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A comparative evaluation of agronomic performance and kernel composition of normal and high sugary corn genotypes (*Zea mays* L.) grown for dry-grind ethanol production

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ABSTRACT

High sugary corn genotypes (*Zea mays* L.) have the potential to reduce enzyme consumption and enhance ethanol yield during dry-grind ethanol production. In the present work, four high sugary and four parent field corn genotypes (HSGs and PFCs, respectively) were studied comparatively to evaluate agronomic performance and kernel composition of the genotypes, as well as predict the effects of these traits on carbohydrate accumulation in the kernels. The corn genotypes were grown over two cropping years (2012 and 2013) under rain fed conditions, and some important agronomic traits and kernel biochemical components were determined. Both HSGs and PFCs varied in the agronomic properties, but these variations were not atypical in HSGs if compared with those of PFCs. The average grain yield ranged from 6.19 Mega gram per hectare (Mg/ha) to 9.43 Mg/ha in HSGs and 5.77 Mg/ha to 10.23 Mg/ha in PFCs. Sugar accumulation in the kernels was found to be negatively correlated with flowering time, grain filling period and physiological maturity of the genotypes. Compared with PFCs, a higher amount of total soluble sugars (TSS) and a lower quantity of starch were recorded in HSGs, which resulted a significant negative correlation between kernel starch and TSS. The agronomic performance and kernel composition of HSGs, particularly with high kernel sugars and low starch contents suggest that these corn genotypes could be promising candidates for producing cost-effective ethanol during dry-grind process.

1. Introduction

Corn (*Zea mays* L.) is one of the most important cereal crops, and being a natural source for carbohydrates, it has been used for producing various food, feed and industrial products since last several decades (Semenčenko et al., 2015). In more recent times, this cereal crop has drawn much attention in fuel ethanol production, as attempts to reduce the dependence on fossil fuels gathers pace globally (Zabed et al., 2014). Corn has been proved to be a promising feedstock for bioethanol production because of its high kernel (grain) and ethanol yields (7.26 Mg/ha and 417 L/ha, respectively) (Gumienna et al., 2016). However, quality and availability of corn are important issues in the sustainability and expansion of this industry, which can partially be done by developing new and promising hybrids (Gumienna et al., 2016; Ramchandran et al., 2015). As attempts to address it, much research efforts have been made in recent years to develop corn hybrids having potential to produce high quality kernels and enhanced amounts of ethanol (Chen et al., 2014; Yangcheng et al., 2013). Although high starchy hybrids have been suggested for increased ethanol yield (Sanchez and Cardona, 2008), high fermentable sugar containing hybrids are considered promising over the formers from both economic and

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Abbreviations: AL, amylose; AM, amylopectin; AT, anthesis time; BLM, black layer maturity; DM, dry matter; EL, ear length; ELD, eight leaves development; ENP, ear number per plant; EW, ear weight; FLD, four leaves development; HSG, high sugary genotypes; GRF, grain filling period; GY, grain yield; K, potassium; KD, kernel depth; KNP, kernel number per plant; LL, leaf length; LNP, leaves number per plant; N, nitrogen; P, phosphorous; PFC, parent field corn; pH, plant height; PY, potential yield; SET, seed emergence time; ST, silking time; TKW, thousand kernel weight; TOM, total organic matter; TOC, total organic carbon; TRS, total reducing sugars; TSS, total soluble sugars.

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technical points of views (Bothast and Schlicher, 2005; Mojović et al., 2013).

In spite of industrial maturity, corn ethanol is still facing some challenges for its long-term economic survival and expansion of the industry. Requirement of amylolytic enzymes for converting starch into glucose before fermentation is considered a cost increasing factor for corn ethanol (Shigechi et al., 2004). The costs of enzymes required for hydrolysis has been reported to be nearly 10–20% of the total production costs (Gregg et al., 1998). As attempts to reduce enzyme consumption, high sugary corn genotypes (HSGs) were developed in previous studies and observed that HSGs had the potential to reduce enzyme consumption during dry-grind ethanol production in a conventional (Zabed et al., 2016a) and a non-cooking method (Zabed et al., 2016b). Despite this promising contribution of HSGs to the dry-grind ethanol production, research efforts are still required to characterize these genotypes, where a particular attention should be given to the agronomic performance and kernel biochemical composition. A comprehensive characterization of HSGs along with the respective parent field corn lines (PFCs) grown under the same conditions will be useful to make a comparison between normal and high sugary genotypes, which further helps to select appropriate growth conditions for HSGs and make these genotypes acceptable or popular to the breeders and producers. Moreover, agronomic and biochemical traits of corn hybrids have direct or indirect effects on the final ethanol yield (Lacerenza et al., 2008; Lorenz et al., 2009; Yangcheng et al., 2013). It has also been reported that kernel compositions of corn affect quality of co-product (distiller's dried grains with solubles, DDGS), which is produced during dry-grind ethanol production and contributes to the overall economy of the plant (Liu, 2011).

Among the agronomic traits, grain yield is the most important parameter, which can vary with hybrids even if all other factors are supposed to be similar (MiJa et al., 2012). Moreover, it has been well documented that grain yield has a significant effect on ethanol productivity (Obuchowski et al., 2010). Other agronomic parameters are interlinked either with grain yield or carbohydrate accumulation in the kernels (D'Andrea et al., 2009), thereby affecting ethanol yield indirectly. Furthermore, several agronomic traits, such as thousand kernel weight have been reported to affect final ethanol yield directly (Swanston et al., 2007), in addition to showing a positive correlation with the amount of kernel starch (Obuchowski et al., 2010). Kernel number per plant (KNP), length and weight of ear, and thousand kernel weight (TKW) are directly correlated with kernel yield (D'Andrea et al., 2008).

The biochemical components of kernels can be divided into two groups, namely, fermentable and non-fermentable attributes. In all types of corn, starch is the major fermentable component, while varying quantities of sugars may also present based on the types of corn (Manikandan and Viruthagiri, 2010; Yangcheng et al., 2013). The non-fermentable components are mainly protein, fat, fiber and ash (Singh, 2012). Although starch is the major carbohydrate in corn, an insignificant correlation has been reported between kernel starch and ethanol yield (Reicks et al., 2009). Because, amylose to amylopectin ratio of starch affects physicochemical properties of starch and can eventually affect the efficiency in the conversion of starch to ethanol (Yangcheng et al., 2013). More recently, a negative correlation has been reported between starch and ethanol yield while studying with 258 corn hybrids (Gumienna et al., 2016). Therefore, it is not reliable to predict ethanol yield directly from the quantity of starch present in the kernels (Swanston et al., 2007). On the contrary, kernel sugars have been found to be correlated significantly with fermentable sugars and ethanol yield (Gumienna et al., 2016; Zabed et al., 2016b).

Taking into account the above facts, the present study aimed to conduct a comprehensive characterization of four HSGs and four PFCs over two cropping years in 2012 and 2013 for agronomic and biochemical traits in light of ethanol production. In other words, HSGs and PFCs were studied comparatively to find the answer of a major research question whether HSGs be suitably grown under the same condition as maintained for normal corn or require extra care for their growth in consideration of the agronomic and biochemical properties. It is hypothesized that HSGs will be able to grow efficiently under the similar growth conditions of PFCs without showing any unusual properties. The major unusual traits in HSGs compared with PFCs may include significantly lower grain yield, longer time for different growth stages (e.g., emergence time, flowering time and maturity), and higher amounts of non-carbohydrate components in the kernels.

2. Materials and methods

2.1. Materials

Seeds of four HSGs, namely, UM.NF-1, UM.NF-4, UM.NF-6 and UM.NF-11, and respective PFCs, such as PFC-1, PFC-4, PFC-6 and PFC-11 were collected from Dr. Golam Faruq, Institute of Biological Sciences, Faculty of Science, University of Malaya, Kuala Lumpur, Malaysia. The major seed quality attributes of the corn genotypes are presented in Table 1. All the chemicals and reagents used in this study were reagent grade and purchased from either Sigma-Aldrich (St. Louis, MO, USA) or Fisher Scientific (Waltham, MA, USA).

2.2. Experimental design

Field experiments and data collection were done in two successive cropping years at the experimental field located near the department of Genetics and Molecular Biology, Institute of Biological Sciences, Faculty of Science, University of Malaya, Kuala Lumpur, Malaysia (3°7′1″N and 101°39′12″E). The soil quality of the experimental field is presented in Table 2.

The corn genotypes were grown in the field in 2012 (September–December) and 2013 (August–November in 2013) under rain fed conditions following randomized complete block design (RCBD) with four replications. An individual experimental plot consisted of 5 rows, 2.5 m long and 0.75 m apart, with hill to hill distance maintained at 0.5 m giving a gross area for each plot of 7.5 m^2. The plots were planted with four seeds in each hill on September 3 (2012) and August 1 (2013). The plots were thinned to 8 plants/m^2 (80,000 plants/ha) by removing extra plants around 10 days after sowing at the three leaf stage (V3) (Cirilo et al., 2009; Gambín et al., 2007). Similar amounts of N, P and K were applied to all plots of each genotype (HSGs and PFCs) in the form of 15-15-15 at a rate of 125 kg/ha prior to sowing the seeds and only nitrogen as urea was applied twice during growth of the genotypes, 15 days after sowing (around the four leaf stage, V4) and on 40 days after planting (15–20 days before anthesis) with 86 and 62 kg N/ha respectively, based on the optimization work of a previous study on the same field. Insects and weeds were adequately controlled throughout the growing cycle of the corn genotypes. Hand weeding was done thrice during the growing season at 4th, 6th and 9th weeks after planting.

2.3. Data collection and analysis for agronomic traits

Duration for seedling emergence (50% of the total plants were visible on the soil surface), 4 leaves development (in 50% plants), 8 leaves development (in 50% plants), anthesis or tasseling (50% of plants showed visible tassel), silking (50% of plants showed visible silk), and maturity (when 50% of the plants showed black layer formation in the kernels) were recorded in each plot during the experimental period (Cirilo et al., 2009). Corn plants were harvested when moisture level reached to around 24–26%. During harvest,