red	Few, large, scattered	Slightly soft	Slightly sweet	Slightly sweet	Fair
	76.7	Red	Red	Many, small, evenly spread	Slightly soft	Sour	Slightly sour	Good
	68.3	Light red	Light red	Many, small, evenly spread	Soft	Sour	Slightly sour	V. good
<b>Abrasion with a TADD</b>								
Seredo (tannin-containing)	100.0 <sup>b</sup>	Brown	Brown	None (flat <i>injera</i> )	Sticky	Bitter	Bitter	Poor
	89.5	Brown	Brown	None (flat <i>injera</i> )	Sticky	Bitter	Bitter	Poor
	83.0	Brown	Brown	Few, small, scattered	Sticky	Bitter	Slightly bitter	Poor
	75.3	Light brown	Light brown	Few, small, scattered	Sticky	Slightly bitter	Slightly bitter	Poor
	66.8	Red	Red	Few, small, scattered	Sticky	Slightly bitter	Slightly bitter	Fair
	54.0	Red	Red	Many, small, evenly spread	Soft	Slightly sour	Slightly sour	Good
IS 2284 (tannin-free)	100.0 <sup>b</sup>	Light brown	Light brown	Few, large, scattered	Crumbly	Slightly sweet	Slightly sweet	Poor
	91.8	Red	Red	Few, large, scattered	Crumbly	Slightly sweet	Slightly sweet	Fair
	83.3	Red	Red	Many, small, evenly spread	Soft	Sour	Slightly sour	Good
	76.7	Light red	Light red	Many, small, evenly spread	Soft	Sour	Slightly sour	Good
	71.2	Light red	Light red	Many, small, evenly spread	Soft	Slightly sour	Bland	V. good

<sup>a</sup>Eyes (gas cells) refer to the honeycomb-like structure of the top surface of *injera* formed due to escaping gas bubbles during baking. <sup>b</sup>Whole flour (undecorticated).

progressive elimination of their diluting effects on the starch concentration, leading to increasingly higher PV, HPV and CPV values of the paste. Flours containing higher amounts of starches have been associated with generally higher paste viscosities (Whistler and BeMiller 1999).

Also, Parker et al (1989) showed that starch plays a major functional role in the formation of the continuous amorphous structural matrix of *injera* during baking. Further, Subramanian and Jambunathan (1990) reported a positive correlation between sorghum *injera* eye quality and the starch content of the flour.

### 3.1.3.3. Compositing

Sensory responses of *injera* made from composite flours of whole tannin-containing sorghum (Seredo) and tef are presented in Table 3.1.3. As stated, *injera* made from 100% Seredo flour had a brown top and bottom surfaces, was flat (without eyes) and had sticky texture, bitter taste and was rated as poor. In contrast, *injera* from 100% tef flour had a white top and white bottom surface, many small evenly spread eyes, very soft texture, a bland aftertaste and was rated excellent. As mentioned, the quality defects of *injera* from Seredo are probably due to a considerable extent to tannins inhibiting the fermentation process. At a low level of compositing with tef (16.7%), there were only a few small and scattered eyes on the surface of the *injera*. Also, other attributes of *injera* such as color, taste and texture were not improved. This can be attributed to minimal dilution of the tannin in the whole Seredo flour. *Injera* of good quality was produced with a 50:50 blend of whole Seredo and tef flour.

At this level of substitution, all the quality attributes of *injera* (color, eye number and distribution, texture, taste and aftertaste) were improved. It appears that as substantial tannin dilution had taken place, this enabled the yeast and lactic acid bacteria to ferment the dough. Additionally, tef appeared to impart its intrinsic flour quality to positively affect *injera* quality. The improvement in *injera* quality continued as the proportion of tef flour in the composite was increased beyond 50%.

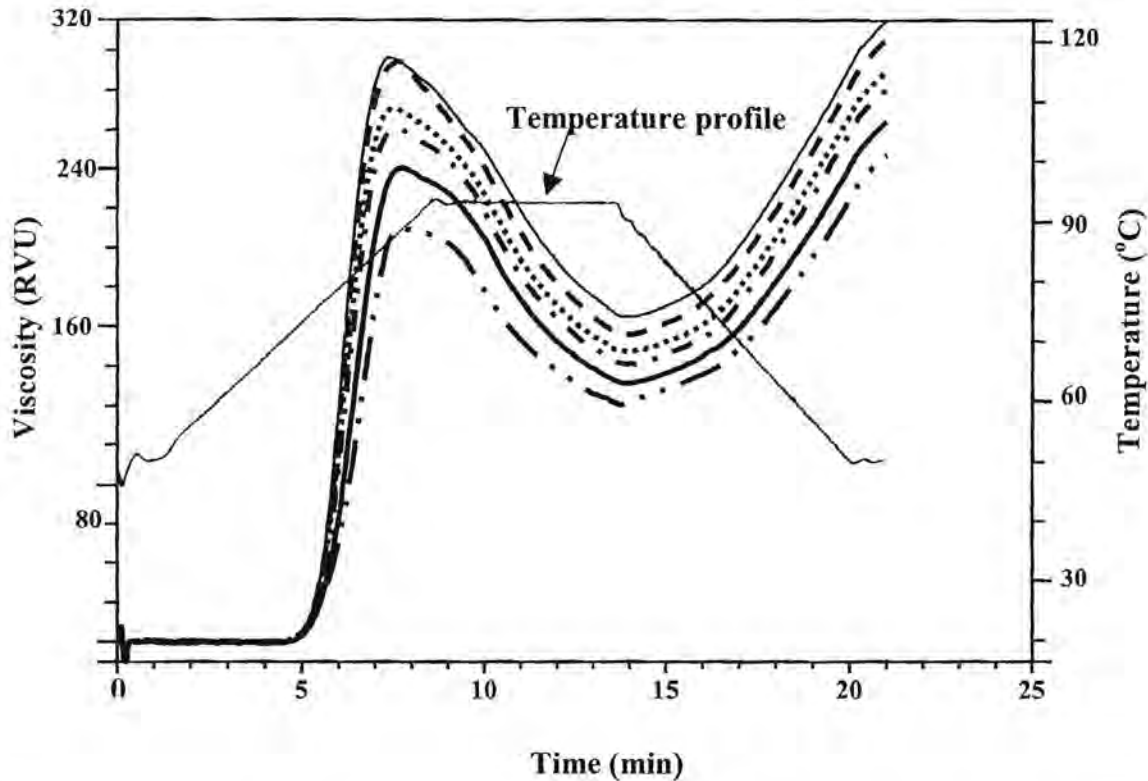
**Table 3.1.3** Sensory panel responses of *injera* prepared from composite flours of whole tannin-containing sorghum (Seredo) and tef flours composited at different proportions

Compositing proportion	<i>Injera</i> quality attributes						Overall rating
	Top surface color	Bottom surface color	Description of the eyes <sup>c</sup>	Texture	Taste	Aftertaste	
100% S <sup>a</sup>	Brown	Brown	Non (flat <i>injera</i> )	Sticky	Bitter	Slightly bitter	Poor
83.3% S + 16.7% T <sup>b</sup>	Brown	Brown	Few, small, scattered	Sticky	Bitter	Slightly bitter	Poor
66.7% S + 33.3% T	Brown	Brown	Few, small, scattered	Sticky	Slightly bitter	Slightly bitter	Fair
50.0% S + 50.0% T	Light brown	Light brown	Many, small, evenly spread	Soft	Slightly sour	Slightly sour	Good
33.3% S + 66.7% T	Light red	Light red	Many, small, evenly spread	Soft	Slightly sour	Slightly sour	V. good
16.7% S + 83.3% T	Light red	Light red	Many, small, evenly spread	Soft	Slightly sour	Bland	V. good
100% T	White	White	Many, small, evenly spread	V. soft	Slightly sour	Bland	Excellent

<sup>a</sup>S = Sorghum (Seredo) whole flour.

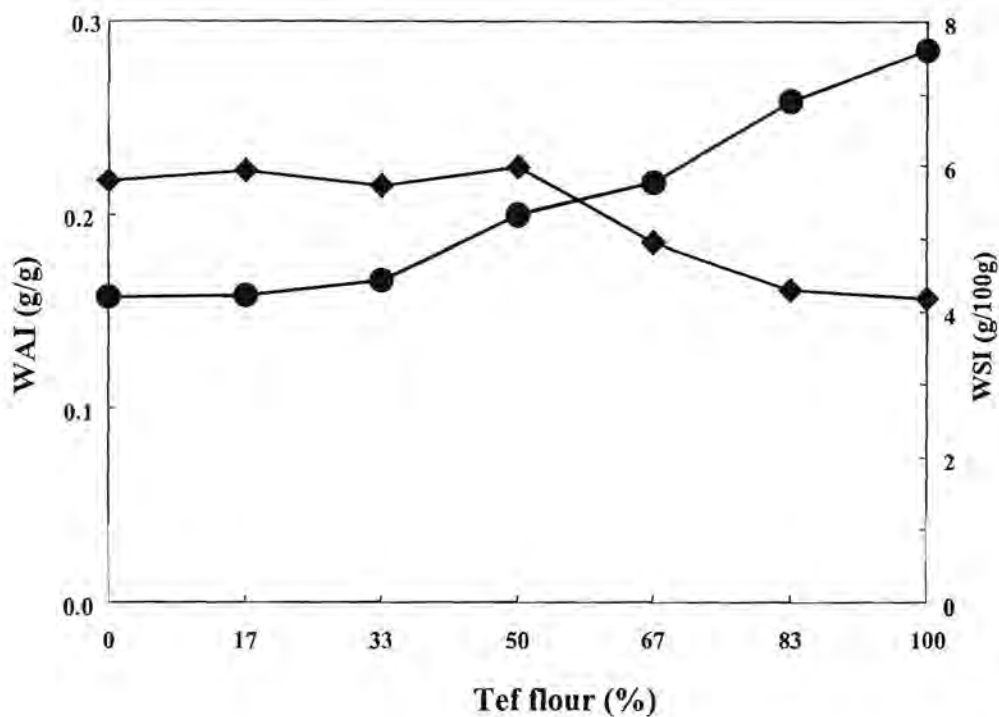
<sup>b</sup>T = Tef whole flour.

<sup>c</sup>Eyes (gas cells) refer to the honeycomb-like structure of the top surface of *injera* formed due to escaping gas bubbles during baking.



**Figure 3.1.3.** Effect of sequential decortication of the tannin sorghum (Seredo) on pasting properties of flours. 54% ——— ; 66.8% - - - ; 75.3% ..... ; 83% - . - ; 89% ——— ; 100% — . . .

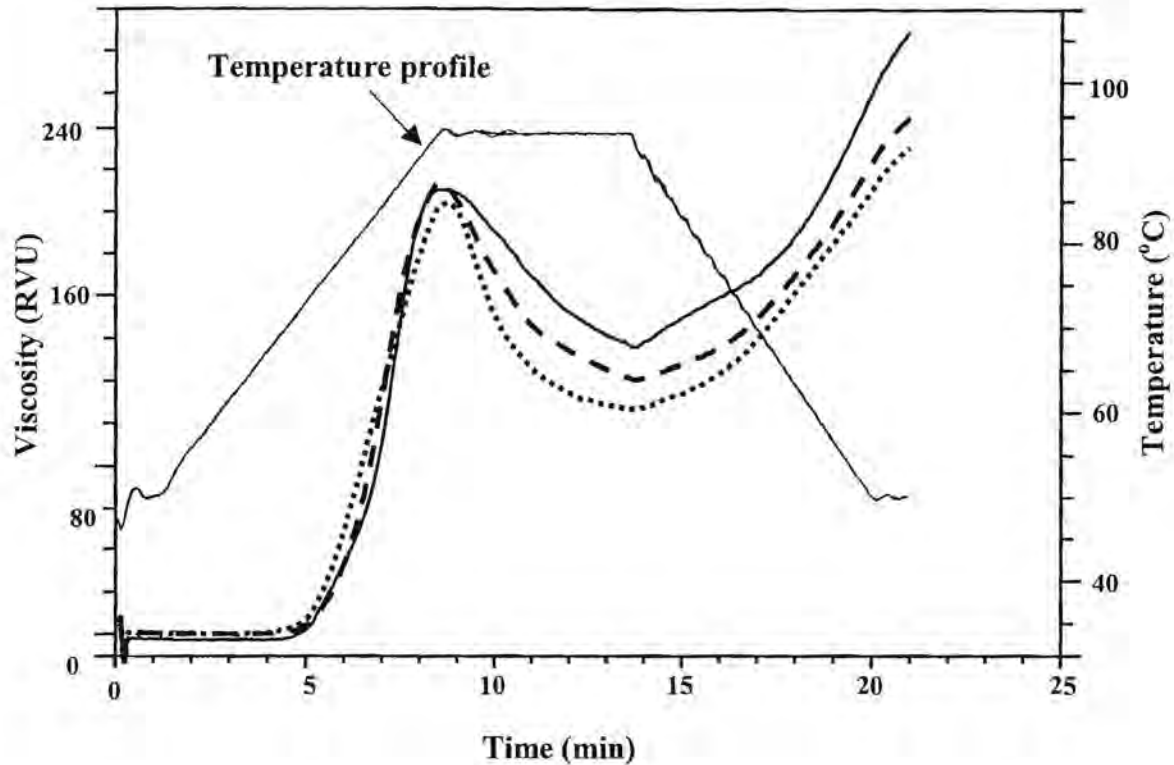
The changes in WAI and WSI when whole flour of Seredo and tef were composited are shown in Figure 3.1.4. As the proportion of tef flour in the composite increased, WSI increased progressively, while WAI declined somewhat. The increase in WSI agrees with the observation that during mixing, tef dough tended to be more sticky compared to sorghum. Water-soluble components in the tef flour could have modified the dough rheology and the texture of *injera* positively. This was manifested by the softer texture of *injera* as the proportion of tef was increased. The decrease in WAI can be attributed to a reduction in damaged starch as the proportion of tef flour increased. Tef starch exists as compound starch granules composed of many tiny granules of 2-6  $\mu\text{m}$  diameter (Umeta and Parker 1996, Bultosa 2002), whereas sorghum starch exists as large single starch granules of about 20  $\mu\text{m}$  diameter (Hoseney 1994). Tef starch granules, because they are so much smaller, are presumably much less prone to damage during milling than sorghum granules and hence would absorb less water.



**Figure 3.1.4.** Changes in water solubility index and water absorption index of composite whole flours of a tannin-containing sorghum (Seredo) and tef composited in different proportions. Circles = WSI; Diamonds = WAI.

Figure 3.1.5 shows that whole Seredo sorghum flour exhibited significantly higher PV, HPV and CPV than whole tef flour. As the proportion of tef flour increased, the PV, HPV and CPV progressively decreased, in contrast to the effect of decorticating Seredo. For the sake of clarity only the 50:50 sorghum:tef curve is shown. The difference in pasting properties of sorghum and tef flours could also be related to inherent morphological differences in their starches. Working with potato, wheat and maize starches Fortuna et al (2000) concluded that the larger starch granules in a population have higher swelling capabilities, while smaller granules have higher resistance to swelling. This may apply to tef starch versus sorghum starch. CPV is related to the ability of the starch paste to form a gel after cooling (Whistler and BeMiller 1999). With high setback, more syneresis is





**Figure 3.1.5.** Effect of compositing the tannin sorghum (Seredo) with tef on pasting properties of the flours. 100% sorghum —; 50% sorghum + 50% tef - - ; 100% tef.....

likely to take place (Newport Scientific 1995). For tef starch, Bultosa et al (2002) reported a low setback viscosity and slow syneresis. This is probably related to the softer texture of tef *injera* compared to sorghum *injera*.

### 3.1.4. Conclusions

Decortication and compositing with tef are both effective ways of improving the *injera* making quality of tannin-containing and tannin-free red sorghum. Decortication improves the color and other quality attributes of *injera* through reduction in the level of non-starch components of the grain. In the case of tannin-containing sorghum decortication also removes some of the tannins, improving *injera* fermentation. Mechanical abrasion is a more effective method compared to hand pounding because acceptable *injera* can be obtained at higher flour extraction levels. Compositing of whole tannin-containing

sorghum flour with tef flour improves *injera* quality primarily by diluting the tannins. Also differences in starch granule characteristics and the higher WSI of tef flour appear to positively influence the quality of *injera*. Compositing seems to be a more useful method of improving sorghum *injera* quality than decortication as it avoids the grain loss associated with decortication.

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### **3.2. Effects of sorghum cultivar on *injera* quality**

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## Abstract

*Injera* is an Ethiopian fermented, leavened flat bread made from cereals, with tef preferred for the best quality *injera*. Because sorghum is less expensive in Ethiopia, there is great interest in improving the quality of sorghum *injera*. Effects of cultivar on *injera* quality were studied using 12 Ethiopian sorghum cultivars of varying kernel characteristics. White tef with good *injera* making quality was included as a reference. *Injera* quality was evaluated using two techniques: descriptive sensory analysis of fresh *injera* and instrumental texture analysis of *injera* stored over a storage period of 48 hr using three point bending rig. Principal Component Analysis (PCA) of sensory data associated fresh *injera* from sorghum cultivars 3443-2-op, 76TI #23, and PGRC/E #69349 of varying endosperm texture, with positive *injera* texture attributes of softness, rollability and fluffiness. Across the two seasons, texture analysis showed *injera* prepared from AW and CR:35:5, both with soft endosperm, required the least force to bend after 48 hr of storage. Bending force was negatively correlated with softness and rollability ( $r = -0.51, -0.52, p < 0.05$ ) and positively with grittiness ( $r = 0.54, p < 0.01$ ) after 48 hr of storage. Sorghum cultivar has an influence on both *injera* making and keeping qualities.

### 3.2.1. Introduction

*Injera* is a fermented leavened, flat Ethiopian traditional bread made from cereals such as tef and sorghum (Gebrekidan and GebreHiwot 1982). Its surface has essentially evenly spaced gas holes, which make up a honeycomb-like structure formed due to the production and escape of gas during fermentation and baking respectively. The bottom surface of *injera* is smooth and shiny. A good *injera* is soft, fluffy and able to be rolled without cracking. It should retain these textural properties after 2 to 3 days of storage, which is traditionally done in a straw basket. A slight sourness is a characteristic taste of *injera*. Because *injera* is a leavened bread made from non-gluten containing flour, it has great potential for commercial production internationally.

*Injera* prepared from flour of tef [*Eragrostis tef* (Zucc.) Trotter], a tiny, millet like grain, is the most preferred. The annual production of tef in Ethiopia is about 1.32 million metric tons (Central Statistical Authority 1998). Of note is the fact that tef commands a higher market price than other cereals in Ethiopia (Seyfu 1993). Sorghum (*Sorghum bicolor* (L.) Moench) is the second most preferred cereal for *injera* preparation in Ethiopia (Gebrekidan and GebreHiwot 1982) with an annual grain production of about 1.82 million metric tons (FAO 2003). Preparing *injera* from sorghum has considerable economic benefits over tef, as sorghum commands a much lower price. However, the problem is that sorghum *injera* rapidly becomes firm and friable upon storage.

Gebrekidan and GebreHiwot (1982) reported that sorghum cultivar differences existed for *injera* making quality and staling property. Yetneberk and Adnew (1985) developed a standard procedure for sorghum *injera* preparation and used it to evaluate the *injera* making qualities of different sorghum cultivars obtained from the Ethiopian Sorghum Improvement Program (ESIP). They confirmed the existence of sorghum cultivar differences for *injera* making quality. It has also been found that the use of composite flour of sorghum and tef improved *injera* texture compared to 100% sorghum (Gebrekidan and GebreHiwot 1982, Yetneberk and Haile 1992). Zegeye (1997) conducted a consumer preference sensory test of *injera* from different cereals and reported that sorghum was accepted as a substitute for tef in *injera* preparation.

The above studies indicate that sorghum cultivar does have an influence on *injera* making and keeping qualities. Thus sorghum cultivars with improved *injera* making quality could probably be selected on the basis of positive *injera* quality attributes. Previous researchers (Gebrekidan and GebreHiwot 1982, Yetneberk and Haile 1992) used a trained panel as consumer panel to evaluate sorghum *injera* making qualities. However, the use of descriptive sensory analysis and instrumental textural analysis to quantitatively evaluate *injera* quality has not been reported. Descriptive sensory analysis detects, identifies, describes and quantifies attribute differences between products and gives information on how raw material and process variables affect sensory characteristics (Stone and Sidel 1985).

The objective of the present study was to determine the influence of sorghum cultivar on *injera* making and keeping quality using descriptive sensory analysis and texture analysis with a view to objectively evaluate sorghum cultivars for selection in sorghum breeding. A wide range of Ethiopian lowland sorghums was used and compared with a white tef cultivar of known good *injera* making quality.

### **3.2.2. Materials and methods**

#### **3.2.2.1. Materials**

Twelve sorghum cultivars IS-777, Aligider Wodifereja (AW), PGRC/E #69441, Seredo, CR:35:5, 3443-2-op, SK-82-022, 76TI #23, Gambella 1107, PGRC/E #69349, [(SC-423xCS-3541)-2-1xRS/R-20-8614-2] (SC-423), [(SC-108-3xCS-3541)-19-1xRS/R-20-8614-2] (SC-108) grown in both the 1999 and 2000 growing seasons, at the Melkassa Agricultural Research Center, Nazareth, Ethiopia were used.

These cultivars had different endosperm texture and pericarp color. Figure 3.2.1 shows the variability in color, size and shape of the 12 sorghum cultivars and a white tef. Seven were white tannin-free, one red tannin-free and four tannin types, as indicated by presence of pigmented testa in the latter (Table 3.2.2). The cultivars are adapted for cultivation in the lowland (high temperature, erratic rainfall) areas of Ethiopia. A white tef cultivar DZ-01-196 with excellent *injera* making quality, grown in 1999 and 2000 at





**Figure 3.2.1.** Variations in color, size and shape of the 12 sorghum cultivars and a white tef.

the Debre Zeit Research Center, Ethiopia, was included for comparison. All sorghum grains were decorticated to about 80% extraction rate, using a carborundum cone abrasive rice pearler (MIAG, Braunschweig, Germany) and milled to flour using Falling Number hammer mill 3100 (Huddinge, Sweden) fitted with a 500  $\mu\text{m}$  opening screen. Tef was not decorticated prior to milling.

#### *3.2.2.2. Kernel characterization*

Sorghum grains were characterized visually for pericarp and endosperm color by comparing with color plates (Rooney and Miller 1982). Glume color was determined by examining the inside of the glume after removing the kernel as described by Rooney and Miller (1982). Pericarp thickness was subjectively rated as thin, intermediate and thick, by scraping through the pericarp with a sharp razor blade. Endosperm texture, the relative proportion of vitreous to floury endosperm, was determined by cutting ten kernels, in halves, longitudinally, and evaluating using a rating scale of 1 (vitreous) to 5 (floury), as described by Rooney and Miller (1982). Grain hardness (extraction rate) was measured by using Tangential Abrasive Dehulling Device (TADD) according to Reichert et al (1982), with extraction rate calculated as percent weight recovered (high recovery indicates harder grain) after abrasion of 20 g of grain for 2 min using sand paper of 60 grit

(Norton type R284 metalite) (Norton Abrasives, Worcester, USA). The presence of a pigmented testa was determined using the Chlorox Bleach Test method described by Waniska et al (1992).

### 3.2.2.3. *Injera making procedure*

A flow diagram of the standardized *injera* making procedure is presented in Fig 3.2.2. The procedure involved milling decorticated sorghum or whole tef grain into a flour, preparation of a dough and fermentation of the dough after adding starter culture, a batter from a previous batch (back slopping) and fermenting at room temperature for about 48 hr. The organisms involved in tef dough fermentation are reported to be Gram negative rods, lactic acid bacteria and yeasts growing in succession (Gashe et al 1982). After fermentation, about 25% of the fermented dough was thinned with 30 mL water and cooked in 200 mL boiling water for 1 min. The objective of gelatinization (cooking) was primarily to bring about cohesiveness of the dough and secondly to provide easily fermentable carbohydrate to leaven the *injera*. The gelatinized batter was cooled to about 45 °C at room temperature and added back to the fermenting dough. After thorough mixing, 100 mL of water was added and the batter was fermented at room temperature for 2-3 hr. Additional water (20 mL) was added to fermented tef dough to bring to batter consistency. Adding back the warm gelatinized starch into the fermented dough promotes the growth of mesophilic microorganisms by raising the fermentation temperature to about 30 °C. About 500 g of the fermented batter was poured in a circular manner, on a 50 cm diameter hot clay griddle (*mitad*), covered and baked for about 2 min.

### 3.2.2.4. *Descriptive sensory analysis*

A panel was trained based on the method described by Einstein (1991). The selected panelists were tested for their ability to detect sweet, sour, bitter and salty tastes (Jellinek 1985). The selected panel consisted of 10 people, as recommended by Stone and Sidel (1985). They were females and males, aged between 20 and 35 years who work at Melkassa Agricultural Research Center.

Nineteen *injera* quality descriptors: Whiteness of top surface, whiteness of bottom surface, redness of top surface, redness of bottom surface, shininess of top surface, eye size, eye evenness and distribution, *injera* softness, stickiness, fluffiness, rollability, grittiness in the mouth, sourness, sweetness, bitterness, sour aftertaste, sweet aftertaste and bitter aftertaste were generated and selected by the trained panel. A score sheet was prepared using the selected descriptors. Each attribute was evaluated using a 10 point numerical scale (0-9) anchored on both sides with verbal descriptions such as 0 = not white, 9 = very white, to allow the panel to score the intensity on a framed common scale. Definitions of the 19 descriptors used for scoring *injera* are given in Table 3.2.1.

The actual product evaluations were performed following good sensory practices according to Lawless and Heymann (1999). Rolled pieces of *injera* (3 cm wide) were presented to the panelists on a tray at ambient temperature (about 25 °C) within 2 hr after baking. A glass of drinking water was provided for rinsing between samples. A maximum of five *injera* samples were served at each session.

#### 3.2.2.5. Texture analysis

Texture analysis of *injera* made from tef and cultivars of sorghum grown for two seasons was performed using a TA-XT2 Texture Analyser (Stable Micro Systems, Godalming, UK). After baking, *injera* samples were allowed to cool for about 30 min at room temperature (about 25 °C). Each *injera* was cut into three equal sized pieces. Each piece was placed in a separate polythene bag of storage and the open end of each bag was folded. The *injera* were stored at room temperature in the dark for 1, 24 and 48 hr. For texture analysis, *injera* samples were cut into strips of 9 cm x 4 cm. Five strips per treatment were measured for maximum bending force using a three point bending rig attachment at a cross-head speed of 0.4 mm/sec for a distance of 10 mm.

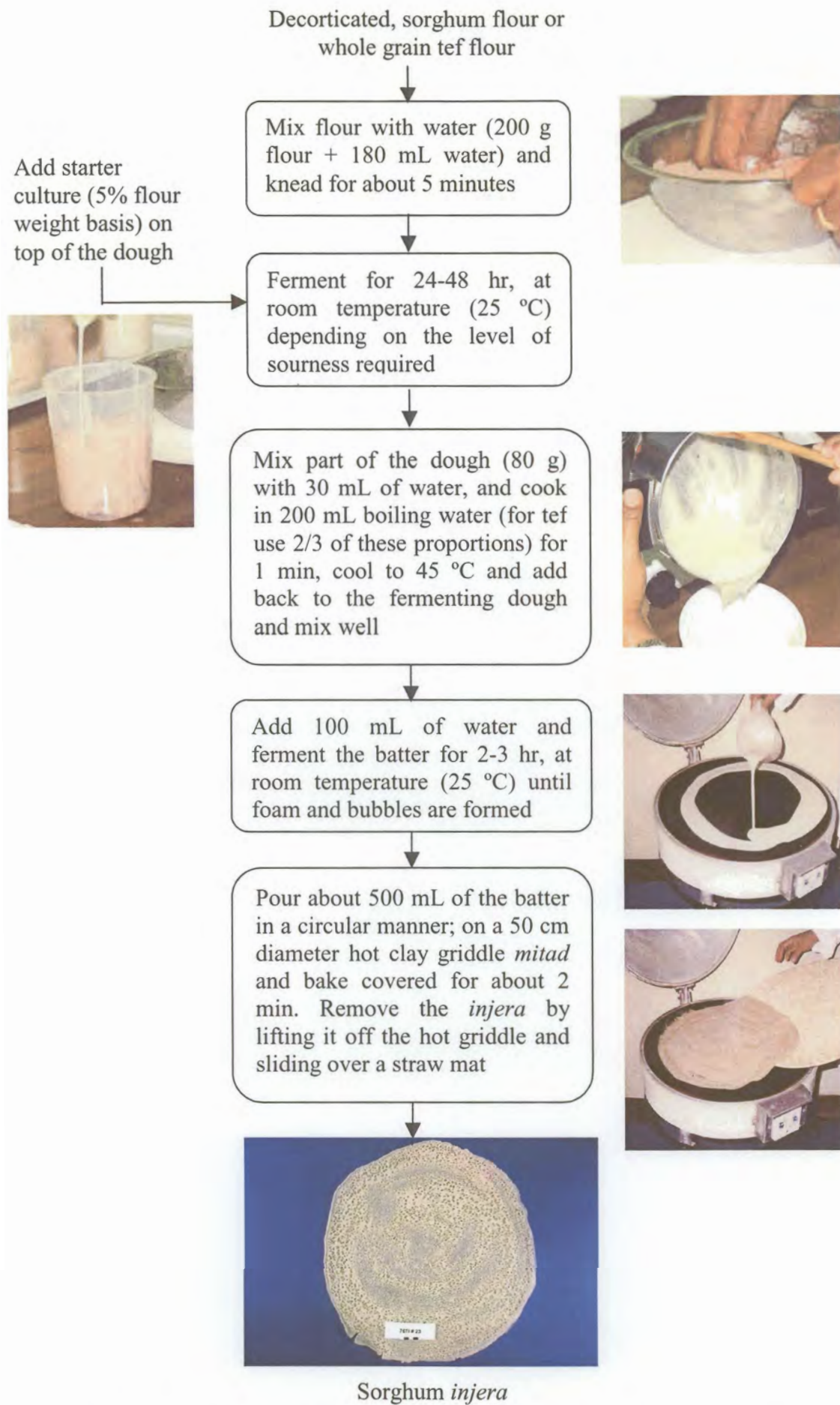


Figure 3.2.2. Flow diagram of standardized *injera* making procedure.

**Table 3.2.1.** Definitions of the 19 descriptors used by the trained sensory panel for scoring sorghum and tef *injera*

<b>Descriptors</b>	<b>Definitions</b>
<b>Appearance</b>	
Whiteness of top surface	Intensity of white color of the top surface of the <i>injera</i> (0 = low and 9 = high)
Whiteness of bottom surface	Intensity of white color of the bottom surface of the <i>injera</i> (0 = low and 9 = high)
Redness of top surface	Intensity of red color of the top surface of the <i>injera</i> (0 = low and 9 = high)
Redness of bottom surface	Intensity of red color of the bottom surface of the <i>injera</i> (0 = low and 9 = high)
Shininess of top surface	Intensity of shininess of the surface of <i>injera</i> (0 = low and 9 = high)
Eye size	Size of the “eyes” (gas holes) on the surface of <i>injera</i> (0 = low and 9 = high)
Eye evenness	Intensity of uniformity of the size of the “eyes” (0 = low and 9 = high)
Eye distribution	Evenness of distribution of the “eye” on the surface of <i>injera</i> (0 = low and 9 = high)
<b>Texture</b>	
Softness	Intensity of softness of <i>injera</i> when lightly pressed between the thumb and forefingers (0 = low and 9 = high)
Stickiness	Intensity of the stickiness of the <i>injera</i> when pressed between thumb and forefingers (0 = low and 9 = high)
Fluffiness	Intensity of fluffiness of the <i>injera</i> when flapped on the fingers (0 = low and 9 = high)
Rollability	Intensity of rollability by rolling the <i>injera</i> strips by hand, samples that are rollable remain rolled (0 = low and 9 = high)
Grittiness in the mouth	Extent of grittiness perceived in the mouth during mastication (0 = low and 9 = high)
<b>Flavor</b>	
Sourness	Intensity of sourness of the <i>injera</i> as perceived in the mouth during mastication (0 = low and 9 = high)
Sweetness	Intensity of sweetness of the <i>injera</i> as perceived in the mouth during mastication (0 = low and 9 = high)
Bitterness	Intensity of bitterness of the <i>injera</i> taste as perceived in the mouth during mastication (0 = low and 9 = high)
Aftertaste sourness	Intensity of residual sourness after swallowing the <i>injera</i> (0 = low and 9 = high)
Aftertaste sweetness	Intensity of residual sweetness after swallowing the <i>injera</i> (0 = low and 9 = high)
Aftertaste bitterness	Intensity of residual bitterness after swallowing the <i>injera</i> (0 = low and 9 = high)

### 3.2.2.6. *Data analysis*

The instrumental texture data were analyzed using multifactor analysis of variance and multiple range analysis. Principal Component Analysis (PCA) of the sensory attributes was conducted using a covariance matrix with cultivar means in rows and attributes in columns.

### 3.2.3. *Results and discussion*

#### 3.2.3.1 *Kernel characterization*

Pericarp colors of the sorghum cultivars ranged from white to red (Table 3.2.2) and were similar for both growing seasons. Pericarp color is genetically controlled (Rooney and Miller 1982). Tef is also known to vary in pericarp color from white to red (National Research Council 1996). White tef is, however, the most preferred grain for *injera* production. The glume color of the sorghum cultivars varied from tan to purple. Glume colors have the tendency to stain the sorghum kernel due to leaching of polyphenolic pigments into the pericarp (Rooney and Miller 1982), which might in turn affect the color of the food product. The pericarp thickness of the sorghum cultivars ranged from thin to thick, while tef pericarp, as observed by Parker et al (1989), was thin and membranous. In the eastern part of Ethiopia, sorghum consumers traditionally use a wooden mortar and pestle to remove the pericarp by hand pounding. An inverse relationship between pericarp thickness and the time required for hand pounding decortication was reported by Scheuring et al (1983). Since sorghum pericarp thickness affects milling performance (Rooney and Miller 1982), cultivars with thicker pericarp are preferred for traditional hand pounding decortication. Endosperm color of all the sorghum cultivars was white.

Four sorghum cultivars, IS-777, PGRC/E #69441, Seredo and CR:35:5 had pigmented testa (Table 3.2.2). The tef cultivar did not have a pigmented testa. The pigments responsible for testa color are polymeric polyphenols known as tannins (Butler 1990). Tannins are known to cause dark color and astringent taste in foods prepared from whole sorghum (Earp et al 1983). As described in Chapter 3.1 *injera* prepared from whole sorghum with pigmented testa had a brown color, “bitter” taste and poor eye quality,

which made it unacceptable to consumers. However, decorticating the sorghum grain with a TADD or compositing the flour with tef at a ratio of 1:1 improved the *injera* quality.

Endosperm texture of two sorghum cultivars, PGRC/E #69349 and SC-108, was relatively vitreous, whereas AW, CR:35:5 and Seredo were essentially floury. The floury endosperm area has loosely packed endosperm cells (Rooney and Miller 1982). For the 1999 growing season grain hardness expressed as % extraction rate varied from 56.5 % (AW) to 84.5% (SC-108). For the 2000 growing season, grain hardness increased and varied from 67.1% (Seredo) to 89.7% (SC-108). It appears that cultivars with a high proportion of floury endosperm generally had lower extraction rate compared to the cultivars with more vitreous endosperm. Lawton and Faubion (1989) also reported that sorghums with softer endosperm had higher rates of loss than did harder sorghums. Sorghum grain hardness is related to the distribution density of protein bodies and matrix in the endosperm (Shull et al 1990). Hard grains had higher milling yields after abrasive dehulling (Mwasaru et al 1988). For sorghum, decortication removes the pericarp and improves the color and quality of *injera* (Yetneberk and Haile 1992). SC-108, a more vitreous sorghum cultivar, had a high extraction rate (89.7%) after abrasive decortication. This suggests that vitreous sorghum cultivars are suitable for mechanical decortication. Because of its small size, tef grain does not lend itself to mechanical decortication and is not decorticated to make *injera*. Since tef *injera* is preferred, the bran of tef does not obviously cause acceptability problems.

Sensory textural attributes of *injera* from sorghum cultivars and tef are shown in Table 3.2.3. Mean softness score of sorghum *injera* ranged from 6.4 (PGRC/E #69441) to 7.7 (AW). Tef *injera* had a higher score of 8.2 for softness. The mean score for stickiness ranged from 1.4 (PGRC/E #69349) to 3.5 (IS-777). Tef *injera* had mean stickiness score of 2.4. The score of sorghum *injera* for fluffiness ranged from 6.5 (SC-108) to 7.5 (AW). Tef *injera* had a higher score of fluffiness 8.1. Sorghum *injera* rollability scores ranged from 6.9 (SC-108) to 7.7 (CR:35:5 and 76TI #23). Tef *injera* had a higher mean score of rollability 8.3. The mean scores for sorghum *injera* grittiness ranged from 1.9 (CR:35:5) to 3.1 (3443-2-op). Tef *injera* had a lower score of 0.8 of grittiness.

**Table 3.2.2.** Pericarp and glume color, pericarp thickness, endosperm color, pigmented testa, endosperm texture and hardness of sorghums and a tef cultivar from the 1999 and 2000 growing seasons

Growing season	Physical variables	Sorghum cultivar											Tef		
		IS-777	AW	PGRC/E #69441	Seredo	CR:35:5	3443-2-op	SK-82-022	76TI #23	Gambella 1107	PGRC/E #69349	SC-423	SC-108	DZ-01-196	
Across seasons	Pricarp color	Red	Red	Red	Light Red	White	White	White	White	White	White	White	White	White	
	Glume color	Purple	Purple	Purple	Purple	Tan	Tan	Purple	Tan	Tan	Tan	Tan	Tan	Tan	
	Pericarp thickness	Thick	Thick	Thick	Thick	Thick	Intermediate	Thick	Intermediate	Thick	Intermediate	Thin	Thin	V. thin	
	Endosperm color	White	White	White	White	White	White	White	White	White	White	White	White	White	ND <sup>d</sup>
	Pigmented testa <sup>a</sup>	Yes	No	Yes	Yes	Yes	No	No	No	No	No	No	No	No	No
1999	Endosperm texture <sup>b</sup>	3	5	3	4	4	3	3	3	3	2	4	2	ND	
	Hardness (%)	71.2cd <sup>c</sup> ±2.9 <sup>e</sup>	56.5a ±1.0	76.8def ±2.8	66bc ±4.9	62.2ab ±4.4	71.7cd ±7.0	80.3fg ±2.4	73.3de ±3.5	73.5de ±0.9	78.8efg ±3.1	63.8b ±5.0	84.5g ±1.8	ND	
2000	Endosperm texture <sup>b</sup>	4	4	3	4	4	3	3	3	2	2	3	2	ND	
	Hardness (%)	73.6bc ±3.0	76.2bcd ±5.1	83.0gh ±4.8	67.1a ±2.0	72.0ab ±2.2	78.2efg ±3.6	80.0fg ±4.5	73.3bcd ±3.8	87.2hi ±2.3	86.3hi ±1.0	83.3gh ±1.6	89.7i ±0.6	ND	

<sup>a</sup>Yes = pigmented testa present, No = pigmented testa absent.

<sup>b</sup>Subjectively rated on a 1 to 5 scale, where 1 = vitreous and 5 = floury.

<sup>c</sup>Values followed by the same letter in the same row are not significantly different ( $p > 0.05$ ).

<sup>d</sup>ND = not determined.

<sup>e</sup>Standard deviation of three replicates.



**Table 3.2.3.** Sensory textural attributes of *injera* from the 12 sorghum cultivars and tef from the 1999 and 2000 growing seasons

Parameter	Season	Sorghum cultivar												Tef
		IS-777	AW	PGRC/E #69441	Seredo	CR:35:5	3443- 2-op	SK-82- 022	76TI #23	Gambella 1107	PGRC/E #69349	SC- 423	SC- 108	DZ-01- 196
Softness	1999	7.2bc <sup>a</sup>	7.8e	7.4cd	7.6de	7.6de	7.4cd	7.0ab	7.1bc	7.7de	6.7a	7.1bc	6.9ab	8.2f
	2000	7.2cde	7.6de	5.5a	7.6de	7.7de	7.0cd	7.4cde	7.3cde	5.9ab	6.8bcd	6.8bcd	6.5abc	8.2e
	Mean <sup>b</sup>	7.2abc	7.7bc	6.4a	7.6abc	7.6bc	7.2abc	7.2abc	7.2abc	6.8ab	6.8ab	7.0ab	6.7ab	8.2c
Stickiness	1999	2.2de	1.8c	1.3b	1.9cd	1.8c	1.0a	2.0cd	1.5b	1.0a	0.8a	1.0a	0.8a	2.4e
	2000	4.9e	3.4bc	1.9a	4.0cd	2.5a	2.3a	2.7ab	4.8de	2.5a	2.1a	2.5a	2.7ab	2.5a
	Mean	3.5cd	2.6bc	1.6a	3.0c	2.2bc	1.6a	2.3bc	3.1c	1.7ab	1.4a	1.8ab	1.7ab	2.4bc
Fluffiness	1999	7.1abc	7.7bcd	7.5abcd	7.0ab	7.3abc	7.6abcd	6.9a	7.2abc	7.8cd	7.0a	7.3abc	7.0ab	8.0d
	2000	6.9cd	7.3d	5.8a	6.6bc	7.3d	7.0cd	7.0cd	7.1d	6.2ab	6.8cd	7.0cd	5.9a	8.2e
	Mean	7.0ab	7.5ab	6.6a	6.8a	7.3ab	7.3ab	7.0ab	7.1ab	7.0ab	6.9a	7.1ab	6.5a	8.1b
Rollability	1999	7.5a	7.8ab	8.0bc	7.9abc	7.7ab	7.8ab	7.6ab	7.7ab	7.8abc	7.7ab	7.7ab	7.5a	8.3c
	2000	7.6cd	7.2abc	6.1a	7.8cd	7.8cd	7.0abc	7.3bcd	7.6cd	6.8abc	7.3bcd	6.9abc	6.3ab	8.3d
	Mean	7.6ab	7.5ab	7.1a	5.3ab	7.7ab	7.4ab	7.4ab	7.7ab	7.3ab	7.5ab	7.3ab	6.9a	8.3b
Grittiness	1999	2.6f	1.5b	2.3de	2.0c	2.5ef	3.1g	2.7f	2.6ef	2.1cd	2.7f	2.6ef	2.4ef	0.7a
	2000	3.2cd	2.7bcd	3.3d	3.0cd	1.3a	3.2cd	2.3bc	1.8ab	3.5d	2.8bcd	2.5bcd	2.8bcd	0.8a
	Mean	2.9bc	2.1bc	2.8bc	2.5bc	1.9ab	3.1c	2.5bc	2.2bc	2.8bc	2.8bc	2.5bc	2.6bc	0.8a

<sup>a</sup>Values followed by the same letter in the same row are not significantly different ( $p > 0.05$ ).

<sup>b</sup>Mean values are mean of the two seasons.

### 3.2.3.2. PCA of sensory attributes

The product of a PCA is a data map illustrating the various relationships among multiple dependent variables and samples (Lawless and Heymann 1999). Principal components (PCs) are orthogonal directions of maximum variance in the original data. The first two principal components described 70% of the total variance in sensory attributes of *injera* made from sorghum and tef from the 1999 growing season (Fig 3.2.3). The abscissa, which corresponds to the first principal component (Destefanis et al 2000), explained 44% of the total variance. Cultivars that were clustered together on the left were the white cultivars, PGRC/E #69349, 3443-2-op, SC-423, Gambella 1107, SC-108 and 76TI #23 (Fig 3.2.3A). The attributes that described *injera* from these white pericarp sorghum cultivars were grittiness, even eye size and distribution, white top and bottom surfaces, sweet and sweet aftertaste (Fig 3.2.3B). On the right plane of the first principal component were the tannin-containing sorghum cultivars (IS-777, PGRC/E #69441, CR:35:5 and Seredo), which were generally characterized as having bitter taste and aftertaste, red top and bottom surfaces, sour taste and aftertaste and stickiness. SK-82-022, a white sorghum cultivar with purple glume color, gave a faint red colored *injera*, possibly imparted by leaching of the glume color through the pericarp into the peripheral endosperm.

The ordinate of the PCA corresponds to the second principal component (Destefanis et al 2000). The second principal component of the 1999 season explained an additional 26% of the total variance (Fig 3.2.3). PC 2 separated cultivars mainly on the grounds of *injera* texture characteristics and appearance of eyes. Cultivars in the upper part of the plot were associated with *injera* of more gritty texture but evenly distributed larger eyes compared to the lower part of the plot (Fig 3.2.3A). Concerning the bottom part of the plot, AW, a red pericarp sorghum cultivar with floury endosperm was associated with soft, and rollable *injera*, while *injera* from tef was characterized by a fluffy texture with a more shiny top surface.

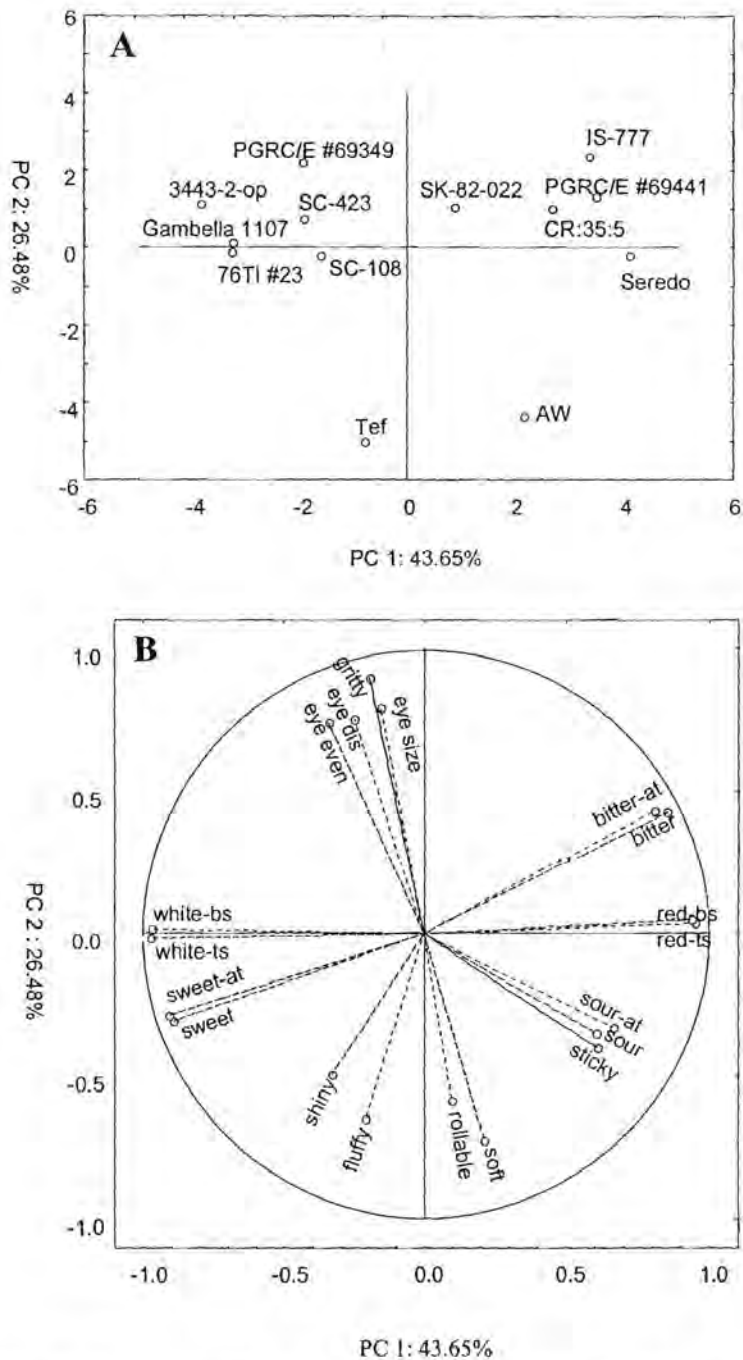
For the 2000 growing season, the first two principal components described less of the total variance (59%) (Fig 3.2.4), compared to the 1999 growing season (70%) (Fig 3.2.3). The third principal component accounted for an additional 15% (Fig 3.2.5), so that 75%

of the total variance in the 2000 data could be explained (Figs 3.2.4 and 3.2.5). The first principal component which described 40% of the variance of the 2000 season, showed a similar trend to the 1999 data. *Injera* from the white pericarp sorghums and tef were characterized by being sweeter and whiter, while *injeras* from the tannin sorghums were characterized by more bitter taste with red top and bottom surfaces (Fig 3.2.4A). *Injera* from the red tannin free sorghum (AW) produced red *injera* and the white sorghum cultivar with purple glume color SK-82-022 produced a faint red colored *injera*, as noted for the previous season.

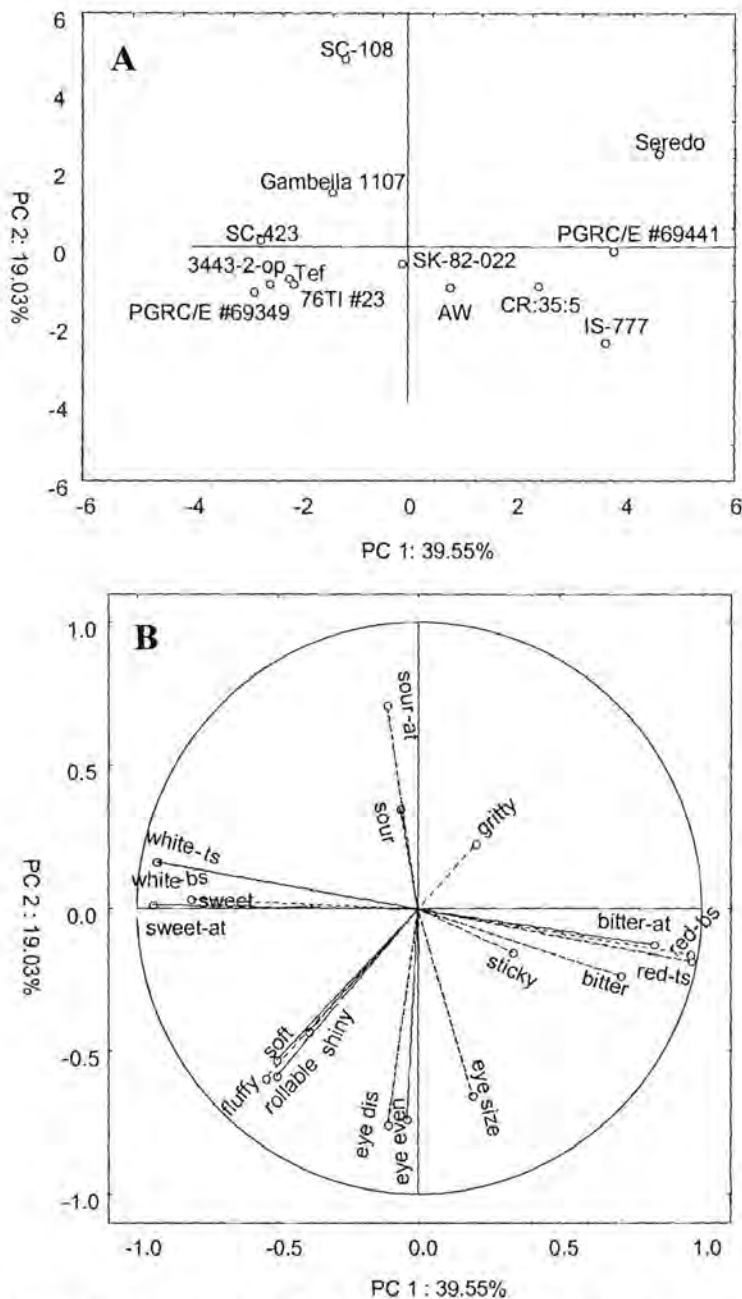
In the upper plot of the second principal component of the 2000 season, a white pericarp-containing sorghum cultivar, SC-108 was associated with *injera* of a more sour aftertaste compared to other sorghum cultivars. This may have been due to the lactic acid fermentation of this cultivar being rapid. In the lower part of the plot, tef was grouped among the white sorghum cultivars (76TI #23, 3443-2-op and PGRC/E #69349). The *injera* attributes associated with these cultivars were sweet taste, positive textural attributes (fluffy, rollable and soft), and shiny surface with more evenly distributed eyes. The endosperm texture of these cultivars varied from intermediate to relatively vitreous.

The third principal component of the 2000 season further explained differences in the texture (softness, rollability, fluffiness and grittiness), eye appearance and sweetness of the *injera* (Fig 3.2.5). White pericarp-containing sorghum cultivars (PGRC/E #69349, 3443-op, Gambella 1107, SC-423 and 76TI #23) were associated with evenly distributed eyes, sweet taste and white top and bottom surfaces. Tef and a white sorghum cultivar with relatively vitreous endosperm (SC-108) produced soft, rollable and fluffy *injera*. Although seasonal variation in cultivar association was observed, both seasons showed similar trends in terms of grouping cultivars with similar *injera* attributes as perceived by the trained sensory panel.

According to Destefanis et al (2000), variables close together in the loading plot are positively correlated while variables lying opposite to each other are negatively correlated. As expected, whiteness and redness of top and bottom surfaces of the *injera* were negatively correlated (Figs 3.2.3B, 3.2.4B and 3.2.5B). *Injera* softness was very



**Figure. 3.2.3.** Principal component analysis of *injera* from 12 sorghums and a tef cultivar grown in 1999. Plot of the first two principal component scores of the cultivars **A**. Plot of the first two principal component loading vectors of sensory attributes **B**. White-ts = whiteness of top surface; white-bs = whiteness of bottom surface; red-ts = redness of top surface; red-bs = redness of bottom surface; eye dis = eye distribution; eye even = eye evenness; sour-at = sour aftertaste; sweet-at = sweet aftertaste; bitter-at = bitter aftertaste.



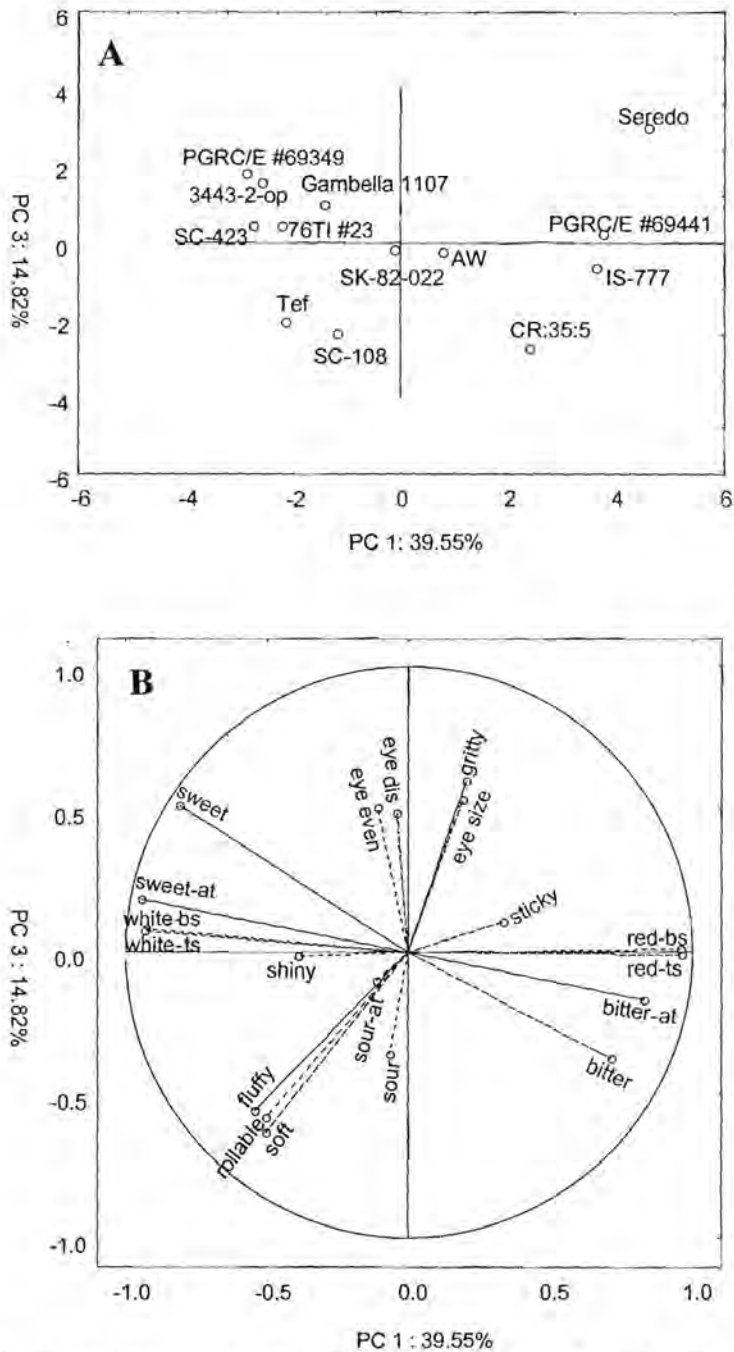
**Figure 3.2.4.** Principal component analysis of *injera* from 12 sorghums and a tef cultivar grown in 2000. Plot of the first two principal component scores of the cultivars **A**. Plot of the first two principal component loading vectors of sensory attributes **B**. White-ts = whiteness of top surface; white-bs = whiteness of bottom surface; red-ts = redness of top surface; red-bs = redness of bottom surface; eye dis = eye distribution; eye even = eye evenness; sour-at = sour aftertaste; sweet-at = sweet aftertaste; bitter-at = bitter aftertaste.

closely associated with rollability (Figs 3.2.3B, 3.2.4B and 3.2.5B). This agrees with the observed tendency of soft *injera* to roll easily. Both characteristics are considered important eating quality attributes of *injera*. Good *injera* is soft and rollable in order to wrap and hold the sauce (*wot*) during consumption (Gebrekidan and GebreHiwot 1982). *Injera* bitterness and bitter aftertaste were negatively correlated with sweetness and sweet aftertaste (Figs 3.2.3B, 3.2.4B and 3.2.5B). The consumption of tannin-rich foods and beverages is associated with astringency or dryness and roughness felt in the mouth (Bacon and Rhodes 2000). It appears as if this sensation was perceived by the panel as bitter.

### 3.2.3.3. Instrumental texture measurement

The maximum force required to bend fresh *injera* and *injera* stored for 24 and 48 hr are presented in Table 3.2.4. *Injera* from tef required the least force in all cases, while *injera* from the sorghum cultivars increased in the force required to bend over the 48 hr storage, indicating that firming (staling) of sorghum *injera* is time dependent. The example of PGRC/E #69349 (a high staler) is shown in Figure 3.2.6. The force required to bend sorghum *injera* varied between cultivars for both fresh and stored *injera* and across seasons. For the 1999 growing season, fresh *injera* from cultivars PGRC/E #69441, SK-82-02 and CR:35:5 required the least force and were similar to *injera* from tef, which had the most bendable *injera*. Fresh *injera* from cultivars SC-423, Seredo, 76TI #23 and 3443-2-op required the highest force to bend. After a storage period of 48 hr, *injera* from sorghum cultivars AW and SK-82-022 required the least force, whereas 76TI #23 and 3443-2-op required the most. This is illustrated in Figure 3.2.7. The low stalers were both tannin-free but varied in pericarp color and endosperm texture. AW had red pericarp color and floury endosperm, while SK-82-022 had white pericarp color and an intermediate endosperm texture. The high stalers were both white, tannin-free cultivars with intermediate endosperm texture.

For the 2000 growing season, fresh *injera* from sorghum cultivars (76TI #23, SK-82-02, CR:35:5, AW and PGRC/E #69441) required the least force to bend, while SC-423 and PGRC/E #69349 required the most (Table 3.2.4). After two days of storage, CR:35:5 and



**Figure 3.2.5.** Principal component analysis of *injera* from 12 sorghums and a tef cultivar grown in 2000. Plot of the first and third principal component scores of the cultivars **A**. Plot of the first and third principal component loading vectors of sensory attributes **B**. White-ts = whiteness of top surface; white-bs = whiteness of bottom surface; red-ts = redness of top surface; red-bs = redness of bottom surface; eye dis = eye distribution; eye even = eye evenness; sour-at = sour aftertaste; sweet-at = sweet aftertaste; bitter-at = bitter aftertaste.

**Table 3.2.4.** Maximum force (N) required to bend *injera* stored at 25 °C over a period of two days from sorghums and a tef cultivar grown for two seasons

Season	Storage time (hr)	Sorghum cultivar												Tef
		IS-777	AW	PGRC/E #69441	Seredo	CR:35:5	3443-2-op	SK-82-022	76TI #23	Gambella 1107	PGRC/E #69349	SC-423	SC-108	DZ-01-196
1999	1	0.18bcd <sup>a</sup>	0.19bcd	0.14ab	0.26gh	0.17abcd	0.24efgh	0.16abc	0.24fgh	0.21def	0.19cde	0.27h	0.21defg	0.12a
		± 0.03 <sup>b</sup>	± 0.06	± 0.03	± 0.05	± 0.06	± 0.05	± 0.15	± 0.08	± 0.05	± 0.08	± 0.08	± 0.08	± 0.01
	24	0.25def	0.18bc	0.23cde	0.21bcd	0.27ef	0.29f	0.15ab	0.36g	0.27ef	0.25def	0.29f	0.20bcd	0.11a
		± 0.05	± 0.03	± 0.06	± 0.06	± 0.03	± 0.05	± 0.02	± 0.17	± 0.06	± 0.04	± 0.05	± 0.03	± 0.02
	48	0.26bcd	0.19ab	0.26cd	0.33d	0.33d	0.47e	0.22abc	0.42e	0.27cd	0.32d	0.32d	0.27cd	0.15a
		± 0.01	± 0.06	± 0.07	± 0.1	± 0.06	± 0.19	± 0.07	± 0.09	± 0.07	± 0.09	± 0.03	± 0.01	± 0.06
2000	1	0.21bc	0.19b	0.20b	0.24cd	0.19b	0.28df	0.19b	0.18ab	0.29f	0.37g	0.43h	0.26de	0.14a
		± 0.03	± 0.04	± 0.06	± 0.03	± 0.05	± 0.06	± 0.03	± 0.03	± 0.04	± 0.05	± 0.09	± 0.05	± 0.05
	24	0.31de	0.23bc	0.37f	0.26cd	0.19b	0.47g	0.26cd	0.36ef	0.32ef	0.54h	0.70i	0.33ef	0.13a
		± 0.06	± 0.04	± 0.06	± 0.04	± 0.06	± 0.11	± 0.03	± 0.09	± 0.08	± 0.02	± 0.03	± 0.05	± 0.05
	48	0.42ef	0.28c	0.52g	0.33cd	0.22b	0.58h	0.33cd	0.45f	0.47fg	0.62hi	0.66i	0.37de	0.15a
		± 0.05	± 0.04	± 0.09	± 0.07	± 0.03	± 0.11	± 0.05	± 0.08	± 0.06	± 0.07	± 0.07	± 0.04	± 0.03

<sup>a</sup>Values followed by the same letter in the same row are not significantly different ( $p > 0.05$ ).

<sup>b</sup>Standard deviation of two *injera* baked on separate days (5 determinations per *injera*).

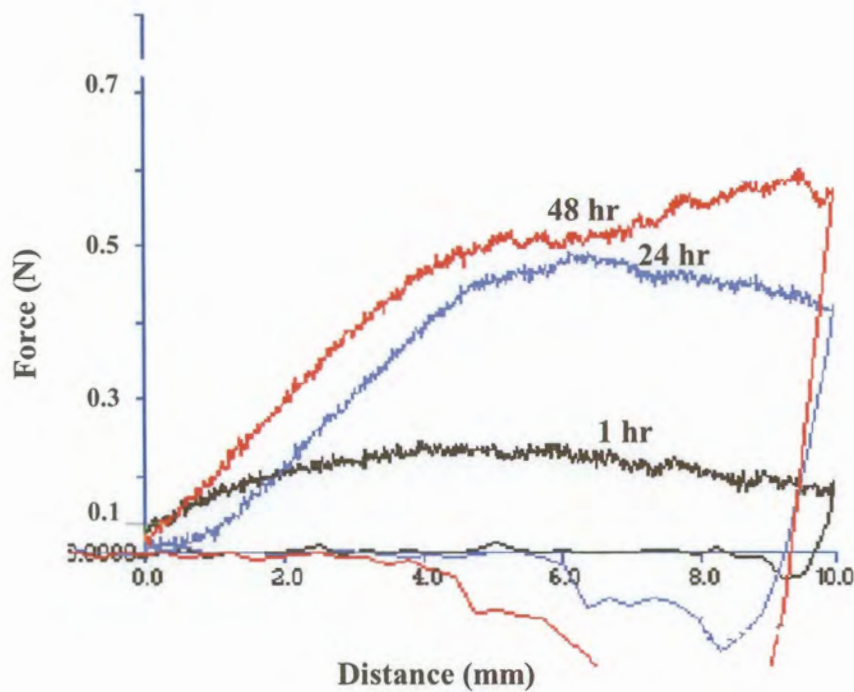


AW required the least force, whereas SC-423 and PGRC/E #69349 required the most. CR:35:5 was a tannin-containing sorghum with floury endosperm. Both the high stalers had white pericarp color with intermediate endosperm texture and were tannin-free. When sorghum endosperm texture data expressed as corneousness (vitreousness) (Murty et al 1982) was related to the shelf life of *injera* made from these cultivars reported by Gebrekidan and GebreHiwot (1982), there was no consistent trend between endosperm texture and staling property. This agrees with the present finding.

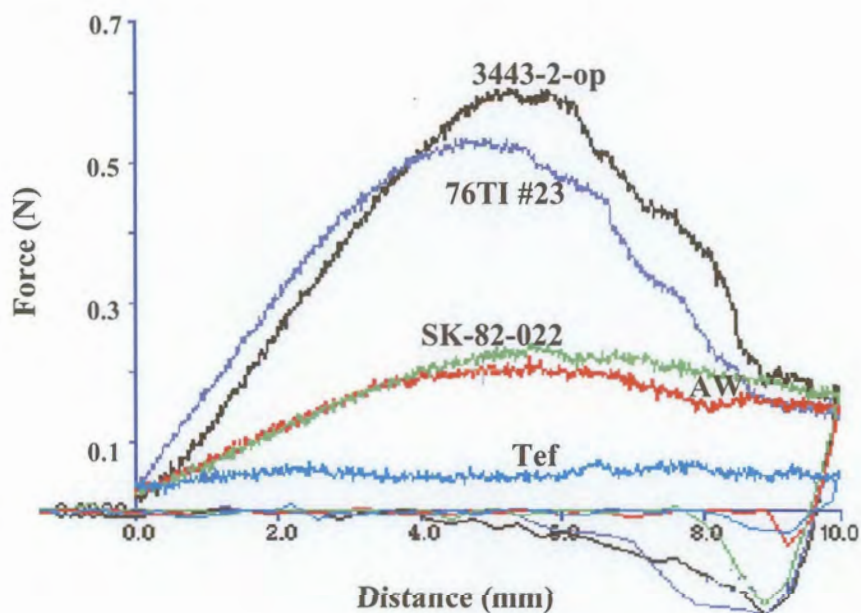
When the means of *injera* sensory textural scores and instrumental texture measurements across seasons for *injera* stored for 48 hr were correlated, bending force was found to be negatively correlated with softness and rollability ( $r = -0.51, -0.52, p < 0.05$ ) and positively with grittiness ( $r = 0.54, p < 0.05$ ). This relationship indicates that as would be expected soft *injera* requires less force to bend. This was clearly demonstrated by the fact that tef *injera*, which is known to be soft, had the lowest bending force throughout the 48 hr of storage. Conversely, *injera* perceived as being gritty (a negative attribute) by the panel required the most force to bend. These relationships show that bending force could be used as an indication of the quality of fresh and stored *injera*.

Across seasons, cultivars AW and CR:35:5 staled least, while cultivars PGRC/E #69349, SC-423 and 3443-2-op staled most. The low stalers had floury endosperm. The high stalers were white with intermediate endosperm and were tannin-free sorghums. *Injera* from AW, one of the low stalers across seasons, was also noted by the sensory panel as being soft and rollable (positive attributes) for the 1999 growing season (Fig 3.2.3). Conversely, for the same season, *injera* from PGRC/E #69349, one of the high stalers across seasons, was perceived as gritty by the sensory panel (Fig 3.2.3).

In the course of the storage trial, it was noted that for both seasons after a storage period of only one day, *injera* from PGRC/E #69349, SC-423, 76TI #23, Gambella 1107, and 3443-2-op, all cultivars giving *injera* with relatively high staling, had a moist bottom surface. Water was probably released (syneresis) out of the *injera* matrix due to



**Figure 3.2.6.** Effect of storage time (1, 24, 48 hr) on maximum force required to bend sorghum *injera* from PGRC/E #69349 (high staler) from the 1999 growing season.



**Figure 3.2.7.** Effect of cultivar on maximum force required to bend *injera* stored for 48 hr from the 1999 growing season.

re-association (retrogradation) of starch components, causing increased firmness, as reported by Lineback and Rasper (1988) with regards to rigidity of wheat bread crumb. Staling is due, at least in part, to the gradual transition of amorphous starch to a partially crystalline, retrograded state (Whistler and BeMiller 1999). Martin and Hosney (1991) suggested wheat bread firming as also being a result of cross-links between starch granule remnants and protein fibrils. During the baking of *injera*, starch granules completely gelatinize and fuse into a continuous amorphous matrix (Parker et al 1989). This amorphous matrix probably transforms to a retrograded state upon storage.

#### **3.2.4. Conclusions**

Sorghum cultivar affects *injera* making quality. AW (floury endosperm), 3443-2-op and 76TI #23 (intermediate) and PGRC/E #69349 (with more vitreous endosperm) were generally associated with soft, rollable and fluffy *injera*, which were positive attributes as perceived by the sensory panel. *Injera* from AW and CR:35:5 (both floury endosperm) required least force to bend after 48 hr of storage. More detailed work on the physico-chemical properties of flours of the 12 sorghum cultivars should be conducted and correlated with *injera* quality. This should lead to identification of specific flour quality parameter(s) that are related to good sorghum *injera* quality. The range in quality of fresh and stored sorghum *injera* seems quite substantial, and indicates that sorghum can be bred for better *injera* making quality.

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The Agricultural Research and Training Project (ARTP) of the Ethiopian Agricultural Research Organization (EARO) supported this research. Senayit Yetneberk gratefully acknowledges the partial financial assistance from the International Sorghum and Millet (INTSORMIL) Collaborative Research Support Program.

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### **3.3. Grain and flour quality of Ethiopian sorghums in respect of their *injera* making potential**

## Abstract

*Injera* is an Ethiopian fermented leavened flat bread made from cereals, with tef preferred for the best quality *injera*. Grain and flour of 12 Ethiopian sorghums and a white tef cultivar grown for two seasons were evaluated in terms of their physico-chemical properties. These were related to the sensory attributes of *injera*. Variability in physico-chemical characteristics among sorghum cultivars and between sorghum and tef were evident. Linear regression correlation analysis between the physico-chemical parameters and sensory attributes of *injera* showed that grain hardness, measured by the Tangential Abrasive Dehulling Device was significantly negatively correlated with the positive *injera* textural attributes of softness and rollability ( $r = -0.70$ ,  $r = -0.66$ ,  $p < 0.001$ ). Endosperm vitreousness, measured by image analysis, was negatively correlated with *injera* softness ( $r = -0.59$ ,  $p < 0.01$ ). Endosperm texture, visually evaluated, was positively correlated with *injera* softness ( $r = 0.59$ ,  $p < 0.01$ ). Water solubility index (WSI) of the flour was positively correlated with *injera* fluffiness ( $r = 0.48$ ,  $p < 0.05$ ), probably due to its influence on the dough rheology. Generally, sorghum cultivars with soft endosperm and higher WSI appeared to produce soft and rollable *injera*. The above grain and flour parameters have potential to be used as indirect indices in predicting the quality of sorghum *injera* in the Ethiopian Sorghum Improvement Program. However, additional research is needed to test larger number of genotypes from multi-location trials to further evaluate their effects on *injera* quality attributes.



### 3.3.1. Introduction

*Injera* is an Ethiopian fermented, leavened flat bread made from cereals, with tef preferred for the best quality *injera* (Gebrekidan and GebreHiwot 1982). Because the major component of *injera* is the flour, the various chemical and other functional components of the flour are potentially important with regard to determining the *injera* making potential of sorghum cultivars. Starch, protein and lipids are the three major components in cereal based food products and the interactions among them in a food system are of importance to functionality and quality (Zhang and Hamaker 2003). During the baking of *injera*, starch is completely gelatinized to form a steam-leavened, spongy matrix, in which fragments of bran, embryo, microorganisms and organelles are embedded (Parker et al 1989). Subramanian and Jambunathan (1990) related panelist responses of sorghum *injera* with grain properties and reported a positive correlation between *injera* texture and starch total setback ( $r = 0.62$ ) and a negative correlation ( $r = -0.63$ ) between “eye quality” (honeycomb structure of the *injera* surface) and protein content.

However, our understanding of the effects of sorghum grain characteristics and flour components to *injera* making quality is very limited. Sorghum genotypes have mainly been evaluated on the basis of the sensory attributes of *injera* without relating it to the grain characteristics and flour components (Gebrekidan and GebreHiwot 1982, Yetneberk and Haile 1992). This approach lacks a deep scientific basis for selecting good *injera* making cultivars and is unable to complement the breeding effort in the Ethiopian Sorghum Improvement Program (ESIP). The influence of cultivars on *injera* making quality has been reported (Gebrekidan and GebreHiwot 1982, Subramanian and Jambunathan 1990), and has been confirmed (Chapter 3.2).

The objectives of this investigation were to determine the physico-chemical properties of sorghums with varying endosperm texture and relate them to the sensory attributes of *injera* set out in Chapter 3.2, in order to identify the specific grain or flour quality parameter(s) to be used as indirect indices in predicting the quality of sorghum *injera* in the ESIP.

### **3.3.2. Materials and methods**

#### *3.3.2.1. Materials*

Twelve sorghum cultivars grown in the 1999 and 2000 seasons at the Melkassa Agricultural Research Center, Ethiopia were used. They were of different endosperm textures and grain colors. A white tef cultivar was included for comparison (see Chapter 3.2).

#### *3.3.2.2. Kernel characterization*

Characterization of kernel and endosperm color, endosperm texture (vitreous or floury endosperm), grain hardness and presence of a pigmented testa were determined by using methods described in chapter 3.2.

Vitreousness of the kernel endosperm was measured by image analysis. Kernels were adjusted to 12 % moisture and allowed to equilibrate for 24 hr, then halved longitudinally using a sharp razor blade. One half of each kernel was fitted on an aluminium stub with the aid of double-sided adhesive tape. Each half was viewed using a stereomicroscope (Nikon, Tokyo, Japan). Images were captured with a digital camera attached to the stereomicroscope. Images were enhanced for contrast using an Adobe Photo Shop 5.5 Program (Adobe Systems, San Jose, USA). The total endosperm and floury endosperm areas were measured using the UTHSCSA, Image Tool Software, version 2.0 (University of Texas Health Science Center, San Antonio, USA). Six half grains were viewed for each cultivar. The results were computed and expressed as percent vitreousness.

Thousand kernel weight (TKW) was determined by weighing 1000 randomly selected unbroken kernels. Test weight was determined as hectoliter weight. Grain and whole grain flour colors were measured in L, a, b units using a Hunter Color Quest 45/0 (Hunter Associates, Reston, USA). Water Absorption Index (WAI) and Water Solubility Index (WSI) were determined according to Anderson et al (1969).

### 3.3.2.3. *Chemical characterization*

Moisture, ash and fat contents of whole grain flours were determined according to AACC approved methods (American Association of Cereal Chemists 2000), methods 44-15A, 08-17 and 30-25, respectively. Protein ( $N \times 6.25$ ) was determined by the Dumas combustion method. Lysine was determined by reversed phase high performance liquid chromatography using the PICO.TAG method, according to the procedure of Bidlingmeyer et al (1984). Total starch was determined by the Megazyme Total Starch Assay Procedure (Amyloglucosidase/ $\alpha$ -amylase method), (Megazyme International, Bray, Ireland). Cultivars with a pigmented testa were treated with very dilute (0.04%) formaldehyde solution (Daiber and Taylor 1982) to react with the tannins, to prevent their subsequent inactivation with the enzymes involved in the total starch assay. Apparent starch amylose content of whole ground grain was determined colorimetrically (Faulks and Bailey 1990) based on preferential binding of iodine by amylose.

### 3.3.2.4. *Pasting properties and gel firmness*

Whole meal flour pasting properties were determined using a Rapid Visco Analyser (RVA) Model 3D (Newport Scientific, Warriewood, Australia) as described in Chapter 3.1. From the RVA pasting curves, peak viscosity (PV), hot paste viscosity (HPV), breakdown viscosity (BV) (PV-HPV), cold paste viscosity (CPV), setback viscosity (SBV) (CPV-HPV), pasting temperature (PT) and peak time duration (PTD) were computed.

Gel firmness of wholemeal flour (6 g, 14% moisture basis suspended in 25 ml distilled water) was determined by pasting the suspension in the RVA. The hot paste was immediately placed into plastic dishes (40 mm diam x 10 mm depth); the depth of each dish had been increased by approximately 5 mm by taping clear adhesive tape around its rim. The dishes were covered and rested for 24 hr at ambient temperature (approx. 25 °C) for gelation to take place. After the tape was removed, the surface gel was scraped off with a sharp knife to level to the container's rim (Takashi and Sieb 1988). The texture of the freshly cut gel surface was analyzed using a TA-XT2 Texture Analyser (Stable Micro Systems, Godalming, UK). A standard single-cycle program was used to compress the gel

for a distance of 5 mm at a cross speed of 0.5 mm/s using a 20 mm cylindrical perspex probe with a flat end. Gel firmness (maximum force required for deformation) was computed from the force-time curve. To compare the elasticity of gels from flours of sorghum cultivar 76TI #23 and the tef cultivar, the gels were stored at 4 °C for 24 hr and removed from the dishes for texture analysis. A standard double-cycle program was used to compress the gels for a rupture distance of 50% of the gel height at a crosshead speed of 1 mm/s using 50 mm cylindrical aluminium probe with a flat end.

#### 3.3.2.5. *Statistical analysis*

The various physical and chemical characteristics, pasting properties, gel texture and microstructure measurements were analysed by one-way analysis of variance (ANOVA) and tested for level of significance by Duncan's multiple range test. Means were compared for relationships by Pearson Product-Moment correlation using Statistica for Windows (Statsoft, 1995).

### 3.3.3. *Results and discussion*

#### 3.3.3.1. *Kernel characterization*

Variation in kernel color has been noted in sorghums, ranging from white to red (Table 3.3.1) and is genetically controlled (Rooney and Miller 1982). Tef varies in kernel color (National Research Council 1996). However, white grain tef was selected for comparison because people prefer *injera* from white tef. Pericarp thickness of sorghums varied from thin to thick, while tef pericarp was thin and membranous (Parker et al 1989). Figure 3.3.1 shows micrographs of the 12 sorghum cultivars with varying pericarp thickness. A thin pericarp is manifested by the presence of a single dominant Z allele, whereas the thick pericarp is determined by two recessive alleles, zz (Rooney and Miller 1982, Scheuring et al 1983). In case of sorghum, decortication removes the pericarp and improves the color and quality of *injera* (Gebrekidan and GebreHiwot 1982, Subramanian and Jambunathan 1990, Yetneberk and Haile 1992) (Chapter 3.1).

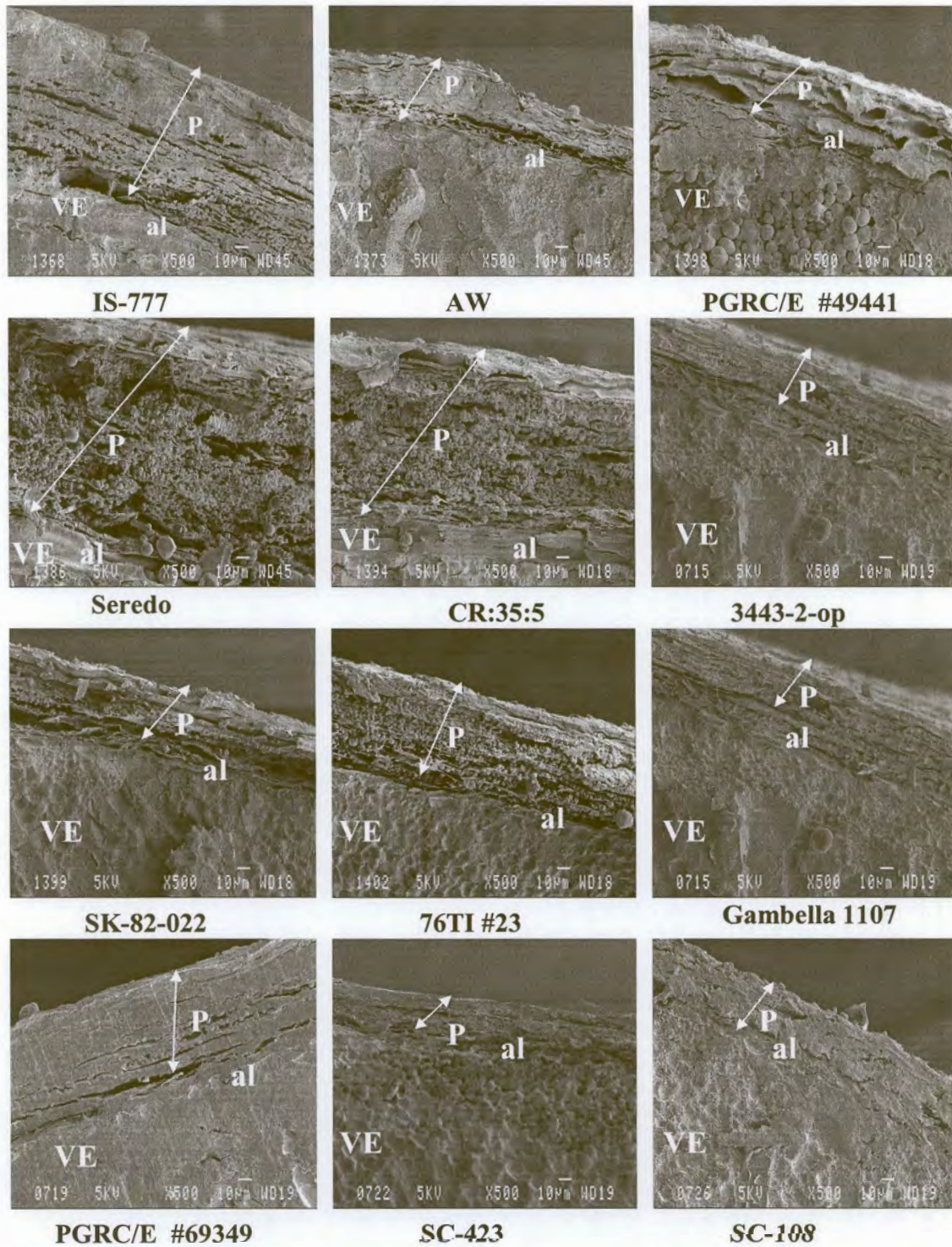
**Table 3.3.1.** Pericarp color and thickness, pigmented testa and endosperm texture of sorghums and tef from the 1999 and 2000 growing seasons (from Chapter 3.2)

Parameter	Season	Sorghum cultivar											Tef	
		IS-777	AW	PGRC/E #69441	Seredo	CR:35:5	3443-2- op	SK-82- 022	76TI #23	Gambella 1107	PGRC/E #69349	SC-423	SC- 108	DZ- 01-196
Pericarp color	Both seasons	Red	Red	Red	Light red	White	White	White	White	White	White	White	White	White
Pericarp thickness	Both seasons	Thick	Thick	Thick	Thick	Thick	Inter mediate	Thick	Inter mediate	Thick	Inter mediate	Thin	Thin	Very thin
Pigmented testa <sup>a</sup>	Both seasons	Yes	No	Yes	Yes	Yes	No	No	No	No	No	No	No	No
Endosperm texture <sup>b</sup> (visual)	1999	3	5	3	4	4	3	3	3	3	2	4	2	ND <sup>c</sup>
	2000	4	4	3	4	4	3	3	3	2	2	3	2	ND

<sup>a</sup>Yes = pigmented testa present, No = pigmented testa absent.

<sup>b</sup>Subjectively rated on a 1 to 5 scale, where 1 = vitreous and 5 = floury.

<sup>c</sup>ND = not determined.



**Figure 3.3.1.** Micrographs of the 12 sorghum cultivars varying in pericarp thickness from the 1999 growing season. P = pericarp; VE = vitreous endosperm; al = aleurone layer.

Visual rating of endosperm texture indicated that three sorghum cultivars PGRC/E #693496, SC-423 and SC-108 possessed essentially vitreous endosperm. Two cultivars, AW and CR:35:5, were essentially floury, indicating the existence of cultivar differences for endosperm texture. Endosperm texture (visual) expectedly highly negatively correlated with test weight, vitreousness (image) and hardness (TADD) ( $r = -0.68$ ,  $r = -0.77$ ,  $r = -0.83$ ,  $p < 0.001$ ), respectively (Table 3.3.2). The inverse relationships indicate that the softer the kernel, the less vitreous and less hard it was. Endosperm texture positively correlated with *injera* softness, a positive textural attribute of *injera* ( $r = 0.59$ ,  $p < 0.01$ ) (Table 3.3.3) indicating that cultivars with soft endosperm tend to make *injera* with soft texture. This agrees with the observation by Rooney et al (1986) that sorghum cultivars with softer endosperm texture produce *injera* with the most desirable texture and keeping quality.

Four sorghum cultivars (IS-777, PGRC/E #69441, Seredo and CR:35:5) had a pigmented testa as revealed by the Chlorox Bleach Test. Tef grain used in this study did not have a pigmented testa. In sorghum the presence or absence of testa is controlled by the complementary  $B_1$  and  $B_2$  genes with the testa present when both the  $B_1$  and  $B_2$  genes are dominant (Rooney and Miller 1982). The pigments responsible for this color are polymeric polyphenols known as tannins (Butler 1990), which cause dark color and astringency in food prepared from whole sorghum with pigmented testa (Bacon and Rhodes 2000). Thus, it is essential to decorticate tannin-containing sorghums to remove the tannin layer or composite with tannin-free tef to act as tannin diluent and improve the *injera* quality (Chapter 3.1).

The L-values (lightness) of the sorghum kernels across seasons ranged from 27.3 (IS-777) to 53.5 (CR:35:5) (Table 3.3.4). Tef had L-value of 53.4. The L-values of sorghum flours ranged from 59.61 (IS-777) to 75.6 (PGRC/E #69349). Tef flour had a mean L-value of 73.7 less light than some of the white sorghum flours. Among the sorghum cultivars, significant differences between cultivars were apparent for grain and flour lightness, redness and yellowness. In sorghum, pericarp color is genetically controlled and can be red, lemon yellow or white (Rooney and Miller 1982). Color is one of the most important considerations in assessment of flour quality in *injera* preparation. Consumers prefer

**Table 3.3.2.** Significant correlations among physico-chemical parameters of the 12 sorghum cultivars across the 1999 and 2000 growing seasons

Parameter	TW	TKW	Protein	Ash	Fat	Starch	Amylose	Vitreousness (image)	Extraction rate (TADD)	WSI	Peak viscosity	Hot paste viscosity	Breakdown viscosity	Final viscosity	Setback viscosity
TW															
TKW	0.42*														
Protein															
Ash	-0.71***														
Fat															
Starch		0.44*													
Amylose		-0.48*													
Vitreousness (image)	0.66***			-0.62**	0.51*		-0.45*								
Extraction rate (TADD)	0.78***			-0.60**				0.65***							
Endosperm texture (visual)	-0.68***			0.62**				-0.77***	-0.83***						
WSI															
WAI		-0.44*						-0.44*		-0.52*					
Peak viscosity					-0.43*	0.51*									
Hot paste viscosity						0.55**		-0.54**			0.86***				
Breakdown viscosity			-0.49*												
Final viscosity		0.57**				0.61**					0.79***	0.84***			
Setback viscosity	0.50*	0.60**				0.54**					0.61**	0.59**		0.93***	
Pasting temperature												0.43*	-0.42*	0.43*	
PTD			-0.69***										0.69***		0.56**
Gel firmness	0.44*		-0.55**	-0.56**									0.57**		0.43*

Level of statistical significance at  $p < 0.05$  (\*),  $p < 0.01$  (\*\*),  $p < 0.001$  (\*\*\*).

TW = Test weight, TKW = Thousand kernel weight, WSI = Water solubility index, WAI = Water absorption index, PTD = Peak time duration.



**Table 3.3.3.** Significant correlations between physico-chemical parameters of the 12 sorghum cultivars and sensory and instrumental textural attributes of *injera* across the 1999 and 2000 growing seasons

<i>Injera</i> attribute	Parameter											
	Test weight	Ash	Starch	Vitreousness (image)	Extraction rate (TADD)	Endosperm texture (visual)	Water solubility index	Water absorption index	Peak viscosity	Hot paste viscosity	Cold paste viscosity	Setback viscosity
Shininess	-	-	-	-	-	-	-	-	-	-	0.46*	0.50*
Eye evenness	-	-	0.52*	-	-	-	-	-	-	-	-	-
Eye distribution	-	-	0.55**	-	-	-	-	-	-	-	-	-
Softness	-0.61**	0.50*	-	-0.59**	-0.70***	0.59**	-	-	-	-	-	-
Stickiness	-	-	-	-	-	-	-	0.45*	0.46*	0.50*	-	-
Fluffiness	-	-	-	-	-	-	0.48*	-0.71***	-	-	-	-
Rollability	-0.54**	0.50*	-	-	-0.66**	-	-	-	-	-	-	-
Grittiness	-	-	-	-	0.42*	-	-	-	-	-	-	-
Bending force (after 1 hr)	-	-	-	-	-	-	-	-	-	-	-	0.43*
Bending force (after 24 hr)	-	-	-	-	-	-	-	-	-	-	-	0.62**
Bending force (after 48 hr)	-	-	-	-	-	-	-	-	-	-	-	0.61**

Level of statistical significance at  $p < 0.05$  (\*),  $p < 0.01$  (\*\*),  $p < 0.001$  (\*\*\*).

- = not significant.

*injera* prepared from white tef or white sorghum cultivars (Gebrekidan and GebreHiwot 1982). Decortication of sorghum increases lightness of the flour possibly through the removal of pigments associated with the bran.

Table 3.3.5 shows TKW, test weight, endosperm texture and hardness (extraction rate) of sorghum and tef. TKW across seasons ranged from 19.8 g for IS-777 to 32.6 g for PGRC/E #69349, indicating cultivar differences for TKW. Grain tef had extremely low TKW (0.33 g) compared to sorghums due to its small size, less than 1.5 mm in length (Parker et al 1989). In general, sorghum cultivars with larger kernel size also had higher kernel weight. TKW positively correlated with test weight ( $r = 0.42$ ,  $p < 0.05$ ) (Table 3.3.2). It seemed that the heavier the kernel the denser it was. However, further verification is needed with respect to this statement due to relatively low  $r$ -value.

Test weight for sorghums across seasons ranged from 70.1 (CR:35:5) to 78.5 kg/hl (PGRC/E #69441). Tef had a higher test weight (86.2 kg/hl) than the sorghum cultivars. This is due to tiny size of tef grain, which compacted in the hectoliter cup with minimum void space between the grains. Among the sorghum cultivars, test weight was highly positively correlated with vitreousness (image analysis) ( $r = 0.66$ ,  $p < 0.001$ ) (Table 3.3.2). This can be ascribed to the tightly packed cells and cellular components of the vitreous region of the endosperm (Shull et al 1990), which probably makes the kernel denser, hence higher test weight. Test weight was highly negatively correlated with ash content ( $r = -0.71$ ,  $p < 0.001$ ). It appears that dense kernels possess lower proportion of bran (rich in minerals) component resulting to lower ash content. Test weight was also negatively correlated with the positive *injera* textural attributes of softness and rollability ( $r = -0.61$ ,  $r = -0.54$ ;  $p < 0.01$ ) (Table 3.3.3). This could relate to the amount and type of protein present in denser kernels to produce *injera* with firmer texture.

Vitreousness (image analysis) of sorghum cultivars ranged from 30.3% for the relatively floury cultivar IS-777, to 55.2% for the most vitreous cultivar SC-108 across the two seasons (Table 3.3.5). Figure 3.3.2 shows section through the 12 sorghum cultivars from the 1999 growing season, illustrating the varying proportions of vitreousness. The endosperm consists of an outer translucent vitreous area and an inner opaque floury area.

The proportions of the two vary from cultivar to cultivar. Such variation within the same season indicates the existence of genetic diversity for endosperm texture. Generally the expression of vitreousness is influenced by the growing environment (Rooney and Miller, 1982), but in any one environment it is possible to distinguish between the genotypes differing in vitreousness. Vitreousness was highly positively correlated with test weight ( $r = 0.66$ ,  $p < 0.001$ ) and negatively with ash ( $r = -0.62$ ,  $p < 0.01$ ) (Table 3.3.2). It appears that vitreous grains have high test weight due to a higher proportion of the dense continuous starch-protein matrix. Low ash content can be attributed to low proportions of bran fraction rich in minerals. Vitreousness was also negatively correlated with *injera* softness ( $r = -0.59$ ,  $p < 0.01$ ) (Table 3.3.3). With few exceptions, cultivars with vitreous endosperm tend to produce firm *injera*. This could be due to less amylose leaching as starch granules are packed into dense protein matrices in the flour particles.

The difference in vitreousness was also reflected in a wide range of hardness (expressed as extraction rate) from 87.1% (SC-108), to 62.3% (AW). This is because the efficiency of removal of the bran depends on hardness, size and shape of the grain (Scheuring et al 1983). Within the sorghum cultivars, hardness was highly positively correlated with test weight and vitreousness ( $r = 0.78$ ,  $r = 0.65$ ,  $p < 0.001$ ) (Table 3.3.2). These positive correlations can be explained by the sorghum grain hardness being related to the distribution density of protein bodies and matrix in the endosperm (Shull et al 1990). Sorghum grain hardness negatively correlated with the positive *injera* textural attributes of softness and rollability ( $r = -0.70$ ,  $r = -0.66$ ;  $p < 0.001$ ) and positively correlated with grittiness, a negative textural attribute of *injera* ( $r = 0.42$ ,  $p < 0.05$ ) (Table 3.3.3). Rooney and Murty (1982) reported that sorghum cultivars with hard, corneous (vitreous) endosperms are undesirable for *kisra* (similar flat bread from Sudan). Grittiness of *injera* can be attributed to the particle size of the flour produced by milling. In maize, hard grain yields larger particle size flour than do softer grains (Pomeranz et al 1986), which may also apply to sorghum.

**Table 3.3.4.** Kernel and flour colors (Lab values) of the 12 sorghum cultivars and tef from the 1999 and 2000 growing seasons

Parameter	Season	Color L a b	Sorghum cultivar												Tef
			IS-777	AW	PGRC/E #69441	Seredo	CR:35:5	3443-2- op	SK-82- 022	76TI#23	Gambella 1107	PGRC/E #69349	SC-423	SC-108	DZ-01- 196
Kernel color	1999		27.3a <sup>a</sup> ±0.2 <sup>b</sup>	35.8c±0.4	32.3b±0.1	37.1d±0.2	52.9h±0.2	49.5f±0.1	45.0e±0.4	53.0h±0.4	54.5j±0.1	53.5i±0.3	54.3j±0.1	52.0g±0.1	53.4g±0.0
	2000	L	27.4a±0.1	35.4c±0.3	33.6b±0.1	39.9d±0.1	54.1j±0.3	54.0j±0.1	52.5i±0.1	47.8f±0.1	49.3g±0.2	50.6h±0.1	45.4e±0.1	47.9f±1.1	53.4i±0.0
	Mean <sup>c</sup>		27.3a±0.2	35.7b±0.4	32.9b±0.7	38.5c±1.6	53.5g±0.7	51.8ef±2.5	48.7d±4.1	50.4de±2.8	51.8ef±2.8	52.0ef±1.6	49.8de±4.9	50.0de±2.4	53.4ef±0.0
	1999		9.4k±0.0	8.2i±0.1	7.7h±0.1	8.7j±0.0	2.3a±0.1	3.5d±0.0	5.7g±0.1	4.1f±0.1	3.3c±0.1	3.4cd±0.1	3.6e±0.1	4.2f±0.1	2.9b±0.0
	2000	a	9.6g±0.2	9.7g±0.0	8.3f±0.1	8.6f±0.2	2.7a±0.2	2.8a±0.0	3.7bc±0.0	4.7e±0.0	3.4b±0.0	3.7c±0.0	4.8e±0.0	4.3d±0.0	2.9a±0.0
	Mean		9.5g±0.2	8.9f±0.8	8.0e±0.4	8.6f±0.1	2.5a±0.2	3.1bc±0.4	4.7d±1.1	4.4d±0.3	3.3bc±0.1	3.5c±0.2	4.2d±0.6	4.2d±0.0	2.9ab±0.0
	1999		8.2b±0.1	11.8c±0.1	7.8a±0.1	13.5d±0.0	14.3f±0.0	15.1i±0.0	13.9e±0.1	14.7h±0.1	15.8j±0.0	16.6l±0.1	17.1m±0.0	15.9k±0.0	14.6g±0.0
	2000	b	8.1a±0.1	13.6d±0.0	9.3b±0.1	14.6e±0.1	13.7d±0.1	15.1g±0.0	14.9f±0.1	13.5c±0.1	15.1g±0.0	15.0g±0.0	15.8i±0.0	15.2h±0.1	14.6e±0.0
	Mean		8.1a±0.1	12.7b±1.0	8.5a±0.8	14.0c±0.6	14.0c±0.3	15.1de±0.0	14.4c±0.5	14.1c±0.7	15.5ef±0.4	15.8fg±0.9	16.4g±0.7	15.6ef±0.4	14.6cd±0.0
Flour color	1999		58.4b±0.1	62.4d±0.0	57.8a±0.1	61.0c±0.0	68.5e±0.0	74.0i±0.0	68.8f±0.1	76.0l±0.0	73.9i±0.0	74.8j±0.0	73.5g±0.1	74.9k±0.0	73.8h±0.0
	2000	L	60.7a±0.0	67.9d±0.1	63.0b±0.1	64.8c±0.1	70.7e±0.0	76.6l±0.1	73.7h±0.0	73.3f±0.1	74.9i±0.0	76.4k±0.0	74.8i±0.0	74.9j±0.0	73.7g±0.0
	Mean		59.6a±1.2	65.7c±2.4	60.4a±2.9	62.9b±2.1	69.6d±1.2	75.3ef±1.4	71.2d±2.7	74.6ef±1.5	74.4ef±0.5	75.6g±0.9	74.2ef±0.7	74.9ef±0.0	73.7e±0.1
	1999		5.8h±0.0	3.5f±0.0	6.6i±0.0	3.8g±0.3	1.3d±0.0	0.1a±0.0	2.3e±0.0	0.5c±0.0	0.1a±0.0	-0.03a±0.0	0.4b±0.0	0.6c±0.0	0.1a±0.0
	2000	a	5.3j±0.0	2.5g±0.0	5.2i±0.0	3.6h±0.0	1.8f±0.0	0.2b±0.0	1.2d±0.0	1.6e±0.0	-0.2b±0.0	0.4c±0.0	0.1a±0.01	0.4c±0.0	0.1a±0.0
	Mean		5.5f±0.3	3.0d±0.6	5.9f±0.8	3.7e±0.2	1.6c±0.2	0.1a±0.0	1.8c±0.6	1.1b±0.6	-0.1a±0.0	0.2a±0.2	0.5a±0.1	0.1a±0.0	0.0a±0.0
	1999		8.9c±0.0	9.4d±0.1	8.5a±0.0	8.5a±0.0	8.6b±0.0	13.0k±0.0	10.7e±0.0	10.8f±0.0	12.4h±0.0	13.0j±0.0	13.6l±0.0	12.7i±0.0	12.0g±0.0
	2000	b	8.8b±0.0	10.2e±0.0	8.6a±0.0	9.2d±0.0	9.2c±0.0	12.6m±0.0	11.0g±0.0	0.8f±0.0	12.3l±0.0	11.4h±0.0	11.8i±0.0	12.2k±0.0	12.0j±0.0
	Mean		8.8a±0.1	9.8b±0.5	8.6a±0.1	8.9a±0.4	8.9a±0.3	12.8g±0.2	10.8c±0.1	10.8c±0.0	12.4def±0.1	12.2de±0.9	12.7ef±1.0	12.5def±0.3	12.0d±0.0

<sup>a</sup>Values followed by the same letter in the same row are not significantly different ( $p > 0.05$ ). <sup>b</sup>Values are standard deviations of three replicates.

<sup>c</sup>Values are the means of the two seasons. L = lightness, a (+) = redness, b (+) = yellowness.

**Table 3.3.5.** Thousand kernel weight, test weight, extraction rate and endosperm texture of sorghum cultivars and tef from 1999 and 2000 growing seasons

Parameter	Season	Sorghum cultivar											Tef	
		IS-777	AW	PGRC/E #69441	Seredo	CR:35:5	3443-2-op	SK-82-022	76TI #23	Gambella 1107	PGRC/E #69349	SC-423	SC-108	DZ-01-196
Thousand kernel weight	1999	20.1b <sup>a</sup> ±0.1 <sup>b</sup>	29.3k±0.1	24.0f±0.3	21.6d±0.0	22.9e±0.1	25.2g±0.0	23.6f±0.3	25.8i±0.4	27.9j±0.4	25.6gh±0.2	20.9c±0.0	21.9d±0.2	0.32a±0.0
	2000	19.5b±0.3	35.3k±0.5	23.8ef±0.6	23.8f±0.0	23.3e±0.2	22.4d±0.1	24.5g±0.1	30.3i±0.1	31.4j±0.1	39.7l±0.4	25.9h±0.1	20.3c±0.0	0.34a±0.0
	Mean <sup>c</sup>	19.8b±0.4	32.3f±3.3	23.9bc±0.5	22.7bc±1.2	23.1bc±0.3	23.8bc±1.6	24.1cd±0.5	28.0de±2.5	29.7ef±1.9	32.6f±7.7	23.4bc±2.8	21.1bc±0.9	0.33a±0.0
Test weight (kg/hl)	1999	72.0c±0.2	71.0b±0.3	78.5h±0.2	69.7a±0.3	69.6a±0.1	74.3e±0.1	73.6d±0.1	77.4g±0.1	70.7b±0.2	74.2e±0.4	69.8a±0.2	75.1f±0.3	86.5i±0.2
	2000	71.9c±0.2	74.1d±0.1	78.5g±0.2	71.5b±0.2	70.6a±0.2	74.5e±0.2	74.1de±0.2	79.2h±0.3	78.0f±0.1	79.9i±0.3	78.7g±0.2	77.9f±0.3	86.0j±0.1
	Mean	71.9abc±0.2	72.6bcd±1.7	78.5f±0.2	70.6ab±1.0	70.1a±0.5	74.4de±0.2	73.9cd±0.3	78.3f±1.0	74.4cde±4	77.0f±3.1	74.3cde±5	76.5ef±1.5	86.2g±0.3
Vitreousness (%) <sup>d</sup>	1999	32.2a±4.2	34.1a±3.2	49.2de±2.7	38.2abc±3.7	33.0a±9.4	35.7ab±1.4	45.7cde±2.0	44.1bcd±1.3	41.5abcd±8.4	51.2ef±2.8	48.6de±5.2	59.2f±12.8	ND
	2000	28.4a±7.8	37.0bc±3.2	47.6de±4.4	30.9ab±4.6	30.8ab±6.9	39.5cd±6.3	37.2bc±6.9	50.1e±1.5	50.6e±1.8	48.4e±5.5	51.4e±3.6	51.1e±3.6	ND
	Mean	30.3a±5.9	35.5abc±3.2	48.4f±3.4	34.5ab±5.5	31.9ab±7.4	37.6bc±4.6	41.5cd±6.5	47.1df±3.5	46.1df±7.4	49.8fg±4.2	50.0fg±4.2	55.2g±9.5	ND
Hardness (%) <sup>e</sup>	1999	71.2cd±2.9	56.5a±1.0	76.8def±2.8	66bc±4.9	62.2ab±4.4	71.7cd±7.0	80.3fg±2.4	73.3de±3.5	73.5de±0.9	78.8cfg±3.0	63.8b±5.0	84.5g±1.8	ND
	2000	73.6bc±3.0	76.2bcd±5.1	83.0gh±4.8	67.1a±2.0	72.0ab±2.2	78.2efg±3.6	80.0fg±4.5	73.3bcd±3.8	87.2hi±2.3	86.3hi±1.0	83.3gh±1.6	89.7i±0.6	ND
	Mean	72.4±1.2	66.4±13.9	79.9±4.4	66.6±0.8	67.1 ±6.9	75.0±4.6	80.2±0.2	73.3±0.0	80.4±9.7	82.6±5.3	73.6±13.8	87.1±3.7	ND

<sup>a</sup>Values followed by the same letter in the same row are not significantly different ( $p > 0.05$ ).

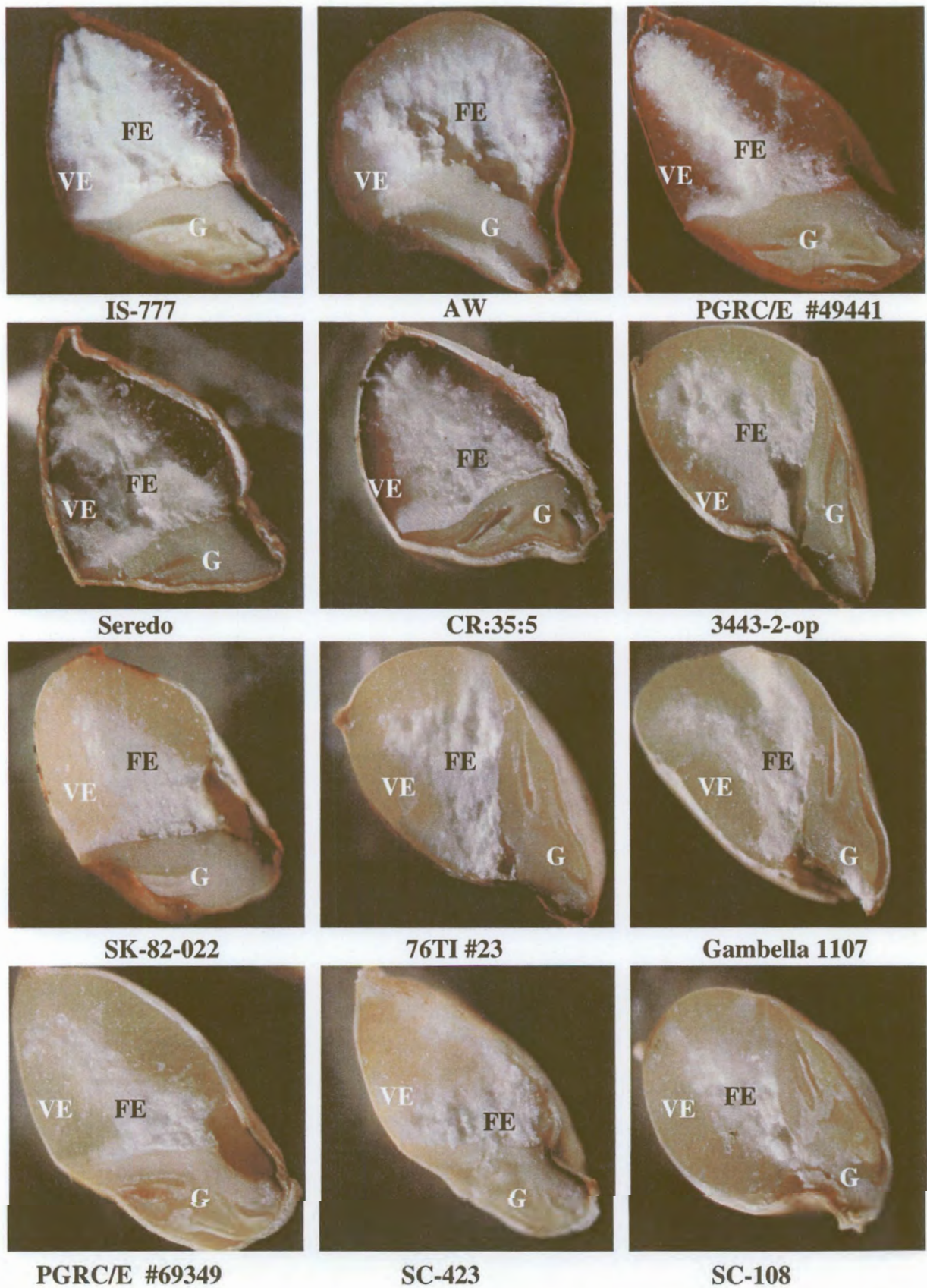
<sup>b</sup>Values are standard deviation of three replicates.

<sup>c</sup>Mean values are mean of the two seasons.

<sup>d</sup>Endosperm texture expressed as percent vitreousness (image analysis).

<sup>e</sup>Extraction rate expressed as percent hardness.

ND = Not Determined.



**Figure 3.3.2.** Micrographs of longitudinal sections of the 12 sorghum cultivars with varying endosperm texture from the 1999 growing season. FE = floury endosperm; VE = vitreous endosperm; G = germ.

### 3.3.3.2. Chemical characterization

Among the sorghum cultivars, protein content across seasons ranged from 10.5% for 76TI #23 to 15.2% for AW (Table 3.3.6). Protein content varied slightly between the seasons. In sorghum, protein content is influenced by cultivar and the environment, with considerable environmental variation (House et al 1995). The protein content of tef was lower (9.7%). These values lie within the range of reported values 7.3%-15.6% for sorghum (Serna-Saldivar and Rooney 1995) and 6.5%-9.3% for tef (Lester and Bekele 1981). Protein content negatively correlated with breakdown viscosity and peak time duration of the flour paste ( $r = -0.49$ ,  $r = -0.69$ ,  $p < 0.05$ ,  $p < 0.001$ ) respectively (Table 3.3.2). The negative relationship between protein and breakdown viscosity suggests that protein matrix inhibited the swelling of the starch granule and conferred added integrity/rigidity to the swollen granules, resulting in lower breakdown viscosity.

The lysine content of sorghums across the two seasons ranged from 1.7 g/100 g protein for 76TI #23 to 2.2 g/100 g protein for SK-82-022. Tef protein contained higher lysine (3.2 g/100 g protein). This value lies within the range of lysine content of tef (2.9-3.9 g/100g protein) as reported by Lester and Bekele (1981). In sorghum most of the endosperm protein is kafirin (prolamin), which is virtually lacking in the essential amino acid lysine (Taylor and Schüssler 1986). Umeta and Parker (1996) reported that the germ of tef grain is large in proportion to the rest of the kernel. The higher lysine content of tef is probably due to more albumins and globulins in the germ, which are relatively rich in lysine as reported for sorghum (Taylor and Schüssler 1986, Serna-Saldivar et al 1994). Tef is not decorticated prior to milling to flour; consequently, *injera* from tef is of better nutritional value compared to *injera* from decorticated sorghum.

The fat content of sorghum cultivars across seasons ranged from 2.8% (IS-777) to 3.8% (PGRC/E 69441), while the fat content of tef was in the lower range (2.8%). Differences in fat content among the sorghum cultivars appear to be due to differences in germ size and amount of oil in the germ, as reported for maize (Hoseney 1994). High fat content in whole flour of sorghum may result to rancidity problem upon storage due to oxidation of unsaturated fatty acids. Fatty acids of sorghum grain lipids are reported to be highly unsaturated (Maestri et al 1996).

Among the sorghum cultivars, ash content ranged narrowly from 1.6% to 1.9%, while tef had relatively higher 2.3% ash content. This could be attributed to the small grain size of tef resulting in a greater proportion of pericarp and aleurone layers that are rich sources of minerals (Serna-Saldivar and Rooney 1995). Among the sorghum cultivars, ash content highly negatively correlated with test weight ( $r = -0.71$ ,  $p < 0.001$ ) (Table 3.3.2). The negative relationship between ash and test weight suggests that cultivars with high test weight have lower proportion of bran resulting to low ash content.

The starch content of the sorghum cultivars across seasons ranged from 70.4% (SC-108) to 76.7% (PGRC/E #69349). Tef had relatively higher starch content (79.5%) and lower protein content. Starch plays a major role in the formation of the continuous matrix of *injera* during baking (Parker et al 1989). The continuous matrix formed due to complete gelatinization of starch during baking results to the structural integrity and texture of the *injera*. Starch content positively correlated with TKW ( $r = 0.44$ ,  $p < 0.05$ ) (Table 3.3.2). It seemed that higher kernel weight would mean more starch is present in the endosperm. Starch content also positively correlated with *injera* eye evenness and distribution ( $r = 0.55$ ,  $r = 0.52$ ,  $p < 0.01$ ,  $p < 0.05$ ), respectively (Table 3.3.3). Although these relationships were not easily explainable, this finding agrees with Subramanian and Jambunathan (1990) who reported a significant positive correlation between starch content and eye quality of *injera*.

The amylose content (amylose as percent of starch in the endosperm) across seasons for the sorghum cultivars ranged from 16.2% for (SC-108) to 25.8% for (CR:35:5). Amylose content of tef was in the lower range (18.8%) compared to sorghum cultivars. Differences in amylose content between seasons were observed for most of the sorghum cultivars. Ring et al (1989) also reported significant differences in amylose content for sorghums grown at different locations or at different seasons in the same locations. Amylose content was negatively correlated with TKW ( $r = -0.48$ ,  $p < 0.05$ ) (Table 3.3.2).

Table 3.3.7 shows that among the sorghum cultivars, WAI across seasons ranged from 1.2 g/g (AW) to 2.1 g/g (Seredo). Tef had lower WAI (1.1 g/g) and as mentioned contained higher starch (79.5%). High starch content may slow water absorption as water



**Table 3.3.6.** Chemical composition (dry weight basis) of whole grain sorghum and tef flours from the 1999 and 2000 growing seasons

Parameter	Season	Sorghum cultivar											Tef	
		IS-777	AW	PGRC/E #69441	Seredo	CR:35:5	3443-2-op	SK-82-022	76TI#23	Gambella 1107	PGRC/E #69349	SC-423	SC-108	DZ-01-196
Protein (%)	1999	13.9i <sup>a</sup> ±0.1 <sup>b</sup>	16.0k±0.1	12.2e±0.1	14.4j±0.1	11.9c±0.0	12.2d±0.0	13.6h±0.1	9.3a±0.1	12.5f±0.0	12.6f±0.0	11.8c±0.1	13.1g±0.1	9.4b±0.1
	2000	14.0gh±0.1	14.4h±0.1	12.8d±1.3	10.1a±0.0	11.8c±0.0	12.7d±0.0	13.3ef±0.1	11.8c±0.0	13.2ef±0.1	10.8b±0.1	9.5a±0.1	13.7fg±0.0	10.1a±0.0
	Mean <sup>c</sup>	13.9f±0.1	15.2g±0.7	12.5cde±0.9	12.2cd±2.4	11.9cd±0.1	12.3cde±0.3	13.4ef±0.2	10.5a±1.4	12.9def±0.4	11.7bc±1.0	10.6ab±1.3	13.4ef±0.32	9.7a±0.4
Lysine (g/100 g protein) <sup>d</sup>	1999	1.6	1.7	1.9	1.8	1.9	1.7	1.6	2.1	1.8	1.8	2.1	1.9	2.5
	2000	2.2	2.9	3.0	2.9	2.2	2.8	2.8	2.6	2.2	2.7	2.8	2.4	3.8
	Mean	1.9	2.3	2.5	2.4	2.1	2.3	2.2	2.4	2.0	2.3	2.5	2.2	3.2
Fat (%)	1999	3.0a±0.0	3.2b±0.1	3.7de±0.0	3.0a±0.1	2.9a±0.0	3.6d±0.4	3.0a±0.1	3.4c±0.4	3.8e±0.1	3.9e±0.0	4.3f±0.0	3.6d±0.0	2.9a±0.0
	2000	2.7a±0.0	3.4ef±0.1	3.9i±0.0	3.2c±0.0	3.5fg±0.1	4.0j±0.1	3.3de±0.1	3.6h±0.3	3.3cd±0.1	3.3cd±0.0	3.4de±0.1	3.6gh±0.1	2.8b±0.0
	Mean	2.8a±0.1	3.3bc±0.1	3.8e±0.1	3.1ab±0.1	3.2b±0.3	3.7de±0.3	3.2b±0.2	3.5c±0.1	3.5cd±0.3	3.6cde±0.3	3.8e±0.1	3.6cde±0.0	2.8a±0.1
Ash (%)	1999	1.9cd±0.2	1.9cd±0.2	1.6ab±0.0	1.8abcd±0.2	1.7abc±0.0	1.8abcd±0.0	1.9d±0.0	1.7abcd±0.1	1.7abcd±0.0	1.8abcd±0.0	1.8bcd±0.0	1.6a±0.3	2.3e±0.0
	2000	1.9e±0.0	1.8c±0.0	1.6a±0.0	1.8c±0.0	1.7b±0.0	1.8c±0.0	1.7b±0.0	1.6a±0.0	1.6a±0.0	1.6a±0.0	1.6a±0.1	1.6a±0.1	2.4f±0.0
	Mean	1.9d±0.1	1.8cd±0.1	1.6a±0.0	1.8bcd±0.1	1.7abc±0.0	1.8bcd±0.0	1.8bcd±0.1	1.6a±0.1	1.6a±0.1	1.7ab±0.1	1.7abc±0.1	1.6a±0.2	2.3e±0.3
Starch (%)	1999	71.0ab±0.4	73.7cd±3.5	74.8cdef±0.2	74.5cde±0.3	76.6efg±1.6	79.8hi±2.1	74.6cdef±0.1	77.6gh±2.6	77.0fg±0.7	75.6defg±0.6	72.7bc±0.9	69.2a±0.8	80.4i±0.9
	2000	71.5a±0.9	73.7abcd±2.2	75bcde±0.5	76.7def±0.8	73.3abc±1.0	74.8bcd±0.7	75.0bcde±0.5	76.1cdef±2.9	74.5abcd±1.8	77.9ef±1.6	72.4ab±4.7	71.5a±0.3	78.6f±0.2
	Mean	71.3a±0.7	73.7bc±2.6	74.9cd±0.4	75.6cde±1.3	74.9cd±2.2	77.3e±3.1	74.8cd±0.4	76.8de±2.6	75.8cde±1.9	76.7de±1.7	72.6ab±3.0	70.4a±1.4	79.5f±1.1
Amylose (% of starch)	1999	24.2gh±1.5	15.5a±0.5	22.6ef±1.0	20.3d±0.4	25.3i±1.5	23.9fg±0.7	24.3gh±0.0	20.6d±0.1	18.0c±0.5	16.3ab±0.4	21.4de±0.3	17.5bc±0.3	18.6c±1.2
	2000	26.3g±2.0	17.5bc±0.1	27.4g±2.4	19.9def±0.2	26.3g±1.6	20.9ef±0.2	21.3f±0.2	19.6de±0.2	16.7b±0.5	17.1b±0.2	19.9def±0.1	15.0a±0.1	19.1cd±0.2
	Mean	25.2g±1.9	16.5a±1.3	25.0g±3.1	20.1cd±0.3	25.8g±1.5	22.4f±1.7	22.8f±1.7	20.1cd±0.5	17.3ab±0.8	16.7a±0.6	20.6d±0.8	16.2a±1.4	18.8bc±0.8

<sup>a</sup>Values followed by the same letter in the same row are not significantly different ( $p > 0.05$ ).

<sup>b</sup>Values are standard deviations of three replicates.

<sup>c</sup>Values are means of the two seasons.

<sup>d</sup>Single determinations.

may be differentially absorbed by the various components within the flour. Differences in WAI also appear to be due to differences in damaged starch during mechanical milling. Damaged starch absorbs more water as opposed to intact starch granules (Whistler and BeMiller 1999). Among the sorghum cultivars WAI was negatively correlated with TKW, vitreousness and WSI ( $r = -0.44$ ,  $r = -0.44$ ,  $r = -0.52$ ,  $p < 0.05$ ), respectively (Table 3.3.2).

WSI of sorghum flours across seasons ranged from 1.4 g/100 g (Seredo) to 2.8 g/100 g (AW). Tef flour had a higher WSI (3 g/100 g). Water-soluble pentosans and other low molecular weight components in tef flour might have contributed to its higher WSI. Whole sorghum flours were reported to contain high level of water insoluble cell wall material (5.3%) (Verbruggen et al 1993) that might have contributed to low WSI.

#### 3.3.3.3. *Pasting properties and gel firmness*

Concerning pasting properties (Table 3.3.8), among the sorghum cultivars, peak viscosity (PV) across the two seasons ranged from 155 (SC-108) to 253 RVU (SK-82-022). The PV of tef was significantly higher (255 RVU). PV indicates the highest apparent viscosity obtained during pasting (Abd Karim et al 2000) and water-binding capacity of starch (Newport Scientific 1995). It appears that tef flour starch had a greater water-binding capacity than sorghum flours upon heating. This is probably due to the smaller tef starch granule size with higher surface area for more water penetration as opposed to the relatively larger sorghum starch granules. Among the sorghum cultivars, peak viscosity negatively correlated with fat content ( $r = -0.43$ ,  $p < 0.05$ ) and positively with starch content ( $r = 0.52$ ,  $p < 0.05$ ) (Table 3.3.2). Lipids in the flour tend to coat the surface of starch granule and act as physical barrier to starch swelling (Whistler and BeMiller 1999). Flours containing higher amount of starch have been associated with generally higher viscosities (Whistler and BeMiller 1999).

It has also been reported that the PV of sorghum starches is affected by growing environment (Beta and Corke 2001).

**Table 3.3.7.** Water absorption and water solubility indices of whole sorghum and tef fours from the 1999 and 2000 growing seasons

Parameter	Season	Sorghum cultivar												Tef
		IS-777	AW	PGRC/E #69441	Seredo	CR:35:5	3443-2-op	SK-82- 022	76TI #23	Gambella 1107	PGRC/E #69349	SC-423	SC-108	DZ-01- 196
Water absorption index (g/g)	1999	1.6fg <sup>a</sup> ±0.2 <sup>b</sup>	1.2a±0.0	1.5cd±0.0	1.8g±0.2	1.4bcd±0.1	1.4bcd±0.1	1.3abc±0.0	1.3ab±0.1	1.4bcd±0.0	1.4abc±0.01	1.6ef±0.1	1.5bcd±0.0	1.3ab±0.1
	2000	1.8g±0.1	1.2b±0.1	1.7efg±0.1	2.4h±0.2	1.8fg±0.2	1.5cde±0.1	1.6def±0.1	1.5cde±0.0	1.4bc±0.0	1.3b±0.1	1.3b±0.1	1.4bcd±0.1	1.1a±0.0
	Mean <sup>c</sup>	1.7e±0.2	1.2ab±0.0	1.6de±0.2	2.1f±0.4	1.6de±0.2	1.5cd±0.1	1.4cd±0.2	1.4bcd±0.1	1.4bcd±0.0	1.3abc±0.1	1.5cd±0.2	1.4bcd±0.1	1.1a±0.1
Water solubility index (g/100 g)	1999	2.3b±0.0	2.5b±0.6	2.4b±0.2	1.4a±0.3	2.3b±0.2	2.4b±0.4	2.8bc±0.2	2.5b±0.2	2.3b±0.4	2.5b±0.2	2.9bc±0.5	2.5b±0.5	3.2c±0.7
	2000	3.3g±0.5	3.1fg±0.3	2.0b±0.6	1.4a±0.2	1.9ab±0.1	2.7cde±0.0	2.9def±0.5	2.6cde±0.1	2.2bc±0.1	2.8de±0.4	2.2bc±0.1	2.4bcd±0.1	2.9def±0.3
	Mean	2.8de±0.7	2.8de±0.5	2.2bc±0.4	1.4a±0.2	2.1b±0.3	2.5bcd±0.3	2.8de±0.3	2.5bcd±0.2	2.2bc±0.3	2.7cde±0.3	2.6bcd±0.5	2.4bcd±0.3	3.0f±0.5

<sup>a</sup>Values followed by the same letter in the same row are not significantly different ( $p > 0.05$ ).

<sup>b</sup>Values are standard deviations of three replicates.

<sup>c</sup>Values are means of the two seasons.

Hot paste viscosity (HPV) positively correlated with starch content and peak viscosity ( $r = 0.55$ ,  $r = 0.86$ ;  $p < 0.01$ ,  $p < 0.001$ ) and negatively with vitreousness ( $r = 0.86$ ,  $p < 0.001$ ) (Table 3.3.2).

Breakdown viscosity (BV) of tef flour paste (135 RVU) across seasons was three times that of SK-82-022 sorghum, a cultivar with the lowest BV (37 RVU). Han and Hamaker (2001) reported that differences in breakdown value were due to differences in rigidity/fragility of the swollen granules. Highly swollen starch granules are fragile and easily broken by stirring, which leads to a decrease in viscosity (Whistler and BeMiller 1999). This agrees with the finding reported in Chapter 3.1. Zobel (1984) also reported that starches with greater shear thinning are more soluble. Among the sorghum cultivars breakdown viscosity negatively correlated ( $r = -0.49$ ,  $p < 0.05$ ) with protein content (Table 3.3.2). This negative relationship suggests the involvement of protein in limiting shear thinning in the sorghum flour pastes.

Cold paste viscosity (CPV) of sorghum cultivars across the two seasons varied. The highest CPV was recorded for the cultivar 76TI #23 (457 RVU) and the lowest for SC-108 (235 RVU). Tef had a CPV of 270 RVU. CPV is related to the ability of starch to gel after cooling (Whistler and BeMiller 1999). It can vary with botanical source of the starches, amylose content and formation of amylose-lipid complex (Whistler and BeMiller 1999). CPV was positively correlated with TKW, starch content, PV and HPV ( $r = 0.57$ ,  $r = 0.61$ ,  $p < 0.01$ ;  $r = 0.79$ ,  $r = 0.84$ ,  $p < 0.001$ ) (Table 3.3.2).

Setback viscosity (SBV) across the two seasons for sorghum flour pastes ranged from 144 RVU (SC-108) to 278 RVU (76TI # 23). Tef flour paste showed lower setback viscosity (150 RVU). This agrees with the finding of Bultosa et al (2002) that tef starch had low setback viscosity compared to maize starch. As stated by Abd Karim et al (2000), setback is likely related to the retrogradation tendency of amylose. SBV was highly positively correlated ( $r = 0.93$ ,  $p < 0.001$ ) with CPV (Table 3.3.2). This strong relationship explains the obvious fact that pastes with high setback giving rise to high cold paste viscosity. Setback viscosity also highly positively correlated with bending force of *injera* stored for 24 hr ( $r = 0.62$   $p < 0.01$ ) and 48 hr ( $r = 0.61$   $p < 0.01$ ) (Table 3.3.3). This long term

retrogradation tendency is probably related to amylopectin molecules with a higher proportion of long chains.

Pasting temperature (PT) across seasons for sorghum flours ranged from 77°C (Seredo) to 85°C (3443-2-op). Tef flour showed substantially lower pasting temperature (71°C). This could probably be due to the higher amylopectin content of tef starch compared to sorghum (Table 3.3.6), as high amylose content is believed to actively inhibit swelling in normal cereal starches (Tester and Morrison 1990). As reported by Parker et al (1989), during the baking of *injera*, starch granules completely gelatinize and fuse into a continuous amorphous matrix in which gas bubbles are trapped. The lower pasting temperature of tef starch might allow faster matrix formation. This seems to favor trapping of numerous gas bubbles in the continuous amorphous matrix, which appears to give the desired textural properties (softness, fluffiness and rollability) of tef *injera*. In sorghum its higher pasting temperature might lead to more gas escape before the formation of the continuous matrix.

Peak time duration (PTD) across the two seasons for sorghum cultivars ranged from 1.7 min (SC-423) to 2.9 min (AW). Tef had a longer PTD (3.5 min), which probably resulted to low HPV (Table 3.3.8) due to shear force rupture and fragmentation of starch granules. Among the sorghum cultivars, PTD was positively correlated with breakdown viscosity and setback viscosity ( $r = 0.69$ ,  $r = 0.56$ ;  $p < 0.001$ ,  $p < 0.01$ ) and negatively with protein content ( $r = -0.69$ ,  $p < 0.001$ ) (Table 3.3.2).

Gel firmness (maximum force required for deformation) of the sorghum cultivars varied from 1.6 N (Gambella 1107) to 3.3 N (76TI #23) (Table 3.3.8). The gel firmness of tef flour was significantly higher (8.4 N). The low gel firmness of sorghum flours compared to tef flour appears to be due to differences in chemical composition. Hosoney (1994) reported that gel firmness is an important determinant of food quality as it influences textural properties. The higher tef gel firmness might be related to its soft *injera* texture. Among the sorghum cultivars seasonal and cultivar differences in gel firmness were also observed. This agrees with Murty et al (1982a) who reported significant cultivar differences for gel consistency of cooled thin porridges from sorghum. Gel firmness

**Table 3.3.8.** Pasting properties and gel firmness of whole sorghum and tef flours from the 1999 and 2000 growing seasons

Parameter	Season	Sorghum cultivar											Tef	
		IS-777	AW	PGRC/E #69441	Seredo	CR:35:5	3443-2- op	SK-82-022	76TI #23	Gambella 1107	PGRC/E #69349	SC-423	SC-108	DZ-01- 196
Peak viscosity (RVU)	1999	210f <sup>a</sup> ±2 <sup>b</sup>	187d±1	155c±3	202e±1	221i±1	217h±0	274k±2	253j±3	214g±1	219hi±0	136b±1.0	125a±1.5	251j±2
	2000	225d±2	234ef±1	196b±0	263h±1	237f±2	227d±1	231e±2	234ef±1	217c±2	255g±5	234f±5	186a±2	260h±1
	Mean <sup>c</sup>	217def±8	210cde±33	176ab±23	233efg±33	229defg±9	222def±5	253g±31	243fg±11	215def±2	237efg±20	185bc±54	155a±33	255g±5
Hot paste viscosity (RVU)	1999	159f±1	142d±1	112c±4	152e±1	164g±5	177h±2	138d±0	179h±8	165g±4	162fg±1	72b±2	61a±1	116c±3
	2000	166c±3	181fg±1	137b±4	213i±1	184g±2	174e±3	192h±0	180f±2	180f±0	179f±4	170d±0	122a±3	124a±4
	Mean	162c±5	162c±21	124b±17	182c±33	174c±12	176c±3	165c±30	179c±5	172c±9	170c±9	121b±54	91a±33	120b±5
Breakdown viscosity (RVU)	1999	52de±2	45bc±2	44b±1	50cd±1	57e±5	40ab±1	36a±2	74g±10	50cd±5	57e±2	65f±1	63f±1	136h±1
	2000	59cde±4	53bc±1	60def±4	51b±2	52b±1	53b±3	39a±3	54bcd±2	36a±2	77g±8	65f±4	64ef±1	135h±4
	Mean	55d±5	49bcd±5	52cd±9	51bcd±2	54d±4	46bc±7	37a±3	64e±13	43ab±8	67e±12	65e±3	63e±1	135f±3
Cold paste viscosity (RVU)	1999	332g±4	360h±1	273d±5	318f±1	362h±1	417j±1	308c±5	456k±2	321f±1	385i±3	181b±2	152a±3	260c±2
	2000	347d±2	394f±5	312b±1	403g±0	358e±2	399fg±2	355e±4	458i±2	358e±4	512j±9	435h±6	319c±1	281a±2
	Mean	340cde±9	377ef±19	292abc±21	361def±47	360def±3	408fg±10	331bcde±26	457g±2	340cde±20	449g±70	308bcd±139	235a±91	270ab±12
Setback viscosity (RVU)	1999	174g±4	218i±1	162de±2	167ef±1	199h±4	239j±1	170fg±5	278k±10	158d±5	223i±3	110b±1	91a±2	144c±1
	2000	182b±5	214d±5	176b±4	191c±1	174b±0	225e±1	163a±4	279g±1	178b±4	334h±13	265f±6	198c±1	157a±3
	Mean	178abc±6	216ed±4	169ab±8	179abc±13	186bc±14	232d±8	167ab±6	278c±6	168ab±12	278e±61	187bc±86	144a±59	150ab±7

**Table 3.3.8 (continued).**

Pasting temperature (°C)	1999	85gh±0	82de±1	83ef±1	79c±4	84fgh±0	86i±1	85gh±0	80cd±0	73b±0	84fg±1	74b±1	74b±2	71a±1
	2000	85ef±1	85ef±0	82d±1	76b±1	81cd±2	85ef±0	85ef±0	84e±2	86f±1	81cd±0	80c±1	80c±1	72a±1
	Mean	85def±0	83def±2	82cde±1	77b±3	83def±2	85f±1	85ef±0	82cd±2	79bc±7	82cdef±2	77b±3	77b±3	71a±1
Peak time duration (min)	1999	1.7a±0	1.8a±0	1.9b±0	2.1d±0	2.1d±0	2.0c±0	2.1d±0	2.4f±0	1.9bc±0	2.1d±0	2.3e±0	2.1d±0	3.5g±0
	2000	1.9b±0	1.6a±0	2.0bc±0	2.1c±0	1.9b±0	1.9b±0	1.9b±0	2.1c±0	1.7a±0	2.4d±0	2.3d±0	2.1c±0	3.5e±0
	Mean	1.8ab±0	1.7a±0	1.9bc±0	2.1d±0	2.0cd±0	1.9bc±0	2.0cd±0	2.2e±0	1.8a±0	2.2e±0	2.3e±0	2.1d±0	3.5f±0
Gel firmness <sup>d</sup> (N)	1999	2.0abc±0.5	2.1abc±0.2	2.6cd±0.1	2.8d±0.4	2.8d±0.3	2.3bcd±0.5	1.8ab±0.5	3.6e±0.2	1.5a±0.2	1.5a±0.3	2.2bc±0.3	2.1bc±0.2	9.1f±0.4
	2000	2.0ab±0.5	1.7a±0.2	3.9e±0.3	2.6bc±0.5	3.1cd±0.3	2.1ab±0.4	1.9a±0.4	3.1cd±0.4	1.6a±0.2	3.2d±0.1	3.6de±0.2	3.1cd±0.2	7.8f±0.5
	Mean	2.0abc±0.4	1.9ab±0.3	3.2fg±0.7	2.7defg±0.4	3.0efg±0.3	2.2abcd±0.4	1.8ab±0.4	3.3g±0.4	1.6a±0.2	2.4bcde±0.9	2.9efg±0.8	2.6cde±0.6	8.4h±0.8

<sup>a</sup>Values followed by the same letter in the same row are not significantly different ( $p > 0.05$ ).

<sup>b</sup>Values are standard deviations of two replicates.

<sup>c</sup>Values are the means of two seasons.

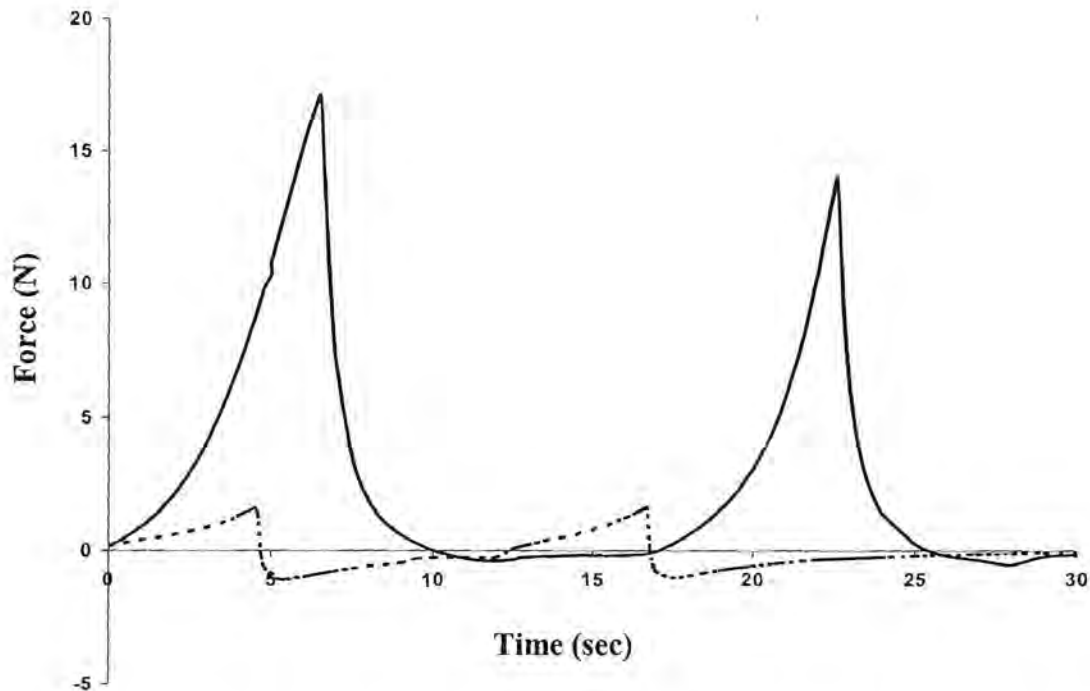
<sup>d</sup>Values are mean of four replication.

RVU = Rapid Visco Analyser Units.

positively correlated with test weight, SBV and BV ( $r = 0.44$ ,  $r = 0.43$   $p < 0.05$ ;  $r = 0.57$ ,  $p < 0.01$ ) and negatively correlated with protein content and ash content ( $r = -0.55$ ,  $r = -0.56$ ,  $p < 0.01$ ) (Table 3.3.2).

Figure 3.3.3 illustrates the properties of double compressed sorghum, and tef flour gels. In the first compression cycle, the breaking point of sorghum flour gel was much lower compared to tef flour gel. Tef flour gel was firmer and required higher compression force to deform as shown by its higher peak. After the deforming force of the first compression cycle was removed and the second compression force was applied, tef flour gel almost returned back to its undeformed condition. It appears that tef flour gel is relatively elastic compared to sorghum flour gel. This can be attributed to higher levels of water soluble component in tef flour and lower setback viscosity of its flour paste that might have increased the soluble phase of the gel and promoted the formation of a stronger network structure. Because rigidity is as a result of starch granule swelling, the binding of solubilized molecules and the formation of physical cross-links resulting from molecular reassociation (Zobel 1984). In terms of physical measurement, rigidity results from the applied stress being stored in the paste rather than being dissipated, such a paste is said to be visco-elastic (Zobel 1984). Tef (DZ-01-196) flour starch had a similar low amylose content as sorghum (76TI #23). The difference in gel texture points to structural differences in the amylopectin component of the starch. During the cold storage of the gels, syneresis was observed for the sorghum gels. This agrees with Wu and Corke (1999) who associated softness of gel with release of water (starch syneresis) during storage.





**Figure 3.3.3.** Double compression of sorghum and tef flour gels aged for 24 hr at 4 °C.  
 Sorghum (76TI #23) ..... and tef (DZ-01-196) ———

### 3.3.4. Conclusions

In terms of chemical composition, compared to tef flour, higher protein, lower protein lysine content, higher fat, and lower starch contents characterize sorghum flours. Sorghum flours showed higher pasting temperature, higher trough (holding strength), lower breakdown (shear thinning), higher final viscosity, and higher setback as compared to tef flour. Sorghum flours formed softer and less elastic gels. Based on linear regression correlation analysis across seasons, physical properties of the grain had a considerable effect on the sensory textural attributes of sorghum *injera*. Cultivars with floury endosperm, relatively lower test weight with low extraction rate were positively correlated with soft and rollable sorghum *injera* texture. Water solubility index of the flour was positively correlated with fluffiness of *injera*. These parameters have potential to be used as indirect indices in predicting the quality of sorghum *injera* in ESIP. However, further research is needed to test large numbers of genotypes from multi-location trials to further evaluate the effects on *injera* quality attributes.

### 3.3.5. References

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