*Original Research Article*

Sequential Path Analysis of Agro-morphological Traits in Pearl Millet [*Pennisetum glaucum* (L.) R. Br.]

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ABSTRACT

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| In the pearl millet breeding program, studies of relationships among the traits at phenotypic and genotypic levels are useful to select desirable traits directly related to seed yield. Using path coefficient analysis, the present study was performed to estimate the phenotypic, genotypic and environmental correlation coefficients between agro-morphological and yield traits and to determine the direct and indirect effects of traits on seed yield per plant.  A total of 67 genotypes of pear millet were studied during the dry and wet seasons of 2023 and 2024 (considered as four environments). The experiment at each season was conducted in randomized complete block design with two replications.  Based on the analysis of variance, pooled results showed that there were positive and highly significant differences (p ≤ 0.01) among the 67 genotypes for all traits. High significant and positive strong correlations at phenotypic and genotypic levels were observed for spike length, seed weight per spike and 1000-seed weigh with seed yield per plant.  According to the sequential path analysis, 1000-seed weight, seed weight per spike and spike length must be prioritized to maximize the production of pearl millet due to its positive correlations as well as direct effects to seed yield. The number of productive tillers per plant had a negative direct effect on seed yield. The combination of correlation and path analysis indicated that improving seed yield in pearl millet can be effectively achieved by focusing on seed weight per spike and 1000-seed weight traits providing valuable foundation for the selection and breeding of high-yielding pearl millet genotypes under the different seasons for increasing pearl millet production and for future research. |

*Keywords: Seed yield, Correlation coefficient analysis, Sequential path analysis, Pearl millet.*

1. INTRODUCTION

Pearl millet [*Pennisetum glaucum* (L.) R. Br.] which is a cross-pollinated and warm-season crop ranks as the sixth most important cereal grain globally. It is a vital crop, sustaining over a third of the world's population (FAO, 2024). Pearl millet is a nutrient-dense staple food grain specifically also utilized as a feed, fodder, construction material and as a biofuel source (Singh and Chhabra, 2018). Therefore, improving pearl millet yield is very important due to its diverse application and its role in global food security and agriculture. A pearl millet improvement program is crucial due to the extensive use of diverse germplasm. It is valuable in crop improvement due to its inherent genetic diversity which provides a rich source of traits for enhancing yield, nutritional value, and resistance to various stresses.

Pearl millet germplasm is the basic raw material to drive crop improvement program. Collection and conservation of millet germplasm provides a continuous supply of raw material for pearl millet improvement. While pearl millet germplasm has been collected and conserved in Myanmar, a small fraction has been exploited for economically important traits. The seedbank at the Department of Agricultural Research, Myanmar conserves 67 millet accessions and limited research has been dedicated to pearl millet germplasm in Myanmar.

Yield is a complicated trait that is influenced by a variety of factors such as polygenes, environment and genetic heterogeneity (Usman et al. 2017). Selecting for grain yield alone is not particularly effective or efficient due to its complexity and interaction with other yield-enhancing traits. Various yield-enhancing characteristics should be considered and then selection based on its components and secondary characters may be more successful and reliable. The study of correlation is to determine the presence or absence of relationships among the different traits used in the investigation. It is a measure to assess the extent and direction of association among the studied traits (Govindaraj et al. 2009). Furthermore, the genotypic correlation coefficient is a measure of the genetic relationship between traits that may assist in selecting which characters should be considered for improvement of seed yield (Patil et al. 2016; Yadav et al. 2020).

The selection of the genotypes based only on correlation may be misleading because its procedures only the mutual association between two variables whereas path coefficient analysis specifies and measures the importance of different components. As noted by Dadarwal et al. (2020) and Swami et al. (2025), path analysis helps in clarifying the causal relationships among traits that influence yield more effectively than simple correlation measures. Genotypic correlation and path-coefficient analyses provide insights into the nature and extent of relationships among traits, facilitating effective selection in breeding. Studies of correlation can also be helpful in breeding programs, although they do not provide enough information about and understanding of the interrelationships of heritable traits. Consequently, this can lead to incorrect information.

Sequential path analysis in pearl millet helps understand the complex relationships between yield and its contributing traits by dissecting the direct and indirect effects of each trait on yield. This method is valuable for identifying key selection criteria for yield improvement in pearl millet breeding programs. It is a statistical method used to understand how traits or variables influence each other over time or in a specific sequence. It helps researchers determine the direct and indirect effects of one variable on another, particularly when the order of these variables is important, such as in plant development. This technique is often used to analyze data where one variable's effect on another is not immediate but rather occurs through a series of intermediate steps (Kozak & Azevedo, 2014).

Perhaps its effects yield directly, or its total influence follows a different path, allowing the contribution of each trait to yield to be identified. This strategy is used as a selection aid for genetic as well as yield improvement in plant breeding programs. The yield traits do not occur independently; rather, they are interconnected and result in seed yield in pearl millet. Sequential path coefficient analysis examines the effects of predictor parameters as first and second-order component variables on a dependent variable such as yield (Usman et al. 2017). It evaluates the connectivity of several yield-relevant characteristics (Haussmann et al. 2012). Therefore, the current study was employed to understand the character association among agro-morphological traits and to estimate the direct effects of various traits on seed yield in pearl millet genotypes.

2. material and methods

**2.1 Experimental Site**

The experiment was conducted at the field of the Department of Agricultural Research (20° 13ʹ N and 96° 38ʹ E at an altitude of 1286 m above sea level), Yezin, Nay Pyi Taw, Myanmar during the dry and wet seasons of 2023 and 2024.

**2.2 Methodology**

A total of 67 pearl millet genotypes (Appendix 1) conserved in the Seed Bank, Department of Agricultural Research (DAR), Yezin, Naypyidaw in Myanmar were characterized during the dry and wet seasons of 2023 and 2024. The experiment was conducted in randomized complete block design with two replications. Each genotype was grown in two rows of 4 meters in length, spaced 45 cm between rows and 10 cm between plants. The recommended agronomic practices and plant protection measures were uniformly applied during the growing season.

**2.3 Data Collection**

A total of 11 agro-morphological traits were recorded based on the descriptors of [Pennisetum glaucum (L.) R. Br.] (IBPGR, 1993). Days to heading were noted from visual observation of planted rows of each genotype. For each trait, ten randomly selected plants from each genotype and replication were used to quantify agro-morphological traits on the basis of individual plants: plant height (cm), stem diameter (cm), total number of tillers, number of productive tillers, spike length (cm), 1000 seed weight (g), seed weigh per spike (g), green fodder yield per plant (g), dry fodder yield per plant (g), seed yield per plant (g). Bartlett’s test was used to assess the homogeneity of variances for all traits across different environments.

**2.4 Data Analysis**

Analysis of variance was computed using (STAR program version 2.0.1) software (https://bbi.irri.org/). Correlation coefficients of the traits were estimated as per method given by Al-Jibouri et al. (1958). The genotypic correlations for all traits were partitioned into components of direct and indirect effects and path coefficients were analyzed using Dewey and Lu (1959). For sequential path analysis, the traits were denoted as first- and second- components based on the two-level relationships.

3. results and discussion

**3.1 Analysis of Variance**

The analysis of variance for all genotypes across environments was shown for all traits (Table 1). Highly significant differences were detected between environments, genotypes and G × E interaction for all traits studied. The extent of the significant differences observed implies that there is a considerable degree of genetic variation among the genotypes evaluated. Any breeding material with high genetic variation has a greater chance of obtaining desirable traits. These significant differences among pearl millet genotypes suggested that they assume genetically difference among the genotypes which are suitable for further breeding program selected to all the characters under study.

**Table 1. Analysis of variance for agro-morphological traits of pearl millet genotypes**

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **SV** | **d.f** | **Mean sum of square** | | | | | | | | | | |
| **DTH** | **PH** | **SD** | **TNT** | **NPT** | **SPL** | **SW** | **TSW** | **GFY** | **DFY** | **SY** |
| Genotype (G) | 66 | 330.72 | 2387.45 | 5.54 | 2.48 | 0.80 | 71.82 | 35.40 | 3.59 | 12381.85 | 4519.37 | 12.41 |
| Pr>F | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* |
| Season (S) | 3 | 1883.22 | 44865.66 | 408.02 | 15.20 | 52.91 | 27.97 | 10.09 | 4.94 | 1557.65 | 175.95 | 12.43 |
| Pr>F | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* |
| G×S | 198 | 19.95 | 508.83 | 2.11 | 0.78 | 0.46 | 2.86 | 2.02 | 0.22 | 17.95 | 68.82 | 0.46 |
| Pr>F | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* | \*\* |
| Pooled Error | 201 | 1.75 | 22.10 | 0.05 | 0.19 | 0.16 | 0.64 | 1.50 | 0.08 | 4.44 | 3.82 | 0.29 |
| CV% | 2.51 | 2.44 | 2.54 | 13.04 | 17.46 | 2.55 | 8.48 | 3.92 | 1.51 | 3.31 | 3.49 |

*Note*: \*\* = significant at 0.01 probability level, *d.f = Degrees of freedom, DTH = Days to heading, PH = Plant height, SD = Stem diameter, TNT = Total number of tillers per plant, NPT= Number of productive tillers per plant, SPL = Spike length, SW = Seed weight per spike, TSW = 1000-seed weight, GFY = Green fodder yield per plant, DFY = Dry fodder yield per plant, SY = Seed yield per plant*

**3.2 Overall Mean Performance of Agro-morphological Traits**

In this investigation, the overall mean and comparison of each evaluated trait are presented in Table 2. In this study all the evaluated traits exhibited considerable variation across the seasons. Within the row values with different letters indicate that the traits performed differently in the environments tested at least significant difference (LSD = 0.05). Days to heading were recorded at ~49 days for dry seasons which were greater than wet seasons of 2023 and 2024. In dry seasons of 2023 and 2024, most of the genotypes took less time to heading which are statistically different from wet seasons. The latest days to heading were recorded for wet season 2023 which was greater than the average value of four environments. Plant height varies depending on the growing seasons. The high plant height was recorded in wet seasons while the shortest plant height (161.11 cm) noted in wet season of 2023 was significantly different from other environments. The stem diameter was wider in wet seasons than in dry seasons. The off-seasons performance for stem diameter was statistically similar, with a mean of 7.25 cm. The performance of stem diameter in the main growing season of pearl millet showed statistically similar, with a mean of 10.29 cm. The number of tillers per plant (total and productive tillers) was highest in wet season of 2024 while the lowest number of tillers per plant (total and productive tillers) was observed in dry season of 2023, suggesting that the development of tillers in pearl millet was favoured during wet conditions due to increased moisture and nutrient availability. The minor variation of spike length was observed across the seasons with the average performance of (7.35 g) suggesting that good seed development was consistent across seasons. Spike length remained relatively stable across seasons with means of 31.35 cm indicated that these traits are less influenced by environments. The main-growing season in both years produced a heavier 1000-seed weight than the off-seasons. The performance of 1000-seed weight remained consistent among the main-growing seasons of pearl millet with a mean of 7.51 cm. The similar performance of 1000-seed weight was shown in off-season, with a mean of 7.20 cm. The three traits; seed weight per spike, green and dry fodder yield showed greater performance in the main- growing season than in off-growing season. The wet season favoured the higher seed yield than the dry season. The wet season of 2023 had the highest seed yield while the dry season of 2023 had the lowest. However, the variation in yield performance is related to the seasonal imbalance of the environmental components. In this study, all traits except plant height exhibited higher mean values in wet or main-growing seasons. The yield traits and biomass accumulation were higher in wet seasons than in dry seasons. The wet seasons may be more favorable for these traits and sufficient moisture in the soil accelerates nutrient uptake by plants, enhancing plant growth and development as well as yield. The other result was also in report with the findings of Solomon et al. (2021) who studied yield performance and stability of pearl millet genotypes across different environments.

**Table 2. Overall mean performance of agro-morphological traits in 67 pearl millet genotypes across the environment**

|  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Traits** | **Dry season,**  **2023**  **Mean ± S.E** | | **Wet season, 2023**  **Mean ± S.E** | | **Dry season, 2024**  **Mean ± S.E** | | **Wet season, 2024**  **Mean ± S.E** | | **Mean** | **LSD** |
| DTH | 49.32 b | ± 1.09 | 56.27 a | ± 0.55 | 49.40 b | ± 1.12 | 55.45 a | ± 0.76 | 52.60 | 1.69 |
| PH (cm) | 203.13 a | ± 1.81 | 165.11 b | ± 0.65 | 202.22 a | ± 3.75 | 199.21 a | ± 4.61 | 192.41 | 5.35 |
| SD (mm) | 7.21 b | ± 0.13 | 10.20 a | ± 0.15 | 7.29 b | ± 0.14 | 10.38 a | ± 0.11 | 8.76 | 0.29 |
| TNT | 3.02 c | ± 0.29 | 3.12 c | ± 0.06 | 3.34 b | ± 0.34 | 3.78 a | ± 0.27 | 3.31 | 0.19 |
| NPT | 1.75 c | ± 0.28 | 2.18 b | ± 0.21 | 2.04 b | ± 0.34 | 3.19 a | ± 0.26 | 2.29 | 0.14 |
| SPL (cm) | 30.71 b | ± 0.43 | 31.60 a | ± 0.30 | 31.37 ab | ± 0.87 | 31.73 a | ± 0.39 | 31.35 | 0.77 |
| SW (g) | 14.13 | ± 1.66 | 14.71 | ± 0.17 | 14.26 | ± 0.64 | 14.63 | ± 0.28 | 14.43 | - |
| TSW (g) | 7.27 b | ± 0.14 | 7.50 a | ± 0.09 | 7.12 b | ± 0.33 | 7.52 a | ± 0.12 | 7.35 | 0.18 |
| GFY (g) | 136.6 | ± 1.09 | 142.91 | ± 0.86 | 137.16 | ± 1.78 | 142.64 | ± 1.61 | 139.83 | - |
| DFY (g) | 58.31 | ± 1.06 | 59.86 | ± 0.45 | 58.01 | ± 2.07 | 60.34 | ± 0.73 | 59.13 | - |
| SY (g) | 26.54 b | ± 0.21 | 27.24 a | ± 0.15 | 26.49 b | ± 0.39 | 27.04 a | ± 0.50 | 26.91 | 0.54 |

*Note: Dry season is the off-growing season and wet season is the main growing season of pearl millet. Within a row, values with different letters indicate that traits performed differently in the environments evaluated with LSD test at p > 0.05.* *DTH = Days to heading, PH = Plant height, SD = Stem diameter, TNT = Total number of tillers per plant, NPT= Number of productive tillers per plant, SPL = Spike length, SW = Seed weight per spike, TSW = 1000-seed weight, GFY = Green fodder yield per plant, DFY = Dry fodder yield per plant, SY = Seed yield per plant*

**3.3 Correlation Coefficient Analysis**

The homogeneity of the variances for all traits across the environments was conducted using Bartlett’s test. Due to the non-significant differences of the variances of most traits, the pooled data analysis was conducted to analyze the correlation and path coefficient among the traits. The genotypic and phenotypic correlation coefficient for seed yield and its components as well as the correlation among the components are presented in Table 3. At phenotypic and genotypic levels, seed yield per plant showed significant and positive correlations with spike length, seed weight per spike and 1000-seed weight indicating that these traits play a critical role in determining the final yield. Similar association results were also observed by Kumar et al. (2022). Days to heading showed significant and positive correlations with plant height, green and dry fodder yield while days to heading showed negative and significant correlations with the number of tillers per plant (total and productive) at both levels.

At genotypic level, the plant height exhibited significant positive correlations with stem diameter and dry fodder yield while significant and negative correlation was observed between plant height and the number of productive tillers per plant at both levels. A similar finding was reported by Bhosale et al. (2021). The green and dry fodder yield were significantly and positively correlated with stem diameter. Fodder yield can be increased through the combination of the late heading, long growth duration, and large stem diameter.

At genotypic level, a significant and positive correlation was observed between the plant height and dry fodder yield which suggesting that increased plant height may enhance biomass production research backed up these finding by Kholová et al. (2012). Both the number of tillers and productive tillers showed negative correlations with days to heading which indicating that early maturity genotypes tend to produce more tillers. This characteristic can enhance potential yield in cultivars suitable for short-duration environments by Deshmukh et al. (2020). In addition, the number of productive tillers per plant was negatively associated with spike length, seed weight per spike and 1000-seed weight. This suggested that increase the number of productive tillers can cause a decrease in the spike length, seed weight and yield. The heavy spikes and large seed size contribute substantially to overall yield making these traits reliable indirect selection criteria. These results have also been reported by Mula et al. (2020). The spike length was positively correlated with seed weight per spike and 1000-seed weight. These results were confirmed by previous findings of Singhal et al. (2023). In this study, the correlation coefficients at the genotypic level are more closely related to the associated phenotypic level in most cases.

The environmental correlations of agro-morphological traits in pearl millet genotypes across environments were displayed in Table 4. In this study, significant and positive correlations were observed in total number of tillers, number of productive tillers, seed weight per spike, 1000-seed weight, green and dry fodder yield with seed yield per plant. The environmental correlation coefficients were lower than the genotypic correlation coefficients indicating that the environmental factors exerted a weaker positive action, than that of the genes, in the phenotypic manifestation of the traits.

**Table 3. Assessment of genotypic correlations (above diagonal) and phenotypic correlations (below diagonal) among agro-morphological traits of pearl millet genotypes**

| **Character** | **DTH** | **PH** | **SD** | **TNT** | **NPT** | **SPL** | **SW** | **TSW** | **GFY** | **DFY** | **SY** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **DTH** | **1.000** | 0.382\*\* | 0.212\*\* | -0.466\*\* | -0.497\*\* | -0.067 | -0.044 | -0.143\* | 0.248\*\* | 0.241\*\* | -0.083 |
| **PH** | 0.182\*\* | **1.000** | 0.347\*\* | -0.134\* | -0.403\*\* | 0.100 | -0.006 | -0.034 | 0.108 | 0.147\* | 0.014 |
| **SD** | 0.096 | 0.074 | **1.000** | -0.121\* | -0.068 | -0.100 | -0.015 | 0.003 | 0.273\*\* | 0.302\*\* | 0.112 |
| **TNT** | -0.205\*\* | -0.040 | -0.045 | **1.000** | 0.765\*\* | -0.184\*\* | -0.108 | -0.110 | -0.119 | -0.172\*\* | -0.081 |
| **NPT** | -0.202\*\* | -0.121\* | -0.059 | 0.619\*\* | **1.000** | -0.138\* | -0.289\*\* | -0.260\*\* | -0.022 | -0.046 | -0.329\*\* |
| **SPL** | -0.066 | 0.086 | -0.098 | -0.099 | -0.067 | **1.000** | 0.189\*\* | 0.249\*\* | -0.187\*\* | -0.144\* | 0.223\*\* |
| **SW** | -0.020 | 0.043 | -0.016 | -0.068 | -0.135\* | 0.158\*\* | **1.000** | 0.942\*\* | 0.088 | 0.109 | 0.705\*\* |
| **TSW** | -0.108 | 0.026 | -0.045 | -0.091 | -0.136\* | 0.197\*\* | 0.796\*\* | **1.000** | 0.019 | 0.037 | 0.723\*\* |
| **GFY** | 0.217\*\* | 0.078 | 0.135\* | -0.061 | -0.008 | -0.170\*\* | 0.083 | 0.022 | **1.000** | 0.987\*\* | 0.118 |
| **DFY** | 0.193\*\* | 0.097 | 0.152\* | -0.065 | -0.011 | -0.106 | 0.085 | 0.027 | 0.960\*\* | **1.000** | 0.115 |
| **SY** | -0.088 | 0.029 | 0.051 | 0.002 | -0.072 | 0.182\*\* | 0.609\*\* | 0.638\*\* | 0.114 | 0.115 | **1.000** |

*Note: \*\*, \* = Significant at 1 % and 5 % levels respectively*

*DTH = Days to heading, PH = Plant height, SD = Stem diameter, TNT = Total number of tillers per plant, NPT= Number of productive tillers per plant, SPL = Spike length, SW = Seed weight per spike, TSW = 1000-seed weight, GFY = Green fodder yield per plant, DFY = Dry fodder yield per plant, SY = Seed yield per plant*

**Table 4. Estimation of environmental correlations among agro-morphological traits of pearl millet genotypes**

|  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Trait** | **DTH** | **PH** | **SD** | **TNT** | **NPT** | **SPL** | **SW** | **TSW** | **GFY** | **DFY** |
| **DTH** | **1.000** |  |  |  |  |  |  |  |  |  |
| **PH** | -0.167\*\* | **1.000** |  |  |  |  |  |  |  |  |
| **SD** | -0.017 | -0.092 | **1.000** |  |  |  |  |  |  |  |
| **TNT** | 0.064 | 0.019 | -0.013 | **1.000** |  |  |  |  |  |  |
| **NPT** | -0.056 | -0.012 | -0.057 | 0.588\*\* | **1.000** |  |  |  |  |  |
| **SPL** | -0.061 | 0.081 | -0.150\* | -0.018 | -0.044 | **1.000** |  |  |  |  |
| **SW** | 0.077 | 0.146\* | -0.023 | -0.040 | -0.075 | 0.007 | **1.000** |  |  |  |
| **TSW** | 0.030 | 0.144\* | -0.120\* | -0.098 | -0.103 | -0.048 | 0.213\*\* | **1.000** |  |  |
| **GFY** | -0.091 | 0.063 | -0.187\*\* | 0.073 | 0.018 | 0.097 | 0.113 | 0.143\* | **1.000** |  |
| **DFY** | -0.144\* | -0.012 | -0.032 | 0.133\* | 0.031 | 0.260\*\* | -0.091 | -0.039 | 0.239\*\* | **1.000** |
| **SY** | -0.113 | 0.076 | -0.017 | 0.142\* | 0.155\* | -0.075 | 0.125\* | 0.234\*\* | 0.164\*\* | 0.123\* |

*Note: \*\*, \* = Significant at 1 % and 5 % levels respectively*

*DTH = Days to heading, PH = Plant height, SD = Stem diameter, TNT = Total number of tillers per plant, NPT= Number of productive tillers per plant, SPL = Spike length, SW = Seed weight per spike, TSW = 1000-seed weight, GFY = Green fodder yield per plant, DFY = Dry fodder yield per plant, SY = Seed yield per plant*

**3.4 Direct and Indirect Effects of Agro-morphological Traits on Seed Yield**

In the current investigation, the path-coefficient analysis was performed to divide the genotypic correlations of various traits with seed yield per plant into their direct and indirect effects. This approach helps clarify the causal relationships among traits influencing yield over simple correlation measures (Kumar et al. 2023). The path analysis at genotypic level divided the association of agro-morphological traits with seed yield per plant into direct and indirect effects in Table 5. According to the path analysis, green fodder yield had the greatest positive direct effect on seed yield per plant, followed by total number of tillers per plant, 1000-seed weight, stem diameter, spike length and seed weight per spike. These traits can be used to develop an ideally effective selection index for improving the seed yield of pearl millet. Contrast result also reported by Patel et al. (2021) who reported similar trait contributions in pearl millet. The number of productive tillers per plant showed negative direct effect on seed yield per plant followed by dry fodder yield, plant height and days to heading. These results were accordance with previous reports of pearl millet research workers findings of Choudhary et al. (2019).

With respect to the yield component traits, the number of productive tillers per plant had a negative direct effect on seed yield per plant and positive indirect effect via total number of tillers per plant indicating the indirect contribution of number of productive tillers to higher seed yield. These results were in accordance with those observed by Deshmukh et al. (2022). Furthermore, the total number of tillers had a positive direct effect on seed yield but a negative indirect effect via the number of productive tillers per plant and other traits, resulting in an overall negative genotypic correlation with seed yield. This suggested complex interactions that increasing total tillers alone can reduce the seed yield without considering tiller productivity. Similar findings were reported by Kumar et al. (2019).

The genotypic correlation coefficients with seed yield varied from strongly and significantly positive to significantly negative. The traits with strong positive correlations and high direct effects including seed weight per spike and 1000-seed weight are essential for yield improvement. On the other hand, seed yield per plant via the number of productive tillers per plant which possesses negative direct effect needs to be carefully managed in breeding programs. This result was similar to the findings of Dadarwal et al. (2020).

**Table 5. Estimate of direct (diagonal) and indirect effects (off diagonal) of agro-morphological traits in pearl millet genotypes**

| **Trait** | **DTH** | **PH** | **SD** | **TNT** | **NPT** | **SPL** | **SW** | **TSW** | **GFY** | **DFY** | **Genotypic Correlation with SY** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **DTH** | **-0.124** | -0.078 | 0.043 | -0.193 | 0.303 | -0.011 | -0.007 | -0.058 | 0.136 | -0.095 | **-0.083** |
| **PH** | -0.048 | **-0.203** | 0.071 | -0.056 | 0.246 | 0.016 | -0.001 | -0.014 | 0.059 | -0.058 | **0.014** |
| **SD** | -0.026 | -0.070 | **0.204** | -0.050 | 0.041 | -0.016 | -0.002 | 0.001 | 0.150 | -0.119 | **0.112** |
| **TNT** | 0.058 | 0.027 | -0.025 | **0.414** | -0.467 | -0.030 | -0.017 | -0.044 | -0.065 | 0.067 | **-0.081** |
| **NPT** | 0.062 | 0.082 | -0.014 | 0.317 | **-0.610** | -0.023 | -0.044 | -0.105 | -0.012 | 0.018 | **-0.329\*\*** |
| **SPL** | 0.008 | -0.020 | -0.020 | -0.076 | 0.084 | **0.164** | 0.029 | 0.100 | -0.103 | 0.057 | **0.223\*\*** |
| **SW** | 0.005 | 0.001 | -0.003 | -0.045 | 0.176 | 0.031 | **0.154** | 0.380 | 0.049 | -0.043 | **0.705\*\*** |
| **TSW** | 0.018 | 0.007 | 0.001 | -0.046 | 0.159 | 0.041 | 0.145 | **0.403** | 0.010 | -0.014 | **0.723\*\*** |
| **GFY** | -0.031 | -0.022 | 0.056 | -0.049 | 0.013 | -0.031 | 0.014 | 0.008 | **0.548** | -0.388 | **0.118** |
| **DFY** | -0.030 | -0.030 | 0.062 | -0.071 | 0.028 | -0.024 | 0.017 | 0.015 | 0.541 | **-0.393** | **0.115** |

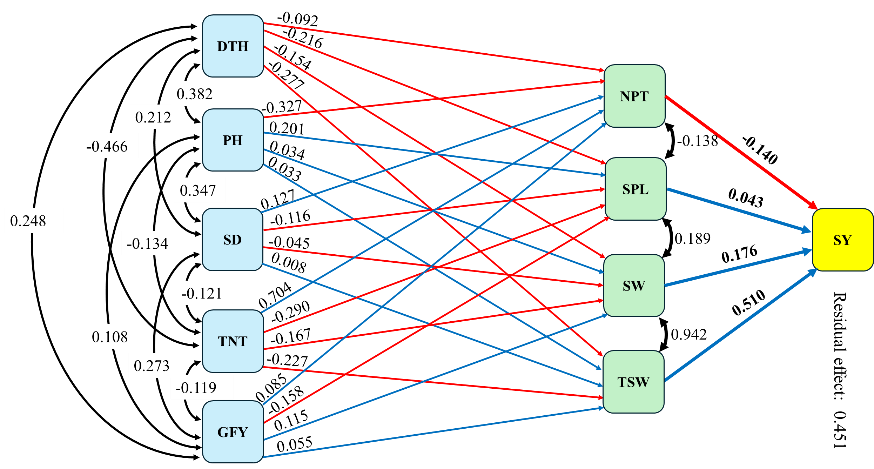
*Residual = 0.34634*

*\*\* = Significant at 1 % level.*

*Note: DTH = Days to heading, PH = Plant height, SD = Stem diameter, TNT = Total number of tillers per plant, NPT= Number of productive tillers per plant, SPL = Spike length, SW = Seed weight per spike, TSW = 1000-seed weight, GFY = Green fodder yield per plant, DFY = Dry fodder yield per plant, SY = Seed yield per plant*

**3.5 Sequential Path Analysis**

The genotypic path coefficient analysis was illustrated in two stages to estimate the association between traits and their cumulative effect on seed yield per plant in pearl millet genotypes. The first order components included vegetative and morphological parameters such as days to heading, plant height, stem diameter and total number of tillers per plant. The four components of yield trait: seed weight per plant, number of productive tillers per plant, 1000-seed weight and spike length, and green fodder yield per plant which were denoted as the major yield determining factors in pearl millet and the second order components. These second order traits served as the ultimate influence on seed yield (SY) by the first order traits as shown in Tables 6 and 7 and Figure 1.



**Figure 1. Genotypic path coefficient diagram representing cause and effect relationships among agro-morphological traits.**

*Path diagram and coefficients of factors on the influence of first order on the second order components and the latter on seed yield (g) per plant of pearl millet genotypes. The single-headed arrow lines in the path diagram reflect the direct effects, whereas the double-headed arrow lines indicate genotypic correlation coefficients. Note: DTH = Days to heading, PH = Plant height, SD = Stem diameter, TNT = Total number of tillers per plant, NPT= Number of productive tillers per plant, SPL = Spike length, SW = Seed weight per spike, TSW = 1000-seed weight, GFY = Green fodder yield per plant, DFY = Dry fodder yield per plant, SY = Seed yield per plant*

**3.6 Effects of First-order Components on Second-order Components**

In the first order component of the path analysis, the seed weight per spike indicated the weak positive direct effect with plant height and green fodder yield but the weak negative direct influence on days to heading, stem diameter and total number of tillers per plant as shown in Table 6. These findings are also reported by Srivastava et al. (2022) who demonstrated that the high statured plant and high green fodder yield in pearl millet are associated with an increase in seed weight per spike and taller plants often have a higher capacity for biomass production which can contribute to greater grain filling and seed development. Green fodder yield is directly related to the overall biomass production by the plant. Higher biomass generally supported the development of larger and heavier seeds. The genetic makeup of different pearl millet varieties in this study determined their potential plant height, fodder yield and seed weight. The number of productive tillers per plant revealed the strong positive direct effect on the number of productive tillers per plant and the significant positive genotypic correlation was observed between these two traits. This relationship is not always straightforward. Several factors such as plant population density, environmental condition, cultivar, sowing date, nutrient and water availability affect both the total number of tillers and the productive tillers produced. Different pearl millet genotypes vary in their tillering capacity and the proportion of tillers that become productive. The weak positive direct effects were observed between the number of productive tillers per plant and other traits such as stem diameter and green fodder yield, respectively. Annamalai (2020) found that the number of productive tillers per plant had positive and significant correlations with number of tillers per plant. The inter-relationship of days to heading and total number of tillers per plant had negative direct effects on 1000-seed weight while the traits; plant height, stem diameter and green fodder yield exhibited the weak positive direct effect on it. Plant height is a significant factor linked to potential yield and increased nitrogen levels can lead to both taller plants and potentially heavier 1000-seed weight. A wider stem diameter is often associated with the taller and more vigorous plants, which could indirectly relate to a higher 1000-seed weight by supporting better plant development and resource allocation for seed production. In path analysis study, Pallavi (2024) also reported days to heading had a direct positive effect on biological yield. In this study, there is a positive direct effect and significant positive genotypic correlation between spike length and plant height. This means that taller plants in the test pearl millet genotypes tend to have longer spikes. The path analysis for the traits revealed that spike length had direct negative effect on days to heading, stem diameter, total number of tillers per plant and green fodder yield. In the tested genotypes of pearl millet, high tillering can lead to short spike on each tiller. This result was also similar by Nehra (2017) who also emphasizes the importance of spike length as a yield component and its relationship with plant height, often showing that longer spikes are associated with taller plants while vegetative traits like stem diameter can have negative or neutral effects on spike length and yield components.

**Table 6. The relationships of first order and second-order components in pear millet**

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Second-order Components** | **First-order components** | **DTH** | **PH** | **SD** | **TNT** | **GFY** | **Genotypic correlation** | |
| **Number of productive tillers per plant** | **DTH** | **-0.092** | -0.035 | -0.019 | 0.043 | -0.023 |  | -0.497\*\* |
| **PH** | -0.125 | **-0.327** | -0.114 | 0.044 | -0.035 |  | -0.403\*\* |
| **SD** | 0.027 | 0.044 | **0.127** | -0.015 | 0.035 |  | -0.068 |
| **TNT** | -0.328 | -0.094 | -0.085 | **0.704** | -0.084 |  | 0.765\*\* |
| **GFY** | 0.021 | 0.009 | 0.023 | -0.01 | **0.085** |  | -0.022 |
| **Spike length** | **DTH** | **-0.216** | -0.082 | -0.046 | 0.101 | -0.053 |  | -0.067 |
| **PH** | 0.077 | **0.201** | 0.07 | -0.027 | 0.022 |  | 0.100 |
| **SD** | -0.025 | -0.04 | **-0.116** | 0.014 | -0.032 |  | -0.100 |
| **TNT** | 0.135 | 0.039 | 0.035 | **-0.29** | 0.035 |  | -0.184\*\* |
| **GFY** | -0.039 | -0.017 | -0.043 | 0.019 | **-0.158** |  | -0.187\*\* |
| **Seed weight per spike** | **DTH** | **-0.154** | -0.059 | -0.033 | 0.072 | -0.038 |  | -0.044 |
| **PH** | 0.013 | **0.034** | 0.012 | -0.005 | 0.004 |  | -0.006 |
| **SD** | -0.01 | -0.016 | **-0.045** | 0.006 | -0.012 |  | -0.015 |
| **TNT** | 0.078 | 0.022 | 0.02 | **-0.167** | 0.02 |  | -0.108 |
| **GFY** | 0.029 | 0.012 | 0.032 | -0.014 | **0.115** |  | 0.088 |
| **1000-seed weight** | **DTH** | **-0.277** | -0.106 | -0.059 | 0.129 | -0.069 |  | -0.143\* |
| **PH** | 0.013 | **0.033** | 0.011 | -0.004 | 0.004 |  | -0.034 |
| **SD** | 0.002 | 0.003 | **0.008** | -0.001 | 0.002 |  | 0.003 |
| **TNT** | 0.106 | 0.03 | 0.027 | **-0.227** | 0.027 |  | -0.11 |
| **GFY** | 0.014 | 0.006 | 0.015 | -0.007 | **0.055** |  | 0.019 |

*\*\*, \* = Significant at 1 % and 5 % levels respectively.*

*Bold letters (Diagonal) indicate the direct effects. Note: DTH = Days to heading, PH = Plant height, SD = Stem diameter, TNT = Total number of tillers per plant, NPT= Number of productive tillers per plant, SPL = Spike length, SW = Seed weight per spike, TSW = 1000 seed weight, GFY = Green fodder yield per plant, SY = Seed yield per plant*

**3.7 Effects of Second-Order Components on Seed Yield**

The effect of the second order component on seed yield is shown in Table 7. According to the path analysis, three traits such as 1000-seed weight, seed weight per spike and spike length had a positive direct association with seed yield. The trait, 1000-seed weight showed the largest value of the positive direct effect on seed yield. Akhare et al. (2023) also reported similar results in their path analysis study on pearl millet genotypes where 1000-seed weight showed a significant positive direct effect on grain yield. The second largest positive and direct contributing attribute was seed weight per spike followed by spike length. Annamalai et al. (2020) found that seed weight per spike and spike length exhibited strong positive correlations and direct effects on seed yield. In this study, the number of productive tillers per plant showed the negative direct effect and genotypic correlation on seed yield per plant. This relationship can be complex and while tillers can contribute to increased yield, an excessive number of tillers can lead to negative effects. When pearl millet plant has numerous tillers, the photosynthetic products are divided among the growing parts of the plant, including tillers. This can result in the less photosynthate being allocated to the development of spikes. There can be a trade-off between the number of productive tillers and the size of the spikes they produce. The pearl millet genotypes which possess high tillering ability can lead to resource competition and reduced allocation of photosynthates to spike and seed development, potentially resulting in negative effects on seed yield. In this study, 1000-seed weight is an important yield attributing trait with the strongest positive direct effect on seed yield in pearl millet.

**Table 7. Effects of Second-order components on seed yield in pearl millet**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Second-order components** | **NPT** | **SPL** | **SW** | **TSW** | **Genotypic Correlation with seed yield per plant** |
| **NPT** | **-0.140** | -0.006 | -0.050 | -0.133 | -0.329\*\* |
| **SPL** | 0.019 | **0.043** | 0.034 | 0.127 | 0.223\*\* |
| **SW** | 0.040 | 0.008 | **0.176** | 0.481 | 0.705\*\* |
| **TSW** | 0.036 | 0.011 | 0.166 | **0.510** | 0.723\*\* |

*Residual are 0.45148*

*\*\* = Significant at 1 % level.*

*Bold letters (Diagonal) indicate the direct effects. Note: SPL = Spike length, TSW = 1000-seed weight, NPT = Number of productive tillers per plant and SW = Seed weight per spike.*

**4. CONCLUSION**

In conclusion, the investigation suggested that 1000-seed weight was the keyattribute among the tested traits in pearl millet due to its strong and positive correlation as well as a high direct effect to seed yield. The seed weight per spike and spike length also showed positive correlation as well as a positive direct effect to seed yield. This implies that above mentioned traits should also be emphasized selection in the case of direct selection for high yielding genotypes. The indirect effects of 1000-seed weight which showed low and positive are also important for the associations with the seed weight per spike, number of productive tillers per plant and spike length indicating that strong positive association was largely due to the direct effect. The 1000-seed weight had the most direct beneficial influence on yield, followed by the seed weight per spike. The number of productive tillers per plant showed a negative direct effect on seed yield. The number of productive tillers per plant with the traits, 1000-seed weight, spike length and seed weight per spike showed negative indirect effects on seed yield. As a result, 1000-seed weight, seed weight per spike and spike length must be prioritized to maximize the production of pearl millet. In general, the combination of correlation and path analysis indicates that improving seed yield in pearl millet can be effectively achieved by focusing on seed weight per spike and 1000-seed weight traits. This research provides valuable foundation for the selection and breeding of high-yielding pearl millet genotypes across diverse agro-climatic conditions and a selection criterion for increasing pearl millet production and for future research.

**DISCLAIMER (ARTIFICIAL INTELLIGENCE)**

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

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Appendix 1. List of pearl millet genotypes and their sources

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| No. | Genotype | Source | No. | Genotype | Source |
| 1 | ICMP-88904 | ICRISAT, India | 35 | ICMP-87101 | ICRISAT, India |
| 2 | ICMP-88130 | ICRISAT, India | 36 | ICTP-8203 | ICRISAT, India |
| 3 | ICMS-7845 | ICRISAT, India | 37 | WCB-77 | ICRISAT, India |
| 4 | ICMS-8021 | ICRISAT, India | 38 | ICH-165 | ICRISAT, India |
| 5 | ICMV-87901 | ICRISAT, India | 39 | ICMS-83506 | ICRISAT, India |
| 6 | ICMS-7703 | ICRISAT, India | 40 | ICH-451 | ICRISAT, India |
| 7 | MC-C-6 | ICRISAT, India | 41 | Variety No.28 | ICRISAT, India |
| 8 | ICMV-81237 | ICRISAT, India | 42 | Kyiywa Sat-1 | ICRISAT, India |
| 9 | ICH-440 | ICRISAT, India | 43 | Kalasat (TaungTha) | TaungTha, Mandaly Region, Myanmar |
| 10 | ICMH-9292 | ICRISAT, India | 44 | ICMH-423 | ICRISAT, India |
| 11 | ICMS-80081 | ICRISAT, India | 45 | Variety No.33 | ICRISAT, India |
| 12 | Sat San Konelun | ICRISAT, India | 46 | ICMS-7835 | ICRISAT, India |
| 13 | Gam-73-K-11 | ICRISAT, India | 47 | NHB-3 | ICRISAT, India |
| 14 | Kalasat(Mahlaing) | Mahlaing, Mandaly Region, Myanmar | 48 | ICMS-8021 | ICRISAT, India |
| 15 | ICTP-8202 | ICRISAT, India | 49 | ICMV-82132 | ICRISAT, India |
| 16 | ICMH-83202 | ICRISAT, India | 50 | SatsanHlesan | DAR, Yezin, Myanmar |
| 17 | ICMH-83401 | ICRISAT, India | 51 | ICH-220 | ICRISAT, India |
| 18 | ICMH-82601 | ICRISAT, India | 52 | ICMS-7835 | ICRISAT, India |
| 19 | IVS-5454 | ICRISAT, India | 53 | ICMD-82113 | ICRISAT, India |
| 20 | LC-7053 | ICRISAT, India | 54 | ICMH-7845 | ICRISAT, India |
| 21 | ICMH-84814 | ICRISAT, India | 55 | ICMH-81814 | ICRISAT, India |
| 22 | NLCH-79 | ICRISAT, India | 56 | ICMS-8004 | ICRISAT, India |
| 23 | IVC-A-82 | ICRISAT, India | 57 | ICMH-81824 | ICRISAT, India |
| 24 | GHB-18 | ICRISAT, India | 58 | ICMV-82113 | ICRISAT, India |
| 25 | ICMS-7857 | ICRISAT, India | 59 | ICMH-9192 | ICRISAT, India |
| 26 | ICMS-7818 | ICRISAT, India | 60 | ICMS-7835 | ICRISAT, India |
| 27 | NELC-P-79 | ICRISAT, India | 61 | ICMV-82123 | ICRISAT, India |
| 28 | ICH-162 | ICRISAT, India | 62 | ICMH-84814 | ICRISAT, India |
| 29 | MBH-127 | ICRISAT, India | 63 | MBH-127 | ICRISAT, India |
| 30 | Kalasat-2 | ICRISAT, India | 64 | Magway millet (Local) | Magway Region, Myanmar |
| 31 | SSC-K-78 | ICRISAT, India | 65 | Nyaung-U millet (Local) | Nyaung-U, Mandalay Region, Myanmar |
| 32 | Pale-1 | ICRISAT, India | 66 | Pearl-3 | DAR, Yezin, Myanmar |
| 33 | ICH-433 | ICRISAT, India | 67 | Pearl-4 | DAR, Yezin, Myanmar |
| 34 | ICMV-83118 | ICRISAT, India |  |  |  |

*Note: DAR = Department of Agricultural Research*