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# Sequential Path Analysis for Evaluating Interrelationships of Yield and Yield-Related Components in Cameroon Highland Composite (CHC202) Maize (*Zea mays* L.)

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**Abstract** This study evaluated the interrelationship between some growth and yield parameters of maize under different fertilizer regimes. The experiment was conducted in Bali Nyonga (North West Region, Cameroon) in 2014. The Cameroon Highland Composite (CHC2020) maize was used under four fertilizer regimes of NPK (20:10:10) at 0 g, 4 g, 8 g and 12 g per plant in a complete block design. Correlation and sequential path analyses were conducted. All variables measured were significantly affected by different fertilizer regimes except the number of plant at harvest ($p$ = 0.538). high correlations were observed between plant vigour (PV) and weight of harvested maize (WHM) (r = 0.543, $p$< 0.0001), number of plants at harvest (NPH) and WHM (r = 0.626, $p$< 0.0001) and plant emergence (PE) and NPH (r = 0.675, $p$< 0.0001). The sequential path analysis (SPA) revealed that the variables could be grouped in to first- and second-order variables. The NPH and PV were first-order variables (*Adj* $R^2$ = 0.654, $p$< 0.0001). The direct effect of plant height (PH), stem diameter (SD) and leaf surface area (LSA) had very small effect on WHM using conventional path analysis (CPA) but had a significant effect via other variables using SPA. The tolerance values (< 0.10) and the variance inflation factor (> 10) for all variables were within acceptable limits for path analysis. This study proposes that PV, NPH and PE as criteria variables for breeding and genetic enhancement of CHC2020 maize. The study also emphasizes the utility of sequential SPA over CPA.

**Keywords** breeding, correlation, genetic, stepwise, tolerance, variance inflation factor

## Introduction

Maize (*Zea mays* L.) is an important cereal all over the globe, serving as the most important food staple for many in central and south America, Africa and China [1-2]. Not only does maize provide food for humans, it also plays a pivotal role in animal feed, brewery and biogas production [2]. The wide uses of maize have sparked varied research interest over many decades with the ultimate goal of increasing production. Researches have been focused on optimizing fertilizer use [3-5], plant density [6-8], pests, disease and weed management [9-12], new varieties [13] and maize intercrop [14-15]. Nevertheless, maize yield is a complex quantitative character which is associated with various interrelated traits. Knowledge of these interrelationships between maize yield and is associated component traits will improve breeding schedules by the use of suitable selected indices [16]. Correlations and path analysis play important roles in elucidating cause and effect relationships between yield and some yield components [17]. For many decades, path analysis has been used in breeding programs to ascertain the relationships between grain yield and its contributing components with the principal objective of identifying those components with significant effects on yield for use as bases for selection [18-20]. In other words, path analysis is a reliable statistical tool which quantifies the interrelationship of various grain yield components and indicate if the influence is directly reflected in the yield or through some other paths, that is, path coefficient analysis shows the extent of direct (path coefficient) and indirect (effect exerted through other independent variables) effects of the causal characters on response character. In many studies involving path analysis, investigators considered the predictor trait as first-order variables to analyze their effects over a response (dependent) variable such as yield [21-22]. Samonte *et al* [20] adopted a sequential path analysis by arranging and analyzing various predictor variables in first-, second-, and third-order path due to multicollinearity for variables, particularly if correlations among some variables were high. The current study was conducted to determine interrelationships among maize yield and related components in order to select characters with high effect on yield in the highlands of Cameroon using CHC202 maize, developed for this part of the country under different nitrogen regimes. The Cameroon Highland Composite (CHC) maize variety was developed by IRAD (Institute of Agricultural Research and Development) Bambui to adapt to highland zones in Cameroon for seed producers. The CHC maize are open-pollinated varieties best suited for use by small-scale farmers who can use the seeds from their own farms for three cycles before buying new seeds [23]. The improvement of such varieties is an ongoing process, thus the need for such a study. It is hypothesized that CHC202's performance in different nitrogen concentrations (following farmers' practices) will elucidate all possible effects (direct and indirect) between yield characters and yield related components relevant to make sound judgments about the breeding schedule of this variety in the region.

## Materials and Methods

### Experimental Site

This experiment was conducted in Bali Nyonga, a village located in Bali sub- Division, North West Region of Cameroon. Bali is rich with antiques that date back to colonial days in Cameroon. Bali lies west of Bamenda, the Capital City of North West Region and it has a population of about 500,000 people (2014 census). The geographical coordinates of Bali are 5053 '0' North, 10 '0' Fast with a humid tropical climate. Annual rainfall ranges between 5.8mm to 10.4mm with average temperatures between 17.0 ºC - 27.0 ºC. The principal activity of the inhabitants was agriculture: chief crops grown included maize, beans, potatoes and vegetables. This research was conducted in 2014 from March to July. Site and soil preparation took place in March; planting in late March harvesting took place in July. Average humidity was 72.2%.

### Experimental Design, Field Layout and Cultural Practices

A randomized complete block design with four blocks and four fertilizer treatments (0g, 4g, 8g and 12g perplant) were used in this experiment. The fertilizer used was a commercial NPK composite in ratio of 20:10:10. There were four blocks, each with a surface area of $38.2m^2$. Each block was divided into raised beds (3 m x 0.4 m x0.4 m). These dimensions were developed based on farmer's recommendation. Each block was separated by a 1.5 mgap. There were two rows with 12 holes of maize per raised bed. Intra-row and inter-rows pacing were 25 cm and 60 cm, respectively. Two maize seeds were planted per hole. Maize seeds were sown on the 25$^{th}$ of March 2014 after two consecutive heavy rain falls. Two fertilizer applications were made in the experiment; on the day of so wing and five weeks after emergence. On the day of sowing, the fertilizer was applied in shallow trenches mixed with soil and the maize seed 3 cm deep. The maize was thinned to 1 plant per hole two weeks after germination. Five weeks after germination, the second application of fertilizer was made: fertilizer was applied in a ring manner around and 4 cm away from the plant. Insect pests and disease were not a major problem in the field: thus no control measure was applied. However, hand weeding and hoeing were done when necessity in all blocks

**Data Collection**

Data was collected on the vegetative stage and at harvest. Data was collected on agronomic (plant emergence, plant height, stem diameter, leaf area index, plant vigour and number of plants at harvest) and yield (weight of harvested maize) parameters. Data was collected using the method described by [5].

**Data analysis**

The data obtained was subjected to normality test and homogeneity of variance using Shapiro-Wilk test and Levene's test, respectively. Analysis of variance (ANOVA) and descriptive statistics (mean and standard deviation) of pooled data across the fertilizer treatments were computed. Correction coefficients were calculated for all possible pairs of variables observed using the Pearson correlation coefficient. A preliminary analysis was done using traditional pat model [24] in which all yield-related characters (all measured characters except weight of maize harvested) were considered as first-order independent variables with weight of maize as the dependent variable. A sequential stepwise multiple regression was performed to group the independent variables into first- and second-order paths depending on their respective contributions to total variation in weight of maize harvested and minimal collinearity. Two common measures, namely, 'tolerance' value and the 'variance inflation factor' (VIF) were used to measure the level of multicollinearity as proposed by [25]. Tolerance value is the amount of variability of the selected independent variable not explained by other independent variables (1 – $R_i^2$, where $R_i^2$ is the coefficient of determination for the prediction of variable *I* by the prediction of variables). The variance inflation factor (VIF = $1/1\text{-}R^2$) represents the extent of effects of the other independent variables on the variance of the selected independent variable. Thus, very large VIF values (above 10) and very small tolerance values (much lower than 0.1) demonstrate high collinearity [25]. The statistical package for social sciences (SPSS ver. 23) was used to conduct all the analyses. A path diagram was drawn using LISREL 8.54 (Scientific Software International Inc.) statistical package.

**Results**

**Descriptive statistics and mean square values of some growth and yield parameters of maize**

The means and standard deviations of measure parameters and means square values, F and *p* values from the ANOVA table are shown in table 1. For all the parameters measured, the effect of different fertilizers was significant ($p < 0.05$) except for the number of plant at harvest ($p = 0.538$). In this study, we hypothesized that different fertilizer levels will affect the maize plant differently as was confirmed by the ANOVA. Heterogeneous response of maize to different fertilizer region is highly reported [3, 5, 26-29]. This implies that the data used for this study covers a broad spectrum of phenotypic response of maize relevant for a path analysis.

**Table 1:** Summary Statistics and mean square values for some growth and yield parameters of maize under different fertilizer regimes

| Parameters | Means | Standard deviation | Mean squares | F | *P* - values |
|---|---|---|---|---|---|
| Weight of harvested maize (kg) | 1.39 | 0.67 | 1.7 | 9.53 | .002 |
| Plant height (cm) | 207.05 | 45.40 | 19395.5 | 16.39 | .0001 |
| Stem diameter (cm) | 2.19 | 0.401 | 1.28 | 11.55 | .0001 |
| Leaf surface area | 495.14 | 186.75 | 460130 | 26.70 | .0001 |
| Plant vigour | 2.96 | 1.16 | 6.511 | 32.45 | .0001 |
| Number of plants at harvest | 7.86 | 2.57 | 3.83 | 0.76 | .538 |
| Plant emergence | 14.54 | 7.15 | 32.73 | 3.732 | .042 |

**Correlation coefficients of seven variables of CHC202 maize**

The correlation coefficients of yield and some yield-related parameters of CHC202 maize grown under different fertilizer regimes are provided in table 2. The pairwise correlation matrix reveals that some correlations were significant and others were not significant. The correlation coefficient between WHM and PV ($r = 0.543$, $p< 0.0001$), WHM and NPH ($r = 0.626$, $p< 0.0001$), NPH and PE ($r = 0.675$, $p< 0.0001$) were high, positive and significant. A significant negative correlation coefficient ($r = -0.232$, $p< 0.0001$) was observed between PV and

PE. Other negative correlations, although not significant (*p*> 0.05) was observed between WHM and SD (r = -0.073, *p* = 0.204), PH and PV (r = -0.025, *p* = 0.422), PV and SD (r = -0.056, *p* = 0.110), NPH and SD (r = -0.056, *p* = 0.331), and PE and SD (r = -0.023, *p* = 0.418). The correlation between PH and WHM was not significant (r = 0.065, *p* = 0.305). Our finding is in contrast with that [30] who recorded a positive correlation between PH and maize yield (r = 0.682, *p*< 0.05). Hannachi et al. [31] reported that PH was not significantly correlated with yield of durum wheat (*Triticum durum* Desf.) under rainfed conditions. On the other hand, [32] reported grain yield of wheat was positively correlated with plant height. The deviation in the correlation coefficients for PH and yield in wheat between [32] and [31] in maize as in the case of the current study and that of [30] are common observations in correlation analyses. Anwar et al. [33] reported a positive and significant correlation between PH and yield while [34] in their study observed a negative and no significant correlation. These deviations are because simple Pearson correlation merely look at how a pair of traits change together [35]. Correlation analysis is a useful tool in plant breeding. However, it fails to bring out the direct and indirect effects of a trait through other traits. Thus, selection of a trait based solely on simple pairwise correlation coefficients without regard for interactions among yield and yield components is erroneous and might mislead breeders [36].

**Table 2:** Pairwise correlation coefficients of seven variables of CHC202 maize grown under different fertilizer regimes.

| Parameters | WHM | PH | SD | LSA | PV | NPH | PE |
|---|---|---|---|---|---|---|---|
| WHM | 1.00 | ns | ns | ns | *** | *** | ** |
| PH | 0.065 | 1.00 | *** | *** | ns | ns | ns |
| SD | -0.073 | 0.420 | 1.00 | *** | ns | ns | ns |
| LSA | 0.177 | 0.407 | 0.397 | 1.00 | ns | * | ns |
| PV | 0.543 | -0.025 | -0.156 | 0.033 | 1.00 | ns | * |
| NPH | 0.626 | 0.083 | -0.056 | 0.205 | 0.003 | 1.00 | *** |
| PE | 0.303 | 0.059 | -0.026 | 0.070 | -0.232 | 0.675 | 1.00 |

WHM – weight of harvested maize, PH – plant height, SD – stem diameter, LSA – Leaf surface area, PV – plant vigour, NPH – number of plant at harvest, PE – plant emergence. Blue colour represents non-significant (ns) correlation coefficients. Red colour represents positive and significant correlation coefficients. Yellow represents negative and significant correlation coefficients. * - *p*< 0.05, ** - *p*< 0.01, *** - *p*< 0.001

**Estimation of direct effects of predictor variables on weight of harvested maize and measure of collinearity in Model 1**

Estimation of direct effects by conventional path analysis, where all the yield-related components were considered as first-order variables (Model 1) with WHM as the dependent variable, and analysis of multicollinearity are shown in table 3. The path model was significant (*Adj* $R^2$ = 0.654, *p*< 0.0001, Durban-Watson = 1.621). All predictor variables exerted a positive direct effect on WHM. The highest direct effect om WHM was from NPH (0.611), followed by PV (0.551). The direct effect of the other variables was relatively lower compared to those of NPH and PV: PH (0.004), SD (0.040), LSA (0.015) and PE (0.019). No negative direct effect on WHM was observed. In a related study conducted by [16], negative direct effects on grain weight per ear was recorded from some maize grain-related characters. Contrary to the current study, [16] recorded a negative direct effect of PH (-0.01) on weight of grain per ear. In addition, they recorded negative direct effects from number of kernel per row (-0.07), and kernel thickness (-0.24) on grain weight per ear. The contrast in the orientation (+ or -) of the direct effect of PH in the current study and that of [16] indicates that orientation of direct effect is variety dependent. The two measure of multicollinearity (tolerance and variance inflation factor) in the current study were within acceptable limits for a path analysis. According to [25], high collinearity is observed when the VIF value is very large (> 10) and the tolerance value is very small (much smaller than 0.1). This was not the case in the current study. It should be mentioned that variables with high collinearity indicate inconsistent patterns of relationships among variables [16]. In such cases, these variables should be removed from the sequential path analysis as was the done by [37].

**Table 3:** Direct effect of first-order predictor variables on weight of harvested maize and measures of collinearity in Model 1 (all predictor variables as first-order variables)

| Predictors | Direct effect | Tolerance | VIF |
|---|---|---|---|
| Plant height | 0.004 | 0.750 | 1.330 |
| Stem diameter | 0.040 | 0.717 | 1.394 |
| Leaf surface area | 0.015 | 0.720 | 1.388 |
| Plant vigour | 0.551 | 0.870 | 1.150 |
| Number of plants at harvest | 0.611 | 0.488 | 2.048 |
| Plant emergence | 0.019 | 0.485 | 2.062 |

**Tolerance, variance inflation and direct effects of the predictor variables (Model 2 – predictors grouped into first- and second-order variables)**

The tolerance, the VIF, and direct effect of predictor variables in model 2 (predictors grouped into first- and second-order variables) are shown in table 4. The sequential path analysis revealed that the variables in the study can be grouped into first- and second-order variables. Number of plant at harvest (NPH) and PV as first-order variables accounted for about 67.5% of the variation in WHM and both the predictor variables also displayed a high and positive direct effect (NPH = 0.622 and PV = 0.541) on WHM. The path analysis of second-order variables over the first-order variables showed that about 44.7% of the total variation for NPH was explained by one character, PE. No direct effect on PV in the path analysis of second-order variable was observed. In [16], first-, second-, and third-order variables were observed while [38], recorded as high as fifth-order variables. In [16], 100-grain weight (100GW) and TNK (total number of kernels per ear) were the first-order variables. In a related study, [38] reported that kernel per ear (KE) and kernel length were first-order variables, accounting for about 46% of the variation in grain yield. It should be noted that both [38] and [16], as well as [39] and [40] worked almost entirely with ear-related (e.g kernel) variables with less or no emphasis on growth parameters. However, it is important to mention that variables other than ear-related, which contribute to above grown biomass (e.g PH, LSA and SD) play an important role in grain yield [41-43]. Thus, a path diagram guided by sequential path analysis will be informative in achieving high yielding maize varieties in genetic enhancement programs.

**Table 4:** Tolerance and variance inflation factor (VIF) values for the predict variables in Model 2 (predictors grouped into first- and second-order variables)

| Response variables | Predictor variables | Dubin-Watson | Adjusted $R^2$ | Direct effect | Tolerance | VIF | Grade |
|---|---|---|---|---|---|---|---|
| WHM | NPH | 1.647 | 0.675 | 0.626 | 1.0 | 1.0 | 1 |
| | PV | | | 0.541 | 1.0 | 1.0 | |
| NP | PE | 2.099 | 0.447 | 0.675 | 1.0 | 1.0 | 2 |

**Path diagram for yield and yield-related characters of CHC2020 maize**

The path diagram in figure 1 shows the interrelations between WHM and other related components. The path coefficients are standardized values. The direct effect of NPH (0.59) and PV (0.55) on WHM are high as indicated by sequential path analysis (Table 4). Stem diameter and PE had a negative effect on WHM via PV, contrary to the low and positive direct effect on WHM as shown in model 1 (Table 3). The path diagram also revealed that LSA had a significant indirect effect (0.13) on WHM through PV compared to low direct effect (0.015) to WHM as seen in model 1. In contrast to model 1 and model 2, path diagram provided a better understanding among various variables and their relative contribution to WHM. Asghari-Zakaria et al. [37] also reported that variables such as stem diameter did not show any significant direct effect on potato tuber yield using conventional path analysis, however, with the aid of sequential path analysis and path diagram, stem diameter showed a significant positive effect on potato tuber yield through other variables (weight per tuber and plant height).

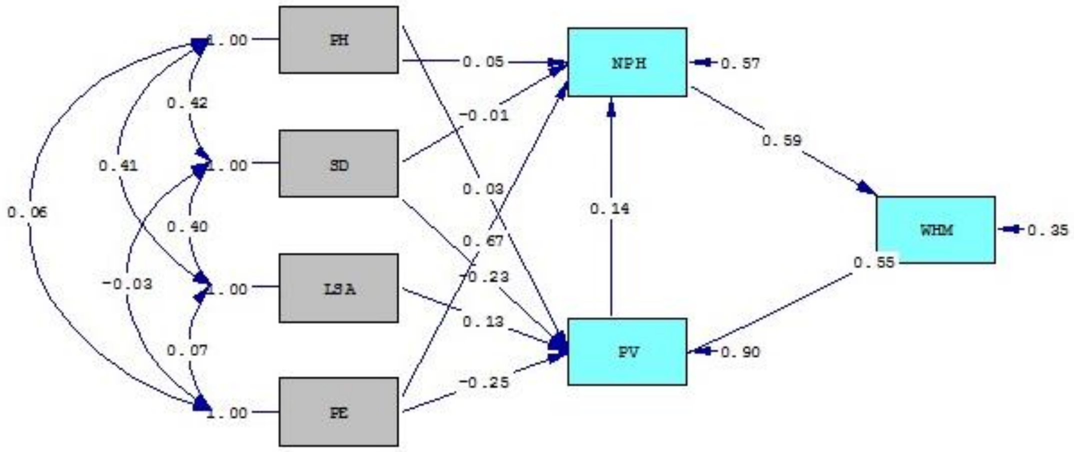


*Figure 1: Sequential path model illustrating interrelationships among weight of harvested maize and related characters*

**Conclusion**

Multiple statistical procedures have been explored to understand the interrelationship among maize yield and yield related variables. This study deviates fundamentally from earlier studies which explore the utility of path coefficient analysis for estimating direct and indirect effects of various characters (particularly ear-related) and maize yield in that, it considers other above ground biomass contributing characters such as plant height, leaf surface area, plant emergence and plant vigour. It was observed that plant vigour and number of plants at harvest had great direct effect on yield as first-order variables. Stem diameter, plant height, leaf surface area and number of plants emerging which had very small insignificant direct effect on maize yield using conventional path analysis was shown to influence yield significantly via other variables. Our study illustrates the usefulness of sequential path model over conventional path model in differentiating between direct and indirect effects of various yield-related variables. In summary, breeding programs of CHC maize should focus on plant vigour and number of plants at harvest. In addition, there is a caveat regarding the applicability of the models in this study since path models are sensitive to germplasm and different production conditions.

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