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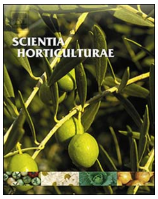

Genetic diversity, population structure and genetic parameters of fruit traits in *Capsicum chinense*

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ABSTRACT

The fruits of *Capsicum chinense* stand out with a wide diversity of sizes, colors, shapes, and levels of pungency. Knowledge about this variability is essential for the success in species conservation and breeding programs. This study evaluated 65 *C. chinense* accessions, from different geographic regions of Brazil, for genotypic fruit traits, correlations between fruit traits, genetic parameters and diversity, population structure, and associations of AFLP markers with fruit traits. Deviance analysis showed significant differences between the accessions for all traits. Heritability in the broad sense (h_g^2) ranged from 0.31 (soluble solids content) to 0.90 (fruit diameter). The accuracy of genotype selection was considered high (≥ 0.70) for most of the traits, except for soluble solids and dry matter contents (0.43 and 0.62, respectively). A high correlation between fresh and dry fruit weight was observed. However, these traits were negatively correlated with dry matter content. Soluble solids content was not correlated with any of the traits. The AFLP markers indicated high genetic variability among accessions. When the groups formed by molecular data and fruit traits were compared, no associations were observed, indicating that both stages of characterization are important. Structure analysis formed two groups, partially concordant with molecular hierarchical grouping. The AFLP markers were significantly associated with several fruit traits and considered interesting candidates for further studies.

1. Introduction

Pepper (*Capsicum* spp.) is one of the most important vegetables in the world due to its high versatility and wide application range in industry, cooking and for decoration (Hulse-Kemp et al., 2016; Silvar and García-González, 2017). It is a genus with wide variability, with 38 described species, of which only *C. annuum* var. *annuum* L., *C. baccatum* var. *pendulum* (Willd.) Eshbaugh, *C. chinense* Jacq., *C. frutescens* L. and C. *pubescens* Ruiz et Pav. are domesticated (Pozzobon et al., 2006; Dewitt and Bosland, 2009; García et al., 2016).

Brazil is considered one of the most important diversity centers of domesticated and wild species of *Capsicum* (Barboza et al., 2005). In this country peppers play an important role in socioeconomic, biological and cultural aspects, due to the wide applicability of fruits in traditional culinary foods and dishes, with regional diversity and sometimes regional uniqueness (Sudré et al., 2010). The wide diversification in the use of *Capsicum* spp. fruits is probably related to the genetic diversity observed in each region of the country.

Among the domesticated species, *C. chinense* is considered the most important in Brazil, with the Amazon Basin as putative center of domestication (Pickersgill, 1971; Moses and Umaharan, 2012; Moses et al., 2014). This species has a wide variability of fruits in terms of size, color, shape, and pungency levels, and is widely used in Brazilian cuisine (Baba et al., 2016). Therefore, knowledge about the variability in this species is essential for successful conservation and its use in breeding programs.

Several studies have addressed the characterization of the genetic diversity of *C. chinense* based on phenotypic and molecular descriptors (Jarret and Berke, 2008; Finger et al., 2010; Moses and Umaharan, 2012; Bharath et al., 2013; Moses et al., 2014; Baba et al., 2016). However, genetic studies for fruit traits are scarce for this species (Sousa and Maluf, 2003; Schuelter et al., 2010; Bharath et al., 2013),

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since most of them focused on *C. annuum*. In an recent evaluation of 264 *C. chinense* accessions from the Caribbean, Bharath et al. (2013) detected wide variability for fruit traits, with the broad-sense heritability (h^2g) varying from 0.52 (pedicel length) to 0.81 (fruit diameter and weight) among accessions. In *C. annuum*, Naegele et al. (2016) observed a high h_a (> 0.90) for most of the 13 fruit traits evaluated in 116 accessions.

In studies based on quantitative traits in *Capsicum* spp., multiple QTL maps have been associated with fruit size, weight, shape, color, and metabolites (Ben-Chaim et al., 2001; Zygier et al., 2005; Yarnes et al., 2013; Naegele et al., 2014; Han et al., 2016; Nimmakayala et al., 2016). Two major QTLs (*fs3*.1 and *fs10.1* – chromosome 3 and 10, respectively) that control fruit shape variation (FS) were identified, accounting for about 60 and 40% of the phenotypic variation, respectively (Ben-Chaim et al., 2001; 2003). For fruit weight (FW), Rao et al. (2003) and Zygier et al. (2005) identified a major QTL (*fw2.1*) located on chromosome 2. These results were corroborated by Han et al. (2016), in an evaluation of an ultra-high density map of crosses between *C. annuum* cultivars, who also identified the importance of these QTLs for the respective traits. However, Nimmakayala et al. (2016) evaluated 96 *C. annuum* accessions by associative mapping, and observed 16 SNPs strongly associated with FW, located in different genomic regions.

In these studies of genetic diversity and QTL mapping in *Capsicum* spp., several molecular markers (e.g., RAPD, AFLP, SSR, and SNPs) were applied. Among these techniques, Amplified Fragment Length Polymorphism (AFLP) is an important tool in view of its wide genome coverage, reproducibility, cost-effectiveness, and sequence information independent genotyping (Zhang et al., 2014). In this study we used AFLP markers addressed to achieve the following objectives: i) determine the genetic parameters associated with fruit traits of *C. chinense* accessions from Brazil; ii) identify correlations between these traits; iii) determine population structure and genetic diversity; and iv) identify AFLP markers associated with the fruit traits.

2. Material and methods

2.1. Phenotyping

We evaluated a total of 65 *C. chinense* accessions from eight Brazilian states (Mato Grosso, Maranhão, Pará, Bahia, Rio de Janeiro, Minas Gerais, Goiás, and Espírito Santo with 26, 11, 11, 6, 4, 3, 2, and 2 accessions, respectively), comprising four geographic regions of Brazil (Mid-West, North-East, North, and South-East, with 28, 17, 11, and 9 accessions, respectively), maintained in the germplasm bank of the Universidade Estadual de Londrina (UEL), Londrina, Paraná, Brazil (Table1).

These accessions were evaluated in a field of the UEL (latitude 23°20'32″ S, longitude 51°12′32″ W; 550 m asl) in two growing seasons. Experiment I was carried out from September to December 2016 and, experiment II, from December–2016 to Mach 2017. The experiment was arranged in a randomized block design, with two replications and 12 plants per plot, in a 0.5 × 0.9 m spacing between plants and rows, respectively. The soil of the experimental area was classified as Red Latosol and the climate was subtropical, mesothermal humid (Cfb), with hot summers, rare frosts and tendency of concentrate drainfall in the summer months.

The soil chemical properties were determined as follows: $pH_{H2O} = 6.60$, $P = 103.65\ mg/dm^3$, $K = 0.28\ cmolc/dm^3$, $Ca^{+2} = 9.0\ cmolc/dm^3$, $Mg^{+2} = 1.7\ cmolc/dm^3$, $Al^{+3} = 0.0$, $H + Al = 2.70\ cmolc/dm^3$, $CEC = 15.0\ cmolc/dm^3$, $V = 80\%$, and $OM = 2.30\%$. The soil was prepared by fertilization with $18\ t\,ha^{-1}$ organic compost one week after transplanting the seedlings, inoculated with Bokashi®, applying 25 g product per plant, manually incorporated and covered with straw. A drip irrigation system was installed and weed growth was inhibited by scattering straw in and in-between the rows.

Table 1
Geographic distribution and morphological descriptors of 65 *Capsicum chinense* accessions in Brazil of gene bank of Universidade Estadual de Londrina (UEL).

ID	Origin	Morphological descriptors		
		Fruit shape	Fruit color at intermediate stage	Fruit color at mature stage
Gen_1	Mato Grosso	Elongate	Green	Red
Gen_2	Mato Grosso	Elongate	Green	Red
Gen_3	Mato Grosso	Elongate	Green	Red
Gen_4	Mato Grosso	Elongate	Green	Red
Gen_5	Mato Grosso	Triangular	Green	Red
Gen_6	Mato Grosso	Elongate	Green	Red
Gen_7	Mato Grosso	Other	Green	Orange
Gen_8	Mato Grosso	Elongate	Green	Orange
Gen_9	Mato Grosso	Almost round	Green	Lemon-yellow
Gen_10	Mato Grosso	Campanulate	Green	Dark red
Gen_11	Mato Grosso	Almost round	Green	Light red
Gen_12	Mato Grosso	Elongate	Green	Red
Gen_13	Mato Grosso	Campanulate	Green	Red
Gen_14	Mato Grosso	Other	Green	Pale orange
Gen_15	Mato Grosso	Elongate	Green	Dark red
Gen_16	Mato Grosso	Almost round	Green	Orange
Gen_17	Mato Grosso	Triangular	Purple	Light red
Gen_18	Mato Grosso	Blocky	Green	Dark red
Gen_19	Mato Grosso	Almost round	Green	Red
Gen_20	Mato Grosso	Campanulate	Purple	Light red
Gen_21	Mato Grosso	Triangular	Green	Red
Gen_22	Mato Grosso	Almost round	Green	Red
Gen_23	Mato Grosso	Elongate	Green	Orange
Gen_24	Mato Grosso	Almost round	Green	Red
Gen_25	Mato Grosso	Elongate	Green	Orange
Gen_26	Mato Grosso	Triangular	Green	Light red
Gen_27	Goiás	Almost round	Purple	Red
Gen_28	Goiás	Almost round	Green	Lemon-yellow
Gen_29	Minas Gerais	Campanulate	Green	Red
Gen_30	Minas Gerais	Campanulate	Green	Light red
Gen_31	Minas Gerais	Campanulate	Green	Dark red
Gen_32	Bahia	Triangular	Green	Red
Gen_33	Bahia	Blocky	Green	Pale Orange
Gen_34	Bahia	Campanulate	Green	Red
Gen_35	Rio de Janeiro	Almost round	Green	Orange-yellow
Gen_36	Pará	Campanulate	Green	Pale Orange
Gen_37	Bahia	Campanulate	Green	Red
Gen_38	Pará	Elongate	Green	Red
Gen_39	Pará	Blocky	Green	Red
Gen_40	Pará	Blocky	Green	Red
Gen_41	Maranhão	Blocky	Green	Red
Gen_42	Maranhão	Campanulate	Green	Red
Gen_43	Maranhão	Triangular	Green	Red
Gen_44	Maranhão	Other	Green	Red
Gen_45	Maranhão	Blocky	Green	Red
Gen_46	Maranhão	Other	Purple	Orange
Gen_47	Rio de Janeiro	Elongate	Green	Red
Gen_48	Espírito Santo	Blocky	Green	Red
Gen_49	Espírito Santo	Elongate	Green	Red
Gen_50	Maranhão	Elongate	Green	Red
Gen_51	Maranhão	Elongate	Green	Orange
Gen_52	Maranhão	Elongate	Green	Red
Gen_53	Maranhão	Elongate	Green	Red
Gen_54	Bahia	Campanulate	Green	Red
Gen_55	Bahia	Elongate	Green	Red
Gen_56	Pará	Triangular	Green	Red
Gen_57	Pará	Elongate	Green	Red
Gen_58	Pará	Campanulate	Green	Red
Gen_59	Pará	Triangular	Green	Red
Gen_60	Pará	Elongate	Green	Red
Gen_61	Pará	Triangular	Green	Red
Gen_62	Pará	Blocky	Green	Red
Gen_63	Maranhão	Blocky	Green	Red
Gen_64	Rio de Janeiro	Campanulate	Green	Red
Gen_65	Rio de Janeiro	Elongate	Green	Red

Three crops were harvested and the following agronomic descriptors evaluated: fruit length (cm), fruit diameter (cm), pericarp thickness (mm), mean fruit weight (g), dry fruit weight (g), fruit dry matter content (%), and soluble solids content (°Brix).