The Application of Tukey's Mean Difference and Dendrogram Statistics to Identify Local Genetic Sources of Corn in Selecting for Prospective Parental Lines

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Abstract. Commercial corn hybrids became expensive for local farmers. Extensive use of corn hybrids since 1980 left numerous obsolete hybrid varieties all over Indonesia. The hybrids became obsolete by the introduction of the new ones. Over time the obsolete hybrids expressed superior traits toward detrimental hindrances in situ such us resistance to pest and disease and drought, required less fertilizers while yielded at par with the new hybrids. The traits were identified, the obsolete hybrids were worthy to select for being local genetic sources (LGS) in a corn breeding program. The research was done to identify the LGS collected from Padang, Palembang, Jogjakarta, West Kalimantan and Lampung. In 2018 the LGS were planted in two rows and open-pollinated among themselves. The seeds were harvested separately to make female lines. A synthetic line was made by mixing 100 seeds of each female line. In 2019 the six lines: five female and one synthetic, and Pioneer36 (P36) F1-hybrid as control were planted in a randomized complete block design with 3 replicates. The data were collected for traits: plant height, leaf number plant⁻¹, time to tasseling, time to silking, ear length, ear diameter, 100 seed weight and seed yield m⁻². The data were variance-analysed and Tukey's HSD was applied to separate and group the lines. A dendrogram was constructed to decide lines suitable to recombine. The result indicated that the Padang, Palembang, Lampung and Synthetic lines could be used as the male parents to recombine with the Jogjakarta and West Kalimantan lines.

Keywords: corn breeding, corn superior-female line, corn synthetic line, dendrogram, local genetic source, Tukey's HSD

1. Introduction

Developing better new varieties of corn had the second importance in plant breeding program in Indonesia after rice [1]. The dependency of animal feeds on corn as raw material increased from year to year following the Government program of increasing animal protein intake in diet to fight stunting among children. As the facts that Indonesia had been introducing hybrid corn varieties since 1975, the corn breeding program had successfully introduced no less than 200 hybrid varieties, exotic and own hybrids as well [2]. The speed of the introduction was rather high, in an average of five hybrids every year. This depleted the parental lines required for corn breeding also rather sooner and impacted on the price of hybrid-corn seeds. The price of the hybrid seeds nowadays was 28-30 times higher than that of 20 years ago.

The introduction of the hybrid corn varieties in 1975 was to fight *Peronosclerospora* downymildew disease [2]. The disease was soil-borne type hard to defeat and fast to disperse. Later the *Fusarium* and *Colletotrichum* also soil-borne diseases infected the corn hybrids. Therefore the hybrids released and marketed should have the resistance against the diseases. And the resistance so complete might not be found easily in one or several parents maintained by the breeders. Following the dispersion of the hybrids, the hybrids released earlier became obsolete. They were of superior recombination only to be put aside due to the release of the newer hybrids. The obsolete hybrids existed, however, planted and managed by the local farmers. In time the hybrids became the gene pool 1 *in situ* having traits well adapted and considered superior locally. The local genetic source (LGS) of corn varieties expressed superiority on resistant to drought, low pH, lower fertilizer requirement, and pest infestation, while maintained acceptable seed yield [3].

In time that the lines beneficial for recombination became less and less due to their extensive use in hybridization, the genetic relationship among the hybrids so developed became closer [4]. This resulted in their superiority against the diseases and unfavorable planting conditions reduced and resulted in using more insecticides and fertilizers. In this concept the use of the LGSs as parent lines in a breeding program was considerable. The study intended to evaluate the potential of the LGSs of corn collected from five Indonesia's provinces as parent lines in the breeding program.

The problems in using the LGSs were each of the LGS might be superior in its localization and would expressed wide differences in preferred characters when planted *ex situ* [5]. The differences mostly due to adaptation ability on a different localization. To overcome the adaptation limitation the LGSs were planted in the first localization in Lampung province before were tested for their performances in the second localization [6]. Data were collected on vegetative and reproductive traits to decide the best LGS or several LGSs for recombination in later programs.

Tukey's mean difference (Tukey's HSD) might be the simplest statistics to differentiate the trait's performance means among the LGSs, however the Tukey's HSD differentiated only for each performance [7]. This led to the problem of deciding which LGSs were prospective based on all tested trait's performances. The study analysis offered a solution to the problem by arranging the trait's performance done with Tukey's HSD into ranks of the LGSs. The performance data would also be used to construct a dendrogram which would explain supposed recombination among LGSs in developing prospective corn hybrids [8].

2. Materials and Methods

The study was done in rainy season, March – June 2018 in Gaya Baru, East Lampung Regency and March – June 2019 in the State Polytechnics of Lampung's research field. The soil in both places was of Red-Yellow Podsolic ultisol having low pH and fertility. Five LGSs were collected from Padang, Palembang, Jogjakarta, West Kalimantan and Lampung provinces. The sixth LGS, a synthetic line was made as a mixture of 100 seeds of each LGSs. In 2018 the LGS were planted in two rows and let open-pollinate among themselves. The seeds were harvested separately to make superior-female lines. In 2019 the six lines: five female and one synthetic lines, and Pioneer36 (P36) F1-hybrid as control were planted in a randomized complete block design with 3 replicates. The data were collected for

traits: plant height (cm), leaf number plant⁻¹, time to tasseling (days after planting; dap), time to silking (dap), ear length (cm), ear diameter (cm), 100 seed weight (g), seed number ear⁻¹ and seed yield m⁻² (kg). The data were analyzed with ANOVA and Tukey's HSD was applied to separate the means so that to construct the rank of the LDSs, respectively. A dendrogram was constructed based on the distances among lines to decide lines suitable to recombine.

3. Results and Discussion

3.1. ANOVA's Mean Squares Analyses for Plant Traits

Table 1a and 1b indicated that there were mean differences for LGS lines meant that that would be averaged value which would ideal for recombination. The CV value ≤ 10 % would indicate that the averaged means fit the ideal expectation whereas ≥ 15 % indicated that the LGSs needed to be reselected to decrease the CV for the trait. The data indicated that the average value for each traits was within the range of new released national corn hybrids, only the standard deviations for 100 seed weight, seed weight ear⁻¹ and seed yield m⁻² were high ≥ 15 %. Furthermore, the data indicated that the traits: plant height, leaf number, days to tasseling, days to silking would not need further improvement because their average values were already within the national range. However, the ear diameter trait would need to increase to 5.0-5.5 cm through a recombination with new hybrid varieties [9].

Table 1a. Mean square analyses for plant traits.

Source of variation	df	Plant height	Leaf number	Days to tasseling	Days to silking	Ear diameter
Replicate	2	832.73	1.92	2.89	2.65	0.01
LGS lines	6	719.15*	7.33**	40.62**	39.42**	0.52**
Error	12	214.89	0.55	5.80	8.05	0.07
Total	20	8558.99	54.45	319.16	338.44	4.02
CV %		8.33	4.91	4.52	5.31	6.41
Average value		176.06 cm	15.11	53.32 dap	53.41 dap	4.14 cm
Standard deviation %		11.47	10.65	7.31	7.52	10.56

^{*} and ** indicated there were mean differences at P \leq 0.05 and P \leq 0.01, respectively

Table 1b. Mean square analyses for plant traits.

Source of variation	df	Ear length	Seed row ear ⁻¹	100 seed weight	Seed number ear ⁻¹	Seed yield m ⁻²
Replicate	2	0.59	0.08	2.63	1162.91	0.002
LGS lines	6	1.86**	4.59**	72.24*	8624.83**	0.13**
Error	12	0.10	0.75	16.48	1292.19	0.02
Total	20	13.50	36.74	636.40	69581.11	0.96
CV %		1.86	6.26	13.69	11.81	16.58
Average value		17.02 cm	13.84	29.65 g	304.36	0.78 kg
Standard deviation %		4.71	9.56	18.57	18.91	27.49

^{*} and ** indicated there were mean differences at $P \le 0.05$ and $P \le 0.01$, respectively

3.2. Mean Difference Analyses and Grouping of the LGS

The Tukey's HSD was used to separate the variable means and resulted in that the traits separated into two classes, except seed yield m⁻² which separated into three classes, in which West Kalimantan LGS was the lowest (horizontal data in Table 2).

Table 2 (vertical data) indicated the superiority of a certain LGS over the others. Following the reasoning in studying the data vertically in which the means having "a" would be considered the best, Table 2 would present the rank of the LGSs as well based on how many "a"s a certain LGS owned [10]. Thus Table 2 indicated that the hybrid P36 would be in rank-1; the Padang, Palembang, Lampung and Synthetic LGSs in rank-2; Jogjakarta LGS in rank-3 and West Kalimantan in rank-4. That the hybrid P36 was in the rank-1, or the best, would not be surprising because it was a new national-hybrid and had been tested and certified to be suitable on a low-pH soil (Red-Yellow Podzolic ultisol) planting in Lampung. Furthermore, the data also indicated that the Padang, Palembang, Lampung and Synthetic LGSs were in the same rank-2 which indicated that the LGSs expressed a good adaptability on the Lampung soil.

Table 2. Mean analysis for plant traits using Tukey's $HSD_{0.05}$ and rank of the LGSs.

	LGS							
Trait	Padang	Palembang	Jogjakarta	West Kalimantan	Lampung	Synthetic	P36	$\mathrm{HSD}_{0.05}$
Plant height (cm)	177.6 ab	166.93 ab	187.47 a	145.27 b	181.43 ab	184.43 ab	189.3 a	41.89
Leaf number	16.2 a	14.87 a	15.67 a	11.8 b	16.47 a	15.67 a	15.13 a	2.12
Days to tasseling (dap)	57 a	54.47 a	52.27 ab	45.67 b	54.6 a	53.73 a	55.53 a	6.88
Days to silking (dap)	56.8 a	54.93 a	52.47 ab	45.8 b	54.4 a	53.8 ab	55.67 a	8.11
Ear diameter (cm)	4.08 a	4.21 a	4.47 a	3.28 b	4.57 a	4.17 a	4.22 a	0.76
Ear length (cm)	17.13ab	17.00ab	16.33bc	15.67c	17.53a	17.57a	17.93a	0.41
Seed row ear-1	13.73 b	13.6 b	13.6 b	12.13 b	16.31 a	13.63 b	13.87 ab	2.48
100 seed weight (g)	30.79 a	30.71 a	31.13 a	18.86 b	33.84 a	30.17 ab	32.02 a	11.60
Seed number ear-1	298.26 ab	399.22 a	248.14 b	247.74 b	292.11 b	345.31 ab	299.72 ab	102.72
Seed yield m ⁻² (kg)	0.8 ab	1.05 a	0.66 bc	0.4 c	0.85 ab	0.9 ab	0.81 ab	0.37
∑a of the LGS	9	9	6	0	9	9	10	
Rank of the LGS	2	2	3	4	2	2	1	

LGS traits (horizontal data) followed by the same letter were not different at Tukey's HSD_{0.05}

3.3. Dendrogram Analysis

A dendrogram was made based on the correlation values among the lines. Table 2 showed that the lines were separated by the Tukey's HSD into 4 ranks and the dendrogram resulted in five cluster: (1) the Lampung line/P36; (2) Padang/West Kalimantan; (3) Synthetic; (4) Jogjakarta; and (5) Palembang (Figure 1). The farthest distance of the lines marked the Lampung line/P36 to Palembang was 0.29 % may indicate that the LGS had one or more common ancestors in the hybridization of each. The Lampung line (ranked 2 in Table 2) which in fact an LGS was closely related with P36 (ranked 1) one of the popular hybrids planted by the Lampung corn-farmers. The natural cross-hybridization may have improved The Lampung line inasmuch to improve its performance.

The dendrogram analysis gave different result from the mean analyses based on the Tukey's HSD. The statistical actuality may give different perspective to a plant-breeder to choose which line would better recombine to another line.

In the Table 2 the LGS's: Padang, Palembang, Lampung and Synthetic might not recombine well since they were ranked the same. They would be better used as the male parents to recombine with the Jogjakarta and West Kalinantan lines. For themselves, they would better off when recombine with the P36 hybrid or other new hybrids.

On the other hand, the dendrogram informed that the Lampung line would be appropriate to hybridize as male parent with the West Kalimantan and Jogjakarta lines; whereas the lines: Lampung, Padang, Palembang and Synthetic could be hybridized as the male or female parents to each other. The P36 as the newer line could be used as male parent to hybridize with the LGS lines.

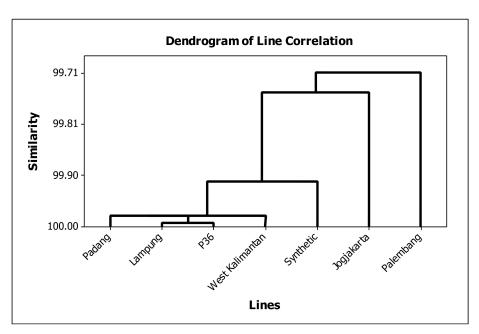


Figure 1. Dendrogram constructed based on the correlations for all traits among lines

4. Conclusions

In testing the Local Genetic Source (LGS) lines collected from Padang, Palembang, Jogjakarta, West Kalimantan, Lampung and Synthetic compared with the P36 one of new released national corn hybrids indicated that the performance of the LGS's was within the range of national standard for traits: plant height, leaf number plant⁻¹, time to tasseling, time to silking, ear length, ear diameter, 100 seed weight, seed number ear⁻¹ and seed yield m⁻². Only the ear diameter would need to increase to 5.0 – 5.5 cm through a recombination with new hybrid varieties. The Tukey's HSD could rank the LSD lines according to the performance of each lines measured of all traits. The performance of lines helped the breeder to rank the lines from the best down to the poor: the hybrid P36 was ranked 1; the Padang, Palembang, Lampung and Synthetic lines were ranked 2; Jogjakarta was ranked 3 and West Kalimantan was ranked 4. On the other hand, the dendrogram differenciated the lines into four groups: the Lampung lines/P36 hybrid; Padang/West Kalimantan; Synthetic; Jogjakarta and Palembang regardless the classification resulted from the Tukey's HSD. Both classification, the Tukey's HSD and the dendrogram would be useful to determine lines to be hybrid parents in a recombination. The Padang, Palembang, Lampung and Synthetic lines could be used as the male parents to recombine with the Jogjakarta and West Kalimantan lines.

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