

Full Length Research Paper

Interrelationships among flower yield and related characters in chamomile populations (*Matricaria chamomilla* L.)

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Awareness of interrelationship between flower yield and its components will improve the efficiency of breeding programs through the use of appropriate selection criteria. This research uses sequential path analyses to determine the interrelationships among flower yield and related-traits. 20 chamomile populations were cultivated to determine the important components of flower yield. Flower yield and its components were recorded. Correlation coefficient analysis revealed that flower yield was positively correlated with all the characters except for flower height (FH), days to start flowering (DF) and days to end flowering (DE). Sequential path analysis identified the harvest index (HI) and biological yield (BY) as important first order traits that influenced flower yield. All direct effects were significant as indicated by bootstrap analysis. The results suggest that HI and BY could be used as selection criterion in selecting for increased flower yield in chamomile.

Key words: Chamomile, sequential path analysis, bootstrap analysis, flower yield.

INTRODUCTION

Matricaria Chamomilla is a species of aromatic annual herbs belonging to the family of Asteraceae (Salamon, 1992). Chamomile is a medicinal plant that is native to Europe (Pourohit and Vyas, 2004) and grows widely in various ecological regions of Iran as a wild plant (Rechinger, 1977). Due to the several applications of chamomile in pharmaceutical, nutritional and sanitary industrials, chamomile was one of the most important commercial plants during the recent decades (Wagner et

al., 2005). Flowers contain apigenine which is used as hair color (Böttcher et al., 2001). Chamomile has medicinal properties such as anti-inflammatory (Pourohit and Vyas, 2004), antispasmodic, antiseptic and therapeutic use (Franke and Schilcher, 2006) and anti-microbial (Letchamo and Marquard, 1992).

The main useable part of chamomile is its flowers. Medicinal value of this plant is for active substances, mainly accumulated in the flowers (McGuffin et al., 1997; Gardiner, 1999). Production of stable quality and quantity of flowers is the most important objective in chamomile breeding, which makes it necessary to breed genotypes with high yield and quality.

Selection between and within populations for a high level of flower yield with high quality depends on the selection criteria to be employed by chamomile breeders. Flower yield is a complex trait determined by several components and highly affected by environmental factors. It can be directly or indirectly affected by changes in yield-contributing characters. It is important to examine the contribution of each of the different components to

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Abbreviations: FH, Flower height; FD, flower diameter; PH, plant height; NF, number of flower per plant; NB, number of branches per plant; NS, number of sepals per flower; DF, days to start flowering; DE, days to end flowering; BYP, biological yield per plant; FYP, flower yield per plant; BY, biological yield; FY, flower yield; HI, harvest index; VIF, variance inflation factor.

give attention to those having the greatest effect on flower yield. Therefore, information on the correlation of plant traits with flower yield is a great importance to a breeder in selecting a superior genotype. Plant breeders prefer yield components that indirectly increase yield (Yasin and Singh, 2010). In chamomile, flower yield is correlated with several traits including number of flower per plant, plant height (Zeinali et al., 2008), number of branches per plant and fresh flower weight can increase yield (Letchamo, 1993; Jamshidi, 2000; Golparvar et al., 2011). Simple correlation analysis may not provide a complete understanding about the importance of each component in determining yield because of the complex interrelationships among traits (Dewey and Lu, 1959). Path coefficient analysis is a statistical technique proposed by Wright (1921) which partitions the estimated correlations into direct and indirect effects, so the contribution of each trait to yield can be estimated. A path coefficient is a standardized partial regression coefficient that measures the direct effect of a predictor variable on the response variable. This permits the separation of the correlation into direct and indirect effects for a set of priori cause and effect interrelationships (Dewey and Lu, 1959). In most studies involving path coefficient analysis, researchers ignored the importance of the causal relationship, and they used a model in which bidirectional causation, among yield components is assumed. This conventional approach might result in multi-collinearity for variables when associations among some traits are high. There may also be difficulties in interpretation of the actual contribution of each variable, as the effects are mixed or confounded because of collinearity (Hair et al., 1995). Although, there are several reports on correlation and path coefficient analysis in chamomile (Pirkhezri et al., 2008; Zeinali et al., 2008), detailed cause and effect relationships using sequential path analysis have not been examined in chamomile. Recently the sequential path analysis was used by many researchers in different crops by organizing and analyzing various predictor variables in different order paths (Mohammadi et al., 2003; Asghari-Zakaria et al., 2006; Feyzian et al., 2009; Sabaghnia et al., 2010; Dalkani et al., 2011).

The main objectives of this research were to determine interrelationships between flower yield and related characters in chamomile by applying sequential path analysis and identifying traits of genotypes which may be useful in breeding of higher-yielding genotypes for improving flower yield.

MATERIALS AND METHODS

The aerial parts of fully flowered 20 populations of *Matricaria chamomilla* were collected from different regions of Iran, Shiraz, Zabol, Isfahan, Tehran, Kazeron, Shoshtar, Andimeshk, Masjedsoleyman, Khoramabad, Poldokhtar, Baghmalek and Haftgel. The German chamomile accessions were grown at Fozve Research Station, Isfahan Agriculture Research Center located in

Ghahderijan (20 km South-west Isfahan 32°36'N and 51°26' E 1612 m above sea level) in October of 2007. The experiment was conducted in a completely randomized block design with three blocks with one plot for each accession, consisting of 6 rows, 2 m long with 20 cm between and 10 cm within lines. The surface layer (0-30 cm depth) of soil had a clay loam texture with pH=7.7 and EC=2.8 dSm⁻¹. Weed control was done by hand as needed. Plots were irrigated every 8 days.

The samples consisted of a 1 m² area; the four center rows of each plot were harvested for yield by hand at the end of flowering, after leaving two rows in the border areas to avoid border effects, and weighted to determine biological yield (BY). Total flower yield (FY) was determined at 1 m² area of plot when the flowers dried at 25°C for 72 h. Harvest index (HI) was recorded from the ratio of flower yield to biological yield. Plant height (PH), flower diameter (FD), flower height (FH), number of branches per plant (NB), number of flower per plant (NF), number of sepals per flower (NS), flower yield per plant (FYP), and biological yield per plant (BYP) were measured from 10 randomly plant samples harvested at the end of flowering. Days to start flowering (DF) were measured when 10% of the plants in plot had at least one open flower and days to end flowering (DE) were recorded when 90% of the plants in plot had no one open flower.

Statistical methods

The datasets were first tested for normality by the Kolmogorov-Smirnov normality test using SPSS version 19 (SPSS, 2010) statistical software. A preliminary analysis was applied by means of conventional path model in which all flower yield-related traits were considered as first-order predictor variables with FY as the response variable. Sequential stepwise multiple regressions were performed to organize the predictor variable into first, second and third order paths on the basis of their respective contributions to the total variation of the flower yield and minimal collinearity using SPSS 19 (SPSS, 2010) statistical software. The level of multi-collinearity in each component path was measured from the "Tolerance" value and its inverse, the "Variance Inflation Factor" (VIF) as suggested by Hair et al. (1995). The tolerance value is the amount of variability of the selected independent variable not explained by other independent variables (1-R²), where R² is the coefficient of determination for the prediction variable by the predictor variables. The VIF indicates the extent of influence of the other independent variables on the variance of the selected independent variable (VIF=1/(1-R²)). Thus, very small tolerance values (much lower than 0.1) or high VIF values (>10) indicate high collinearity (Hair et al., 1995; Mohammadi et al., 2003). Direct effects of flower yield characters in different order paths were estimated by the procedure suggested by Williams et al. (1990). Partial coefficients of determination (analogous to R² of linear regression) were calculated from the path coefficients for all predictor variables. To estimate the standard error of path coefficient, bootstrap analysis (Efron and Tibshirani, 1993) was performed by Amos 19 (SPSS, 2010) statistical package, followed by the standard *t* test to analyze the significance of path coefficients.

RESULTS AND DISCUSSION

Simple correlations

The simple correlation coefficients computed between different pairs of traits indicated there were high positive

Table 1. Pairwise correlation coefficients between 13 traits of 20 chamomile populations.

Trait	FH	FD	PH	NF	NB	NS	DF	DE	BYP	FYP	BY	FY
FD	0.24											
PH	0.55*	0.36										
NF	0.40	0.43	0.21									
NB	0.56*	0.34	0.28	0.87**								
NS	0.42	0.48*	0.51*	0.43	0.27							
DF	-0.42	-0.37	-0.38	-0.1	-0.16	-0.23						
DE	-0.21	-0.43	-0.44	-0.22	-0.28	-0.31	0.98**					
BYP	0.25	0.41	0.19	0.21	0.33	0.1	0.2	0.1				
FYP	0.4	0.7**	0.36	0.45*	0.43	0.33	0.06	-0.3	0.71**			
BY	0.08	0.57**	0.15	0.42	0.18	0.25	0.26	0.19	0.42	0.75**		
FY	0.28	0.57**	0.46*	0.68**	0.48*	0.58**	-0.08	-0.21	0.48*	0.6**	0.6**	
HI	0.29	0.36	0.5*	0.54*	0.48*	0.53*	-0.35	-0.45*	0.33	0.25	0.08	0.84**

* Significant at the 0.05 level

** Significant at the 0.01 level

FH, flower height; **FD**, flower diameter; **PH**, plant height; **NF**, number of flower per plant; **NB**, number of branches per plant; **NS**, number of sepals per flower; **DF**, days to start flowering; **DE**, days to end flowering; **BYP**, biological yield per plant; **FYP**, flower yield per plant; **BY**, biological yield; **FY**, flower yield; **HI**, harvest index.

correlations between flower yield and all of the measured traits except for FH, DF and DE. The highest correlation for flower yield was between HI and FY ($r=0.84$). As shown in Table 1, there was a statistically significant and high positive correlation between FD and FYP, between FD and BY, between NF and NB, between DF and DE, between FYP and BYP and between BY and FYP.

Significant positive correlation were detected between FY with NF ($r=0.68$) and NB ($r=0.48$). These results were in close agreement with those of Jamshidi (2000), Pirkhezri (2010), Zeinali et al. (2008) and Golparvar et al. (2011) who reported that a positive and significant correlation between FY with NF and NB. In this study, the correlation between FY with DF and DE was not significant. These results was not consistent with Pirkhezri et al. (2010), Zeinali et al. (2008) and (Golparvar et al., 2011) who reported that a positive and significant correlation between FY with DF and DE. This may be the nature of the genotypes used in this experiment. Correlation of FY and PH was positive and significant. These findings are consistent with Pirkhezri et al. (2010) and Zeinali et al. (2008) who reported a significant and positive correlation between FY and PH.

Conventional and path coefficient analysis

To determine the relative importance of the characters, the data were subjected to conventional path analysis. The results pertaining to direct effects of components traits on chamomile flower yield, where yield-related traits were considered as first order variables, with flower yield as the response variable are presented in Table 2.

Results from this analysis indicated high multi-collinearity for some traits such as NF, NB, DF and DE. On the basis of "tolerance" and VIF values, besides the magnitude of direct effects, HI and BY were considered as first order variables. This procedure was gain performed separately taking HI and BY as dependent variables to find out second order variables for FY. Similar procedure was followed to determine the third order variables. Estimation of direct effects by sequential path analysis were considered, where flower yield-related traits, as grouped into first, second and third-order variables, with flower yield (Model II). Analysis of multi-collinearity indicated the sequential path analysis a better understanding of the interrelationships among the various characters and their relative contribution to FY (Figure 1). The results of tolerance and VIF values for predictor variables indicated a considerable reduction of VIF values in model II compared to model I (Table 3). Sequential path coefficient analysis in present study minimized collinearity measures of all variables, thus facilitating detection of the actual contribution of each predictor variables in different path coefficients, with negligible confounding effects and interferences. The superiority of sequential path over the conventional path analysis in minimizing collinearity measures and identifying actual contributions of each component in different path components are similar to those found in other crop studies (Mohammadi et al., 2003; Asghari-Zakaria et al., 2006; Feyzian et al., 2009; Sabaghnia et al., 2010; Dalkani et al., 2011) indicating that it should be very effective in obtaining favorable results.

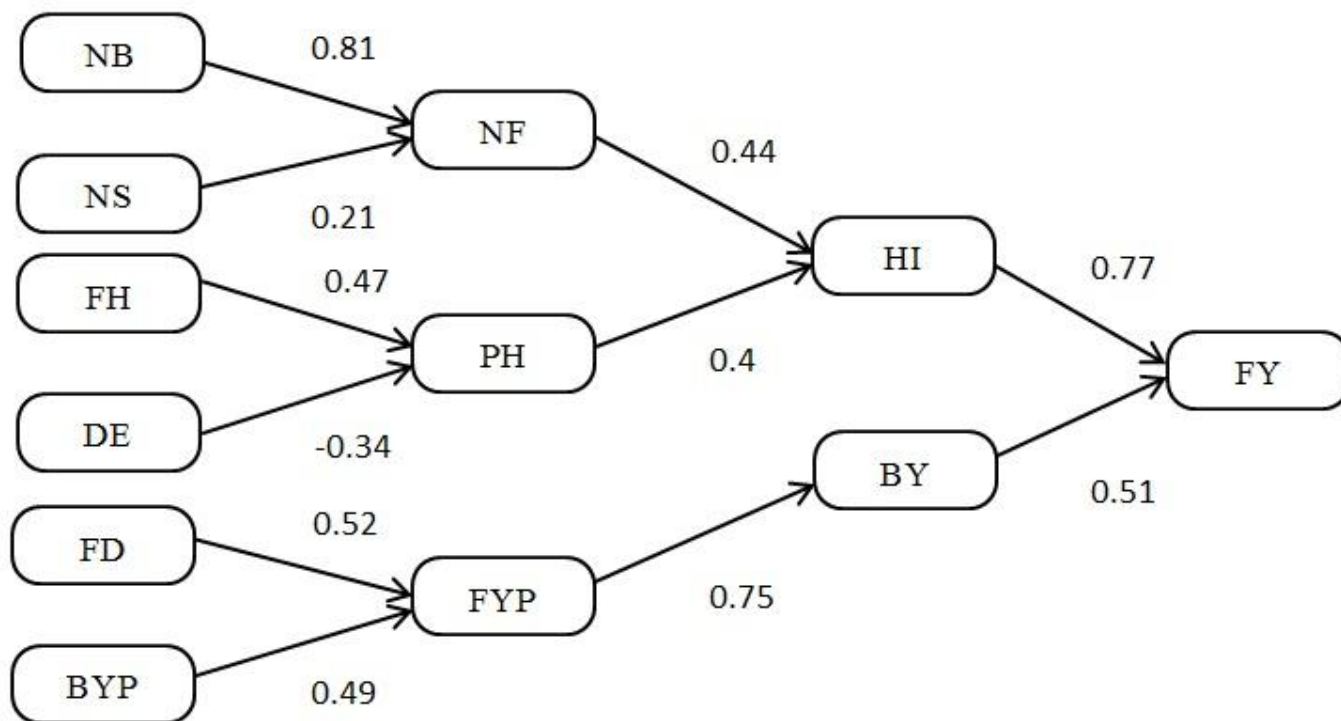
In statistical analysis, bootstrap technique provides estimates of the standard error, confidence interval, and

Table 2. Direct effect of first-order variables on the flower yield of 20 chamomile populations and two common measures collinearity in conventional path model.

Trait	Direct effect	Tolerance	VIF*
FH	0.020	0.346	2.892
FD	0.026	0.235	4.255
PH	0.025	0.358	2.794
NF	0.213	0.074	13.599
NB	-0.160	0.087	11.479
NS	0.003	0.400	2.498
DF	0.009	0.013	75.990
DE	0.072	0.013	77.377
BYP	0.028	0.233	4.296
FYP	-0.006	0.109	9.161
BY	0.438	0.150	6.682
HI	0.761	0.274	3.646

* VIF: variance inflation factor

FH, flower height; **FD**, flower diameter; **PH**, plant height; **NF**, number of flower per plant; **NB**, number of branches per plant; **NS**, number of sepals per flower; **DF**, days to start flowering; **DE**, days to end flowering; **BYP**, biological yield per plant; **FYP**, flower yield per plant; **BY**, biological yield; **HI**, harvest index.

**Figure 1.** Sequential path analysis diagram illustrating the interrelationships among various traits contributing to flower yield.

the distribution of any statistic. In this study, the mean direct effects estimated from a set of 200 bootstrap samples were in close agreement with the observed direct effects of the various traits (Table 4). The low standard error for all direct effects and the low bias also

indicated robustness of the sequential path compare to the conventional path. The *t* test of significance, using standard error values achieved from bootstrap resampling revealed that all direct effects were significant. HI and BY, as first order variables, accounted

Table 3. Measures of collinearity values (tolerance and VIF*) for predictor variables in conventional path model (Model I) (CPA, all predictor variables as first-order variables) and sequential path model (Model II) (SPA, predictors grouped into first, second and third-order variables).

Predictor variable	Response variable	Tolerance		VIF	
		CPA	SPA	CPA	SPA
HI	EY	0.274	0.85	3.646	1.177
BY		0.233	0.899	4.296	1.112
NF	HI	0.074	0.957	13.599	1.045
PH		0.358	0.957	2.794	1.045
FYP	BYF	0.109	1	9.161	1
NB	NF	0.087	0.929	11.479	1.077
NS		0.4	0.929	2.498	1.077
FH	PH	0.346	0.954	2.892	1.048
DE		0.013	0.954	77.377	1.048
FD	EYP	0.235	0.833	4.255	1.201
BYP		0.233	0.833	4.296	1.201

*VIF: variance inflation factor

FH, flower height; **FD**, flower diameter; **PH**, plant height; **NF**, number of flower per plant; **NB**, number of branches per plant; **NS**, number of sepals per flower; **DE**, days to end flowering; **BYP**, biological yield per plant; **FYP**, flower yield per plant; **BY**, biological yield; **HI**, harvest index.

Table 4. Estimation of standard error values of path coefficients using bootstrap analysis.

Predictor variable	Response variable	Adj. R ²	Direct effect	Bootstrap		
				Bias	Mean	SE
HI	EY	0.99	0.77	-0.017	0.756	0.073
BY			0.51	0.014	0.526	0.087
NF	HI	0.48	0.44	-0.014	0.43	0.134
PH			0.4	0.005	0.401	0.136
FYP	BYF	0.56	0.75	-0.005	0.746	0.087
NB	NF	0.78	0.81	0.006	0.811	0.073
NS			0.21	-0.008	0.203	0.093
FH	PH	0.41	0.47	0.037	0.512	0.239
DE			-0.34	-0.104	-0.445	0.172
FD	EYP	0.72	0.52	-0.001	0.52	0.135
BYP			0.49	0.017	0.51	0.107

FH, flower height; **FD**, flower diameter; **PH**, plant height; **NF**, number of flower per plant; **NB**, number of branches per plant; **NS**, number of sepals per flower; **DE**, days to end flowering; **BYP**, biological yield per plant; **FYP**, flower yield per plant; **BY**, biological yield; **HI**, harvest index.

for nearly 99 of variation in flower yield that indicates the influence of the HI and BY traits as first order variable (Table 4). Both these variables also displayed high and positive direct effects on FY. The direct effect of HI (0.77) on FY was found to be relatively higher than that of BY (0.51). The results of sequential path analysis, when the second-order variables were used as predictors, and the first-order variables as response variables, indicated that NF and PH positively influenced the HI and accounted for more than 48% of the observed variation. The FYP positively influenced BY and accounted for more than 56% of the observed variation. When the third-order

variables were used as predictors and second-order variables as response variables, the results indicated NB and NS positively influenced the NF and accounted for about 78% of observed variation. Also, FH positively and DE negatively influenced PH and accounted for about 41% of observed variation. The FD and BYF positively influenced FYP and accounted for about 72% of observed variation (Table 4).

This study demonstrated that the utility of the sequential path analysis over the conventional path analysis is use in discerning the direct and indirect effects of various yield-related characters. When all the traits

were used as first-order variables as per conventional path model, we detected the occurrence of moderate to severe multi-collinearity for salient yield components. This accentuated the inadequacy of the conventional path model in determining the actual effect of each predictor variable on the response variable. For instance, when the conventional path model was applied, NF had a negligible effect on FY, indicating that this trait had no significant effect on FY. However, the sequential path model clearly indicated that NF had a positive and significant effect on FY through HI. Such effects could not be detected through the conventional path model because of high multi-collinearity of traits, such as NF, NB and DE.

In this study, the traits HI, BY, NF, PH, FYP, NB, NS PH, DE, FD and BYP were identified as the first, second and third order variables. Zeinali et al. (2008) found number of flower per plant and plant height as most interesting traits to use as an indirect selection criteria for flower yield. Our results are largely in agreement with the above observations, as reflected by positive and significant direct effects of NF and PH on HI, and HI on FY. Letchamo (1993) and Jamshidi (2000) reported the traits of number of flower per plant, flower weight and number of flowering branches as the dry flower yield components. These results besides our results, revealed the importance of HI, BYF, NF, PH and FYP as the selection criterion for chamomile flower yield. Therefore selection for increasing flower yield through these traits might be successful.

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