5. SUMMARY

The aim of the present investigation was to estimate a RAPD fingerprint for each inbred line as well as estimating heterosis, genetic components and their interaction with nitrogen levels for: date of 50% tasseling, 50% silking, plant and ear heights, ear length, ear diameter, no. of rows/ ear, no. of kernels/ row, 100-kernel weight and grain yield/ plant. Also, a comparison between methods of diallel cross analysis was considered for all the studied traits. Nine yellow inbred lines namely: M.1012 (P_1), M. 106 (P_2), M.103 (P_3), M. 100 (P_4), M. 161 (P_5), M. 120-B (P_6), M. 1006 (P_7), M. L-56 (P_8) and M. 313A (P_9), representing wide range of variability in most of the studied traits were used. Three experiments were carried out; the first experiment was laboratory experiment. The Random Amplified Polymorphic DNA (RAPD) techniques and genetic analysis (PCR analysis) were used to determine the genetic diversity among parental inbred lines. The second experiment was a half diallel set of crosses involving nine inbred lines evaluated under two nitrogen fertilizer rates (60 and 120 kg N/ fed) at two separate experiments to study the type of genetic variance and comparing among methods (Griffing M-2, M-4, **Jones 1965 and Hayman 1954.** The third approach was six populations P_1, P_2, F_1, F_2, Bc_1 and Bc_2 of the three crosses i.e P_1xP_5 (high diversity), P_1xP_9(moderate diversity) and P_4xP_9 (low diversity), designated in the text as the first, second and third cross, respectively were studied under 120 kg of nitrogen rate/ fed. The six populations of each particular cross were evaluated

at a separate experiment. Each of the five experiments were grown in a randomized complete block design with three replications at the Agricultural Research and Experimental Station of Fac. of Agric., Moshtohor. Data were recorded on individual plant basis for all studied traits. The data were genetically analyzed by the procedure developed by **Griffing (1956)** Method 2 and 4 model I, **Hayman (1954a and b), Jones (1965), Jinkens (1954) and Allard (1956)** for diallel cross analysis, and **Gamble (1962)** for the six population's analysis. The obtained results could be summarized in three topics as follows:

A- The first experiment

DNA fingerprinting using Random Amplified Polymorphic DNA (RAPD)

a. RAPD polymorphism

1. The results revealed that, nine primers (A12, A13, A14, A15, A17, A18, A19, A20 and Q11) gave polymorphic amplification products scored a total of 397 amplified DNA bands. The number of bands varied from (28) bands for (A15) primer to (71) bands for (A12).
2. Total number of fragments varied from 4 fragments for (A13) to 11 for (A12).
3. Number of polymorphic fragments varied from 2 for A19 to 9 for (A18 and Q11)
4. The nine RAPD primers generated 397 scorable bands across nine inbred lines. These primers produced a total of 66 reproducible fragments, from which 39 (59.05%) were

polymorphic. The size of fragments ranged from 181 bp to 2305 bp.

b. Genetic similarities and clustering.

1. The lowest genetic similarity was (0.55) detected between P1 and P5 also, obtained between P8 and P9. While, the highest genetic similarity was (0.90) scorbed between the two parental inbred lines P4 and P9. The over all mean for genetic similarity between the parental inbred lines was (0.698).
2. The data collectively distinguished two main clusters. The first main cluster consists of eight inbred lines P_1, P_2, P_3, P_4, P_6, P_9, P_8 and P_7 and this cluster separated into two sub-clusters: the first sub-cluster contained six inbred lines P_1, P_2, P_3, P_4, P_6 and P_7. Meanwhile, the second sub cluster contained two inbred line P_8 and P_9. In addition, the first sub-cluster divided into two sub-sub clusters the first sub-sub cluster contained P_1 and P_2. While, the inbred lines P_3, P_4, P_6 and P_7 were belonging to the second sub-sub cluster as well as inbred lines P_4 and P_6 being closely. The second main cluster contains the inbred lines only P_5.

B. The second experiment:

a. Analysis of variance, mean performance and heterosis.

1- Nitrogen rate mean squares were significant for all traits under study except for no. of rows/ear, with mean values in high rate being higher than those in low rate of nitrogen for all traits.

2- Hybrids mean squares were significant for all traits in both rates of nitrogen as well as the combined analysis in both methods of Griffing. Insignificant interaction mean squares

between hybrid and nitrogen rates in both methods of Griffing (M2 and M4) were detected for all traits studied except, days to 50% tasselling, days to 50% sillking and ear height and shelling percentage in M4

3- The parental inbred lines no. 1, 6, 9 and 4 had the highest mean values of ear weight and grain yield/ plant in the combined analysis.

4- None of the hybrids surpassed the late or the highest performing inbred lines for tasseling and silking dates revealing that all hybrids were shifted to wards the earliness direction. The earliness of tasseling and silking dates was detected by crosses P_1xP_3, P_1xP_6, P_1xP_8, P_2xP_3, P_2xP_6, P_3xP_6, P_3xP_7, P_6xP_7 and P_6xP_9.

5- The crosses P_3xP_4, P_4xP_6, P_4xP_5 and P_3xP_8 in the combined analysis, P_3xP_4 and P_4xP_6 in low nitrogen rate had significant superiority over the best check hybrid (S.C. G. 155) for grain yield/ plant

6- Significant mean squares for parents *vs*. hybrids were detected for all the studied traits in both nitrogen rates as well as in the combined analysis as an indication of average heterosis. Insignificant interaction between mean squares due to parent vs. crosses and nitrogen rate were obtained for all traits except days to 50% tasseling, days to 50% silking dates and plant height. The best crosses for earliness were P_3xP_6 and P_3xP_7. As for plant and ear heights, the highest negative heterotic effects were detected by two crosses, P_1xP_7 and P_4xP_7 compared to the two check varieties (S.C. Pioneer 3084 and S.C. G. 155). For grain yield/ plant, the cross P_3xP_4 gave significantly positive

heterotic effects relative to S.C. G. 155 in both nitrogen rates as well as the combined analysis. Also, the crosses P_3xP_4 and P_4xP_6 in low nitrogen rate, P_3xP_4, P_3xP_8 and P_4xP_5 in high nitrogen rate and P_3xP_4, P_4xP_5 and P_4xP_6 in the combined analysis had significant out yielded the best check hybrid (SC G. 155). In addition, the crosses P_1xP_4, P_1xP_6, P_2xP_3, P_2xP_4, P_2xP_8, P_3xP_5, P_3xP_6, P_3xP_7, P_3xP_8, P_4xP_5, P_4xP_8, P_5xP_6, P_5xP_7, P_6xP_7 and P_8xP_9 in low nitrogen rate, P_1xP_4, P_1xP_5, P_1xP_6, P_2xP_4, P_2xP_5, P_2xP_6, P_2xP_8, P_3xP_5, P_3xP_6, P_3xP_7, P_4xP_6, P_4xP_8, P_6xP_7 and P_8xP_9 in high nitrogen rate and P_1xP_4, P_1xP_5, P_1xP_6, P_2xP_4, P_2xP_5, P_2xP_6, P_2xP_8, P_3xP_5, P_3xP_6, P_3xP_7, P_4xP_6, P_4xP_8, P_5xP_7, P_5xP_8, P_6xP_7, P_7xP_8, P_7xP_9 and P_8xP_9 in the combined analysis gave insignificant out yielded the best check hybrid S.C. G. 155.

7- The correlation coefficients between genetic diversity (GD), and each of mean performance and heterosis relative to check variety S.C. G 155 for grain yield/ plant were positive (r = 0.06 and 0.03), respectively. The results indicate that RAPD marker can be used as a tool for determining the extent of genetic diversity among maize inbred lines. The results indicated that RAPD marker can be used as a tool for determining the extent of genetic diversity among maize inbred lines and for genotypes into different groups but when used a large number of primers to detect the variation over all DNA or used a new marker like SSR or AFLP.

b. Estimation of Genetic components

b.1 Griffing method 2 model 1, 1956.

1- The mean squares due to general and specific combing ability were highly significant for all traits except mean square of GCA for shelling% at low nitrogen rate.

2- The ratio of GCA/SCA were equal unity for days to 50% sillking at high nitrogen rate, plant height at low nitrogen rate and the combined analysis, ear length at the combined analysis and no. of rows/ear at the low nitrogen rate. GCA/SCA ratio equal unity was obtained. High ratios which largely exceeded the unity were obtained for days to 50% tasseling at low rate of nitrogen fertilization, days to 50% sillking at low nitrogen rate as well as combined analysis, plant height, ear length and shelling% at high nitrogen rate, ear height at both nitrogen rates as well as the combined analysis, and no. of rows /ear at high nitrogen rate as well as the combined analysis. For the other remain traits, GCA/SCA ratios, were less than unity.

3- The mean squares of interaction between nitrogen rates and both types of combining ability were significant for days to 50% tasseling and silking and ear height and shelling %.

4- As for ear length and ear weight/plant, insignificant mean squares of interaction between nitrogen rates and GCA along with significant SCA x nitrogen rate were detected. On the other hand, insignificant mean squares of interaction between nitrogen rates and both combining ability were obtained for ear diameter, no. of rows/ear, no. of kernels/ row and grain yield / plant.

b-2 Griffing 1956 method 4 model 1

1- The mean squares due to general and specific combining ability were highly significant for all traits except, the mean squares due to SCA for ear diameter and shelling% in the high nitrogen rate and no. of kernels/row in the low nitrogen rate.

2- The mean squares of interaction between rates of nitrogen fertilization and both types of combining ability were insignificant for all studied traits except days to 50% tasseling, dayes to 50% silking, ear height, ear weight/ plant and shelling percentage.

General combining ability effects:

1- The parental inbred line no. 1 exhibited the highest significant negative ($\hat{g}_i$) effects for; days to 50% tassling and silking and ear height plant height in both nitrogen rates as well as the combined analysis in both methods in the two method (G. M-2 and 4).

2- The parental inbred line no. 4 seemed to be a good combiner in both methods of analysis for; no. of kernels/ row, ear weight/ plant and grain yield/plant in both nitrogen rates as well as the combined analysis. However, it exhibited either significant undesirable or insignificant ($\hat{g}_i$) effects for the other traits.

b-3 Specific combining ability effects ($\hat{S}_{ij}$):

As for days to 50% tasseling; the crosses P_1xP_8 and P_2xP_3 in both methods of analysis and P_2xP_6, P_3xP_7, P_5xP_9, P_6xP_7 and P_8xP_9 only in method-2 had the best desirable $\hat{S}_{ij}$ values, in both

nitrogen rates as well as the combined analysis. With regard to days to 50% sillking; the crosses P_2xP_6 and P_3xP_7 in both methods of analysis and P_5xP_7, P_5xP_9, P_6xP_7, P_6xP_9, and P_8xP_9 in method 2 exhibited the highest desirable $\hat{S}_{ij}$ effects in both nitrogen rates and the combined analysis. For grain yield/plant, the crosses P_3xP_4, P_8xP_9, P_1xP_6, P_7xP_9, P_1xP_9, and P_3xP_8 had the highest significant desirable $\hat{S}_{ij}$ effects at both nitrogen rates as well as the combined analysis, in both methods of analysis.

b-4 The half diallel cross (Method of Jones, 1965):

1- The (a) component which primarily testes the significance of additive effects were significance in all traits in both nitrogen rates and the combined analysis. Significant of the component (b) was detected for all traits in both nitrogen rates as well as the combined analysis. The ratio of (a)/(b) was coincided with that obtained from GCA/SCA from Griffing method-2.

2- The (b1) values were significant for all traits in both nitrogen rates and the combined analysis. The significant (b2) values were obtained for all traits in both nitrogen rates as well as combined analysis, except days to 50% tasseling, ear height and 100-kernel weight in low rate of nitrogen. Significant (b3) values were detected for all traits in both nitrogen rates and the combined analysis except ear diameter in high rate of nitrogen and the combined analysis and number of kernels/ row and shelling percentage at low and high nitrogen rates, respectively.

3- The mean squares of interaction between nitrogen rates and both types of additive (a) and non-additive (b) were significant

for days to 50% silking and tasseling and ear height. As for plant height, 100-kernel weight and shelling percentage, the mean squares of interaction between nitrogen rate and (a) component were significant, however, insignificant (b) component by nitrogen rates mean squares were detected. As for ear length, ear weight/ plant and grain yield/ plant insignificant mean squares of interaction between nitrogen rates and (a) component along with significant (b) component with nitrogen rates were detected. On the other hand, insignificant mean squares of interaction between nitrogen rates and both (a) and (b) components were obtained for other traits.

b-5 Hayman's method (1954b):

1-With the exception of 100-kernel weight, ear weight/plant and grain yield/plant in both nitrogen rates, days to 50% tasseling, number of kernels/row and shelling percentage in low rate of nitrogen and ear height in high nitrogen rate, the additive component (D) reached the significant level of probability for all traits. Highly significant values and larger in magnitude for the dominance component (H1) were obtained for all traits in both nitrogen rates. Significant (h^2) values were detected for all the studied traits. The result showed the presence of over dominance for all the studied traits (D/H1).

2- The Values of (H2/4H1) which largely deviate from one quarter were obtained for number of rows/ear and shelling percentage in both nitrogen rates and number of kernels/ row at high nitrogen rate. On the other hand, values of (H2/4H1) were nearly one quarter for other traits. The symmetry vs.

asymmetry in gene frequency was also examined by estimating the (F) component. The same conclusion could be drawn from the corresponding proportion $(4DH1)^{0.5} + F)/ (4DH1)^{0.5} – F)$ and F values.

3- Low heritability values were detected for all the studied traits in both nitrogen rates.

b-6 Graphical analysis:

1- Significant regressions were obtained in all traits (in both nitrogen rates) and the slope of the regression lines significantly from unity. With the exception of shelling percentage in low nitrogen rate, the regression lines were intercept the (Wr) axis below the origin, suggesting over dominance. The array points scattered along the regression line for all traits in both nitrogen rates. The correlation coefficient between parental mean (Yr) and (wr+vr) for each array were significant positive values for days to 50% silking at both nitrogen rates, days to 50% tasselling in high rate of nitrogen and insignificant and high positive value in low nitrogen rate. For the other traits, significant negative correlation estimates were obtained. The parental inbred lines P_1 and P_7 for ear weight and grain yield contained the most dominant genes responsible for the expression of both traits. Meanwhile, both inbred lines P_3 and P_8 seemed to carry the most recessive ones in both nitrogen rates.

2- For shelling% the parental inbred lines P_1 and P_4 in low nitrogen rate and P_7 and P_9 in high nitrogen rate contained the most dominant genes. However, P_5 and P_7 in low and P_3 and P_2

in high nitrogen rates, seemed to contain the most recessive genes.

b.7 Analysis of genetic-environmental interactions:

1- For plant height, ear length and shelling% mean squares of parents and parent by nitrogen rate were found to be significant. However, insignificant parent and parents x nitrogen rates mean squares were detected for 100-kernel weight and ear weight. On the other hand, the other traits significant mean squares associated with parents along with insignificant mean squares associated with parents by nitrogen rates mean squares were detected.

2- Significant nitrogen rates (N) mean squares were detected for days to 50% tasseling, ear diameter, no. of kernels/ row and 100-kernel weight. However, the other traits, insignificant nitrogen rates mean squares were detected. The dominance mean squares were significant for all traits. Significant mean squares associated with dominance by nitrogen rates interaction was found for days to 50% tasseling, number of kernels/row and 100-kernel weight.

3- Significance of the array mean squares were detected for all traits except; days to 50% tasseling and sillking, ear length and shelling percentage.

4- Insignificant variance of array x nitrogen were obtained for all traits, indicating that the position of the parental arrays on the graphs were constant in different nitrogen rates. Significance of the dominance x arrays was obtained for all traits except days to 50% tasseling and silking and shelling percentage.

5- The insignificance of the dominance x arrays x nitrogen rates components of variation were detected in all traits except plant height.

b.8 Comparison between methods of analysis:

b.8.1-Comparison between combining ability effects (M2 and M4 of Griffing)

The correlation coefficient between ($\hat{g}_i$) and ($\hat{S}_{ij}$) effects in two methods (Griffing's method-2 and 4) was significant for all traits.

b.8.2-Relative efficiency of the four methods Griffing M2 and M-4, Jones 1965 and Hayman1954)

Additive

1- The F. test between the methods used in this study revealed that Griffing's method-2 and 4 and method of Jones reached similar results.

2- The correlation coefficient of additive effects in four methods for all traits used in this study was highly significant between the four methods i.e. Griffing's method-2 and 4, **Jones 1965 and Hayman 1954b**

3- Insignificant F. test for additive x nitrogen interaction was obtained between the four methods (Griffing's method-2 and4, **Jones 1965** and **Allared 1956** used in this study for most traits.

4- Significant correlation values of additive x nitrogen interaction between four methods except between Griffing method 4 and **Allard 1956** was obtained between Griffing's method-2 and 4, **Jones 1965 and Allard 1956.**

Non additive

1- For comparison between the four methods (i.e G2/G4, G2/H, G4/J, G4/ Hayman and J/ Hayman), the results indicated that significant F. test was detected for all traits in both nitrogen rates as well as the combined analysis except Griffing Method 2 and **Jones 1965** in all traits in both nitrogen rates as well as the combined analysis, where M-2 did not differ than **Jones 1965.** Also, insignificant F. test was obtained between **Hayman 1954** and each of Griffing M-2 and **Jones 1965** for 100-kernel weight in the combined analysis, indicating that method of **Hayman 1954** and each of Griffing M-2 and **Jones 1965** was similar in the combined analysis for 100-Kernel weight.

2- The correlation coefficient of non-additive effects derived from four methods of diallel analysis over all traits in both nitrogen rates as well as the combined analysis were highly significant.

3- The efficiency of the four diallel methods of analysis for non-additive x nitrogen interaction based on F. test was obtained between the four methods (Griffing m-2, m-4, Jones and Allard) used in this study for all traits except days to 50% tasseling and sillking, plant height, ear height, ear diameter, on. of rows/ear, no. of kernels/row, ear weight/plant, grain yield/plant and shelling%. Where, method of Allared was differed from the other methods i.e. Griffing m-4, m-2 and Jones. This result indicating that the three methods of analysis (Griffing method-2 and -4 and Jones reached similar results.

4- Significant correlation values of non-additive x nitrogen and SCAxN/SCA between four methods were obtained for Griffing's method-2 and 4 and Jones.

Error

For error mean squares insignificant F. test was detected between Griffing's method-2 and 4 for all traits except days to 50% tasseling in the combined analysis and shelling percentage in high nitrogen rate and the combined analysis. Insignificant F. test between the four methods was obtained for most traits. The correlation coefficient values of error mean squares between the four methods of all traits were significant.

C. Six population's analysis. (Experiment III).

1- Significant genetic variance was detected for all traits in the three crosses and therefore, other genetical parameters were estimated.

2- Highly significant negative heterotic effects for days to 50% tasselling and silking in the three crosses. Highly significant positive heterotic effects were detected for all the other traits.

3- The results indicated that (P) values exceeded the unity in all cases. Over dominance towards the lower parent was detected for days to 50% tasselling and silking suggesting that earliness dominated lateness. Over-dominance towards the higher parent was detected for all other trait under test.

4- Inbreeding depression was significant negative for no. of days to 50% tasselling and silking in the three crosses. Meanwhile, significant positive inbreeding depression was detected for other cases except shelling percentage in the first cross.

5- Significant F2 deviation (E1) and backcrosses deviation (E2) were obtained for most traits.

6- The additive gene effects (a) were significant for no. of days to 50% silking, ear height, ear weight/plant, grain yield/ plant and shelling percentage in the three crosses, no. of days to 50% tasseling in the first and third cross, plant height and 100-kernel weight in the first and second cross, ear length and no. of rows /ear in the third cross and ear diameter in the first cross. The dominance gene effect (d) was highly significant for all traits in the three crosses except the shelling percentage in the third cross. Additive x additive (aa) epistatic type of gene action was significant for all traits, except ear height and ear length in the first cross. Also, additive x dominance gene effects were significant for all traits except no. of rows/ear in the three crosses, days to 50% silking in the first and second cross, no. of kernels/row in the first and third cross, and ear length and shelling percentage in the second cross. Dominance x dominance gene effects were significant for all traits in the three crosses except shelling % in the third cross.

7- High genetic coefficient of variation was detected for ear diameter, no. of kernels/row, ear weight/plant, grain yield/plant and shelling percentage in the three crosses. However, moderate values were obtained for plant height, ear height, no. of rows/ear and 100-kernel weight, in the three crosses. Days to 50% silking and tasseling in the three crosses, had low values of G.C.V. %.

8- High heritability value in broad sense in the three crosses except 100-kernel weight. For the exceptional case, moderate

heritability values were obtained in the three crosses. High heritability in narrow sense was detected for ear diameter and shelling percentage in the three crosses, no. of rows/ear, days to 50% tasseling and silking in the first cross and ear weight/ plant and grain yield/plant in third cross. Moderate to low heritability values in narrow sense were detected for the other cases in the three crosses. For days to 50% tassling and no. of rows/ear in the first cross and ear diameter in the second cross, heritability values in narrow sense were high in magnitude and nearly equal its corresponding value in broad sense. For other cases in the three crosses narrow sense heritability values were much lower than those of broad sense.

9- With the exception of days to 50% tasseling and silking in the second and third cross and plant height in the second cross, the results indicated that the predicted genetic advance expressed as the percentage of the mean was moderate to high for all the studied traits. For the exceptional cases, low GA% was low.