Malt Quality Traits of Different Barley Genotypes from High Growing Potential Areas of Ethiopia

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Abstract

This study was conducted to assess malt quality traits of different barely genotypes grown at different locations. Sixty barley samples were collected from Holeta, Debre-Birhan and Bekoji that used for quality analysis. Genotypes FBPVT and NPPT had higher protein content. Locations (Holeta and Debre-Birhan) showed the lowest protein content with 8.93% and 8.31%, respectively than Bekoji which had 11.70%. Genotypes FBNVTN, FBNVTOG and MBNVTOG had higher friability value than other genotypes. On the other hand, FBPVT and FBNVTOG exhibited higher beta-glucan content. Holeta and Debre-Birhan locations showed higher extract and friability contents whereas Bekoji had lower extract content and friability. Contrarily, the highest protein and beta-glucan contents were obtained from Bekoji location. In general, the protein and extract contents, and friability obtained in the present study were in acceptable range according to international malt quality traits standards. Thus, these quality traits could be used to select appropriate genotype for further breeding program.

Keywords: Barley, Genotypes, Malt, Traits

Introduction

Barley (*Hordeum vulgare* L.) is the fourth most important cereal crop worldwide after wheat, corn and rice (Marwat *et al*, 2012). It is a crop of ancient origin in Ethiopia and the country is considered as a center of diversity for barley, because of the presence of great diversity in ecology (Birhane, 1991). In the country barley has a long history of cultivation in the highlands (Firdissa *et al*, 2010). Barley has the ability to adapt and survive in a wide range of environmental conditions, but the diversity of barley types found in Ethiopia is probably not expanded in any other region of comparable size (Bekele, 1983). Even though it is most important crop for food in Ethiopia, but it is used mostly for the production of malt (Ullrich, 2002). Barley malt is mainly used as a source of fermentable sugars for alcoholic fermentation for the production of beer (Kreisz, 2009).

Barley is a complex mixture of many organic components that include protein, starch, water, oil, fib4.25 136136(olt)-11(y)20stac49(ha)-5(irde)5es and aes Deofs

of barley in particular, modifies the grain components during the controlled steeping, germination and drying processes (Bamforth *et al*., 1993). However, varying the malting process conditions influences the level to which the carbohydrate and protein constituents are modified, which in turn influences beer processing and product characteristics.

The ability to predict grain quality for different purposes in early generations would be of great benefit to breeders and brewers, allowing for selection of suitable genotypes to deliver product of the highest quality. The fact the production of malt barley is restricted to some specific areas is advantageous with respect to transport, storage and research (Kunze, 2004). However, the problem arises in the selection of suitable cultivars for each region that meet the required quality specifications. Breeding of new cultivars therefore requires the evaluation of many quality characteristics and the testing and selection of thousands of breeding lines, starting with early generation material in the breeding program. Therefore, the barley genotypes grown in different locations in Ethiopia needs to be analyzed for their quality traits to categorize it as malt and food purposes. The barley breeding programs conducted in the country by regional and federal agricultural research centers mainly depends on the selecting materials based on the quality traits and agronomical data to judge it as malt or food. Therefore, the present study was designed to assess malt quality traits of different barely genotypes grown at different locations in Ethiopia.

Materials and Methods

Study site and sampling

The samples used in this study were from barley breeding program trials of Ethiopian Institute of Agricultural Research Centers having the mandate of barley growing and breeding potential around central highland areas of Ethiopia. Accordingly, the barley samples were collected representing a range of breeding generations grown at different environments throughout barley growing highland areas specifically from Holeta, Debre Birhan and Bekoji. Sixty samples from 2018-year trials of malt barley were collected from the pre-specified growing areas from different plots depending on the agronomical data and source of genotypes from where they originated as well as the history of their quality.

Sample preparation

A 150g per sample was taken after manually cleaned and graded for biochemical composition and spectral analysis. Before malt quality traits analysis, the malt barley samples were malted according to Phoenix Automated Micro malting system (Phoenix Bios stems, Adelaide, Australia) designed to process 300g of 24 barley samples per batch (Nilsen and Panozzo, 1995). After kilning the rootlets were removed from the malted samples by using mechanical malt cleaner that had

been reconfigured to simultaneously process eight 250 g samples (Fraser Fabrications Pty Ltd, Malaga Western Australia. Then, the barely samples were ground using Mill3100 grinder (Perten Instruments, Hagersten, Sweden) to pass through 0.5mm. But, for malt friability determination the malt sample was not grounded, because the friabilimeter machine itself grounds the sample for the ratio of friability measurement.

Malt quality analysis

Malt quality traits were analyzed at the Food Science and Nutrition Laboratory, Holeta Agricultural Research in collaboration with VLB Institute in Berlin (Germany) for the quality traits.

Malt extract content

Malt extract content was determined according to a small-scale version of the European Brewery Convention (EBC, 1998) Methods Manual, Section 4.9.1. Fine grind malt was extracted using a hot water mashing bath (SIEMENS Mashing Machine, Germany). For extraction, 50 g of finely ground malt was mixed with 200 mL of distilled water and mashed at 45°C with continuous stirring. After 30 minutes of mashing, the temperature was increased by 1°C/min until 70°C. As temperature reaches 70°C, 100 mL distilled water was added. After 1 hour, the mash was cooled to 30°C and adjusted to a volume of 515 mL or a weight of 450g. The extract was filtered using whatman 12cm filter paper into 500ml cylinder and specific gravity was measured at 20^{o}C using a DMA5000 density meter (Anton Paar GmbH, Graz, Austria). Therefore, the following formula was used to calculate the final result. E = P (800+M)/(100-P); where, E= Extract content, P= Wort Density (^{o}Plato), M= Malt Moisture content.

Total malt protein

The malt protein content was determined using kjeldhal method (Digester SBS 2000, Distillation Unit 5000DL, Food ALYT GimbH, Germany) according to (AOAC, 2005). For analysis, one gram ground sample of malt barley was measured and transferred into completely dry kjeldhal flask. Ten gram of kjeldhal tablet was added to the sample inside the flask. Twenty milliliter of 98% concentrated sulphuric acid was mixed with the sample. The sample digestion was started by connecting the kjeldhal flasks with the digestion rock. The digestion was completed when the brown color of the sample completely disappeared. After the digested sample was cooled, 100 ml of distilled water and 80 ml of sodium hydroxide (32%) were added and distilled into 25 ml of excess boric acid containing 0.5 ml of screened indicator. The distillate was titrated with 0.1N hydrochloric acid to the methyl red end point. The protein was calculated by using this formula: CP%= [(T-B)*14*6.25)]/ [W (100-MC)]; where CP=Crude Protein, T= Volume of HCl used in Titration, B= Blank used as control and W= Weight of sample taken for analysis.

Malt friability

Malt grain samples were analyzed using a friability measuring machine (Pfeuffer Friabilimeter GmbH, Germany) which used a pressure roller to grind the sample against a rotating screen. Low, medium and high friability malts were tested according to EBC method 4.15 (EBC, 1998). Fifty gram malt sample was run in the friabilimeter for 8 min and the non-friable fraction was weighed to get the final result.

Malt β-glucan

The malt β-glucan content was determined using the Megazyme kit method (Megazyme, Bray, Ireland) according to EBC, 1998 Method 4.16.1. For the analysis, 100 mg sample was suspended and hydrated in a buffer solution of pH 6.5 and incubated with purified lichenase enzyme and filtered. An aliquot of the filtrate was then hydrolyzed to complete with purified β-glucosidase. The D-glucose produced was assayed using a glucose oxidase/peroxidase reagent. The final prepared aliquot was measured by spectrophotometer at absorbance 510 nm against reagent blank within one hour. Finally, the beta-glucan was calculated using the formula; B-glucan (%W/W) = ΔA * (F/W) * 27; Where, ΔA= Absorbance after β-glucosidase treatment (reaction) minus reaction blank absorbance, F = Factor for the conversion of absorbance values to μg of glucose, W = The calculated dry weigth of the sample analyzed in mg.

Moisture content

Barley grain dry matter content was determined according to (AOAC, 2005) international standard method from grain flour prepared using the above sample preparation method. Five gram of barley flour was taken using a sensitive analytical balance and oven dried at 105^{O}C temperature for 3 hours. After the dried sample was cooled in a desiccator, the final measurement was taken using the same analytical balance to get the result using the following known formula for moisture content. $MC\% = (W_i-W_f)/W_i*100$; where W_i is initial weight, W_f is Final Weight

Data Analysis

The collected data was analyzed using SAS statistical software version 9. The main effects of the factors were compared using ANOVA statistical tool. The means were separated to their significance level using LSD at $p<0.05$.

Results and Discussion

Malt extract content

Genotypes Variation: Statistically, a significant difference was obtained between genotypes at $p<0.05$ for malt extract contents (Table 1). Genotypes MBNVT N and MBNVT OG had higher extract content with 80.70% and 80.67% respectively than other genotypes. Similar finding reported by Swanston et al. (2014) observed

that the extract yield varied depending on genotypes which affect the extent of enzymatic degradation and the solubility of grain components after malting and mashing. During malting, enzymes that have an impact on the degradation of substrates were either synthesized or cleaved from their bound forms. The range of enzymes produced included those that degrade cell wall components, proteins and starch. This is also influenced by the nature of the genotypes performance to produce enough enzymes during such processing. As the objective for most maltsters is to maintain high extract levels and yet somehow achieve relatively high extract content according to EBC standard from 70-80% based on genotypes.
Location Variation: Significant effect was observed between growing locations (Holeta, Debre-Birhan and Bekoji) for malt extract content at ($p<0.05$). Higher extract content was observed with Debre-Birhan and Holeta locations with a value of 80.3% and 79.91% respectively whereas Bekoji showed lower extract (76.78%). This variability could be from environmental, growing conditions, temperature, available nitrogen and moisture (Fox et al., 2003).

Protein content

Genotypes Variation: The protein content of barley throughout genotypes varied from 8-16%. The EBC standard range malt barley protein content ranges from 9-11.5%. Genotypes had a significant effect on protein content at $p<0.05$) (Table 2). Accordingly, genotypes FBPVT and NPPT gave higher protein content with 10.37% and 10.50% respectively than the other genotypes. This could be due to the genetic makeup the genotypes. In the brewery, standard protein content is not needed to be higher as well as lower, but need to be in the range of 9-11.5%.
However, in this study since some food barley genotypes were included in the samples and the protein content showed less than 9%. Emebiri et al. (2007) reported that protein variability occurred due to genotypes variability. The authors also reported negative correlation between protein and extract, a positive correlation between protein and diastatic power, using a low protein breeding population, mean that the quality traits correlation of genotypes could affect the protein variation among genotypes.

Location Variation: A Significant difference was observed between locations (Holeta, Bekoji and D/Birhan) at $p<0.05$ (Table 2). Accordingly, Bekoji location showed higher protein content (11.70%) than Holeta and Debre-Birhan locations with 8.93% and 8.31% protein values respectively. This might be due to the differences in growing conditions (soil, moisture, temperature). This variability was also reported by Emebiri et al. (2007) that barley type (one and two rowed, malt and food type) and a parental irrelative affects protein content. In addition, the authors reported that protein variability occurred due to environment and nitrogen fertilizer application. Thus, the protein content would be varied with genotypes, soil type, growing season, agricultural practices and amount of rain fall.

β

β

Percent of friability

Genotypes Variation: Measuring the friability of commercial malt has increasingly been used as an indicator to malting and brewing quality as well as trouble shooting on samples of poor malt quality. A significant difference was observed between the genotypes in their percent friability at $p<0.05$ (Table 2). Genotype MBNVT OG (73.00%) had higher friability followed by FBNVT OG (68.67%) and FBNVT N (68.33%), whereas genotype FBPVT (45.33%) gave lower friability compared to others. Friability potential of genotypes needs to be higher in breeding lines for the purpose of barley malt commercial as set European Brewery Convection (EBC, 1998). The lower and varied value of friability occurred among genotypes could not be only the genotype variation but also occurred because of relationship with other malt quality parameters and malting process as reported by (Chapon *et al.*, 1978).

Location Variation: Significant variation was observed between locations on the friability at $P<0.05$ (Table 2). Higher friability was observed at Holeta (76.71%) and followed by Debre-Birhan (71.57%) whereas Bekoji (42.57%) showed lower friability.

β-Glucans content

The major constituent of barley endosperm cell wall is -D-(1-3), (1-4)-glucans (75%), with a minor component identified as arabinoxylans (20%) (Fincher and Stone, 1986). The range in barley for glucan is 2 to 10% of total grain weight (Henry, 1987).

Genotypes Variation: The mean values for β-Glucan content was significantly differed among genotypes at $P<0.05$. Genotypes, FBPVT and FBNVT OG had the highest β-glucan content with values of 862.7 mg/L and 885 mg/L, respectively as compared to the other genotypes. But, the significant variation between genotypes is very important for further breeding works.

Location Variation: The same trend as in genotypes was observed for between location that the β-glucan mean value was significantly differed at $p<0.05$. Bekoji gave higher β-glucan content (958.1mg/L) as compared to Holeta and Debre-Birhan locations with mean values of 437.3 mg/L and 650.1mg/L respectively. Higher value of β-Glucan content is not needed for malt commercial since it contributes undesirable effect in other malt quality. The same as in genotypes significant variation between locations is very important to consider factors in breeding programs.

Henry (1985) reflected both genotypes and location influenced the content of β-glucan as it has been shown to have a relationship with other malt quality traits. Importantly, high β-glucan levels may not result in higher or lower extract but relate to other malt quality traits such as Kolbach Index (ratio of soluble to total protein), viscosity or the speed of filtration (Evans *et al.* 1999). There was a contradicting idea between that the higher β-glucan content as lower the amount of extract and indirectly contributing for reducing extract content rather having direct relation with wort speed of filtration and viscosity.

Moisture content

The moisture content of barley is 8-15% on average. The moisture content can vary between 12% in very dry harvesting conditions and over 20% in wet conditions. More precisely, it is less than 13% in the South region of the European Brewery Convention (EBC) barley malt and it is more than the 16% in the North region, where consequently the barley should be dried before long term storage.

Genotypes Variation: Barley must have moisture content below 15% for long term storage. But, this study did not show a significant variation in moisture content among the genotypes. However, in the contrary some scholars found significant variation among genotypes for moisture content in the accepted range from 8.65-10.93%. Moreover, moisture levels need to be low enough to inactivate the enzymes involved in seed germination as well as to prevent heat damage and the growth of disease microorganisms. Quality and germination capacity may also significantly deteriorate (Plankinton *et al*., 2014).

Location Variation: As shown in Table 2 a non-significant difference was observed between locations at $p>0.05$. Moreover, the determination of the

moisture content is important when the amounts of the other components are related to the dry weight (Kunze, 2004, Vijaya, 2003).

Conclusion and Recommendations

Overall, the result of the present study showed that genotypes FBPVT and NPPT had higher protein content within the acceptable range. Extract content was higher for NPPT, MBPVT, MBNVTOG and MBNVTN genotypes. Genotypes, FBNVTN, FBNVTOG and MBNVTOG had higher percent of friability than other genotypes. FBPVT and FBNVTOG genotypes exhibited the highest β-glucan content. Holeta and Debre-Birhan locations showed higher extract and friability content whereas Bekoji location gave lower extract and friability content. On the other hand, higher protein and β-glucan contents were obtained from Bokoji location. In addition, protein, extract and friability traits were in acceptable range according to international malt quality traits standards. Therefore, these quality trait will be used to select appropriate genotype for further breeding program. However, all barley growing areas were not included in this research work so future studies are needed to investigate effect of diverse growing conditions on malt quality trait of these genotypes.

References

AOAC. 2005. Official methods of Analysis. 17th edn. Association of Official Analytical Bamforth CE and Barclay AHP. 1993. Malting technology and the uses of malt. In: *Barley: Chemistry and Technology* (edited by A.W. MacGregor & R.S. Bhatty). Pp. 297-354. St. Paul, Minnesota, USA: American Association of Cereal Chemists, Inc.

Berhane L. 1991. Barley Dependence Cereal in Ethiopia, IAR, News Letter Agricultural Research (2):3.

Chapon L, Gromus J, Erber HL and Kretschmur H. 1980. Physical method of the determination of malt modification and their relationship to wort properties. European Brewery Convention, Monograph VI - Symposium on the relationship between malt and beer, Helsinki, 45-70.

Chapon L, Maucourt, JM and Gobert J. 1978. The friabilimeter an apparatus for objective measurement of malt friability. 32: 160–167.Chemists, Washington DC.

Duffus CM. and Cochrane MP. 1992. Grain structure and composition. In: Shewry, P.R. (Ed.), Barley: Genetics, Biochemistry, Molecular Biology and Biotechnology. CAB International, pp. 291e317.

Edney M, MacLeod A and LaBerge D. 2014. Evolution of a quality testing program for improving malting barley in Canada. *Canadian journal of plant science*. (2014) 94.

Emebiri LC, Moody DB, Black C, van Ginkel M and Hernandez E. 2007. Improvement in malting quality barley grain yield by manipulation of genes influencing grain protein content. Euphytica 156, 185-194.

European Brewery Convention. 1998. *Analytica EBC*. 5th ed. Verlag Hans Carl, Nurnberg, Germany.

Evans DE and Hejgaard J. 1999. The impact of malt derived proteins on beer foam quality Part I: The effect of germination and kilning on the level of protein Z4, Protein Z7 and LTP1. Journal of the Institute of Brewing 105, 159-169.

Fox G, Panozzo J, Li C, Lance RC, Inkerman P, and Henry RJ. 2003. Molecular basis of barley quality, Australia journal of agriculture research. 54, 1081–1101.

Fridissa E, Woldeyeseus S, and Heinrich G. 2010. On farm Diversity and Characterization of Barley (*Hordeum vulgare L.*) Landraces in the Highlands of West Shewa, Ethiopia vol.8:025 034.

Henry RJ. 1985. Use of a scanning near-infrared reflectance spectrophotometer for assessment of the malting potential of barley. *Journal of the Science of Food and Agriculture*, 36, 249-254.

Kreisz S. 2009. Malting. In: *Handbook of brewing: Processes, Technology, Markets* (edited by H.M. Eßlinger). Pp. 147-164. WILEY-VCH Verlag GmBH & Co. KGaA, Weinheim.

Kunze W, "Technology Brewing and Malting" VLB, Berlin. 2004.

Marwat SK, Hashimi M, Khan KU, Khan MA, Shoaib M and Fazal-ur-Rehman. 2012. Barley (*Hordeum vulgare L.*) A Prophetic Food Mentioned in Ahadith and its Ethnobotanical Importance. American-Eurasian Journal of Agricultural and Environmental Sciences. 12(7). P. 835–841.

Nilsen M. and Panozzo J. 1995. In: *Leaping ahead with Near Infrared Spectroscopy*, Ed by Batten, G.D., Flinn, P.C., Welsh, L.A. and Blakeney, A.B. NIR Spectroscopy Group, Australia, p. 174-177.

Plankinton Ave and Milwaukee. 2014. Malting barley quality requirements, American Malting Barley Association, Inc.vol. (414) 272-4640.

Swanston JS, Wilhelmson A, Ritala A, Gibson BR. 2014. Malting, brewing and distilling. In: Shewry PR, Ullrich SE, eds. *Barley: Chemistry and Technology. St. Paul: AACC International, 193 222.*

Ullrich SE. 2002. Genetics and breeding of barley quality attributes. In: *Barley Science: Recent advances from molecular biology to agronomy of yield and quality* (edited by G.A. Slafer, J.L. Molina-Cano, R. Savin, J.L. Araus & I. Romagosa). Pp. 115-142. Binghamton, New York: The Haworth Press.

Vijaya Raghavan GS. 2003. "Commercial Grain Dryers" in Handbook of Postharvest Technology Cereals - Fruits, Vegetables, Tea, and Spices, Amalendu Chakraverty, Indian Institute of Technology Kharagpur, India, pp. 167-183.