

Full Length Research Paper

Analysis of influencing factors in determining characters for photosynthesis in *Lonicera japonica* Thunb.

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This paper examined eight additional parameters relating to photosynthetic rate of *Lonicera japonica* Thunb. Correlation analysis, regression analysis and factor analysis were used to analyze the relationship between net photosynthetic rate (Pn) and environment factors, which included stomatal conductance (Sc), intercellular CO₂ concentration (Ci), transpiration rate (Tr), air temperature (Ta), leaf temperature (Tl), air CO₂ concentration (Ca), relative air humidity (RH), photo flux density (PFD), etc.. The results showed that the diurnal Pn of *L. japonica* Thunb. It may vary with double peak curves; there was no significant midday depression from June to August. The stepwise regression analysis which introduced characters into a multiple regression equation in the order in which they contribute to Pn, together with factor analysis was used to analyze the same dependence structure. The 8 causes were grouped into the basic factors which generate the dependence structure. The result suggested that high photo flux density (PFD), high Tl and low RH may result in the high Pn. Increasing Tr and PFD, accelerating the hot air flow, would be the most effective way of increasing Pn. The analysis result would assist the medicinal herb grower by adjusting environment factors level to regulate Pn of *L. japonica* Thunb. and improve output and quality of flower.

Key words: *Lonicera japonica* Thunb., environment factors, net photosynthetic rate, interrelationships.

INTRODUCTION

Lonicera japonica Thunb. is a genus of woody plants (family Caprifoliaceae) that grow extensively in Europe, Asia and North America. *L. japonica* Thunb. was recorded in China Pharmacopeia (2005) with the Chinese name Jinyinhua. The aqueous extract from *L. japonica* Thunb. flower has been used in Chinese traditional medicine for treating fever, arthritis and infectious diseases for thousands of years. This plant had been shown to display a wide spectrum of biological and

pharmacological activities such as antibacterial, antiviral (Houghton et al., 1993), antioxidant and inhibition of the platelet activating factor (Kim et al., 1994). *L. japonica* contains anticomplementary polysaccharides and polyphenolic compound, the polyphenolic compounds mainly come from flowers (Chang and Hsu, 1992).

The initiation of flowering, that is, the conversion of shoot apical meristems from vegetative to reproductive development, was a critical event in the life cycle of higher plants. Under good light condition, the plants could obtain high photosynthetic capacity, which was beneficial for the flowering and fruit setting of *Cypripedium flavum* (Zhang et al., 2005). The consideration of the effect of environment factors, such as daily carbon gain, supplies moisture and gas flow on photosynthetic capacity was necessary and closely correlated with photosynthetic capacity. Widodo et al. (2003) discovered photosynthetic responses to atmospheric carbon dioxide concentration

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Abbreviations: Pn, Net photosynthetic rate; Ci, intercellular CO₂ concentration; Ca, air CO₂ concentration; PFD, photo flux density; Sc, stomatal conductance; Tr, transpiration rate; Ta, air temperature; Tl, leaf temperature; RH, relative air humidity; WUE, leaf water use efficiency; ANOVA, analysis of variance.

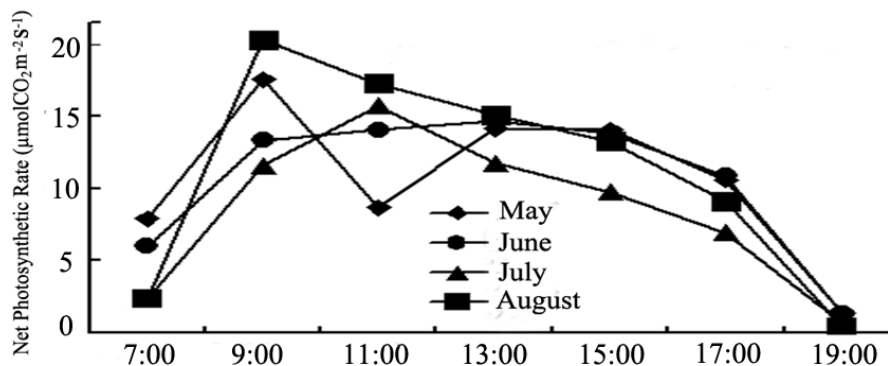


Figure 1. The diurnal variation of Pn in *L. japonica* Thunb. from May to August.

enrichment as well as drought stress, and rising atmospheric carbon dioxide concentration and predicted changes in rainfall frequency and intensity could have considerable impact on crop growth and yield. *L. japonica* Thunb. was easily adapted to low light conditions, that physiological adaptability of *L. japonica* Thunb. to low-high environments might be related to climbing mechanics (Gregory and Alan, 1998; Hair et al., 1998; Jennifer and Katherine, 1999).

Little was known about the interactive effects of these environmental variables on the fundamental processes of photosynthesis of *L. japonica* Thunb.. The major purpose of this study was to use both regression analysis and factor analysis to determine the dependence relationship between Pn and 8 independent environmental variables, to find a set of consistent and efficient estimates of element parameters in variables factors and residual variables in relation to Pn of *L. japonica* Thunb. The different regression analyses to determine the best fit equations describe the relation of Pn and different environment factors.

MATERIALS AND METHODS

Measurement of photosynthesis

The study was conducted at Huaihua University medicinal garden (E109.95, N27.55, altitude 838 m). Different varieties of *L. japonica* Thunb. came from eight different places of China (*liaoling, Shandong, henan, hebei, zhejiang, hunan, guizhou* and *guangxi*). From May to September 2010, the mean monthly air temperature and total precipitation were 16.4°C and 1160 to 1450 mm, respectively. Compared with the climatic data during 1979 to 2009, the average air temperature was similar, while the precipitation was lower. The plants were shaded by nylon netting to control sunlight, and irrigated during the entire growing season. In 2010, the new leaves emerged above on March 18, and flower appeared on May 20. Fruit setting occurred between June and September. Pn was measured using a LI-Cor 6400 portable photosynthesis system (LI-Cor, NE, USA) on days 30 (April 17), 60 (May 17), 90 (June 16), 120 (July 16), 150 (August 15), after leaf emergence, respectively. Photosynthesis was conducted on the second leaf from the top of the plants, and was measured once per 2 h from 7:00 am to 7:00

pm. These measurements were replicated using four to eight single; the plant was randomly selected to measure.

Statistical analysis

All the percentage values were arcsine transformed and counts were subjected to square root transformation. The relationships between photosynthesis and environment factors were studied by linear regression analysis (SPSS14.0 software); statistical significance level was assumed at $P < 0.05$. The currently used factor analysis followed the method used by Cattell (1965). The method basically reduces a large number of correlated variables to a small number of uncorrelated variables or factors. It was assumed that the set of observed correlated variables form a multivariate normal distribution and that the factors were orthogonal. In this method a multiple regression equation is obtained by adding one independent variable at a time according to their relative importance in determining the dependent variable.

The factor analysis started from the correlation matrix of 8 independent traits which was experimentally obtained. The correlation of each test with itself would give diagonal values of unity and these were replaced by communalities. The communality was the amount of the variance of a variable accounted for by the factors taken together. The maximum likelihood method was used to determine the communality values and the number of factors. Communalities were fixed by taking the highest correlation in each row or column of matrix. These values were used as approximate values of the loadings. Iteration continues until the values converge at the best approximation. When the contribution of a factor to the total variance was less than 10%, the process stopped. Thus only the more important factors, in order of increasing triviality, were retained. Here, the varimax rotation method proposed by Walton (1971) was used.

RESULTS

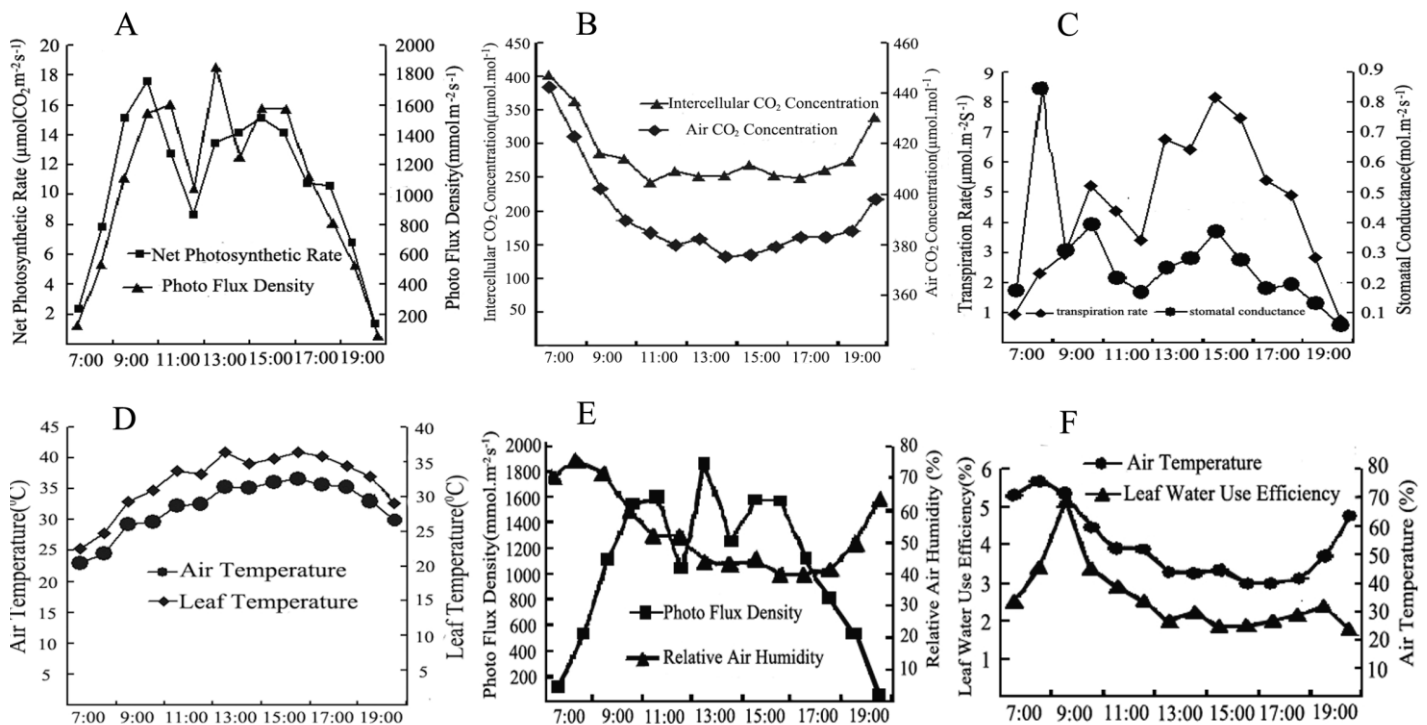
Change comparison

The photosynthetic responses of *L. japonica* Thunb. were similar among different months. The photosynthetic capacity of *L. japonica* Thunb. changed with time in every day, Pn peaked from 9:00 to 11:00 and then decreased (Figure 1). The results showed that the diurnal Pn of *L. japonica* Thunb. in May varied with double peak curves.

Table 1. Pearson correlation coefficients matrix of 9 characters in relation to photosynthesis in *L. japonica* Thunb.

	Pn	Sc	Ci	Tr	TI	Ca	Ta	RH
Sc	0.358							
Ci	-0.807(*)	0.183						
Tr	0.944(**)	0.726	-0.893(**)					
TI	0.684	-0.284	-0.971(**)	0.839(*)				
Ca	-0.728	0.353	0.978(**)	-0.803(*)	-0.960(**)			
Ta	0.484	-0.337	-0.833(*)	0.711	0.935(**)	-0.834(*)		
RH	-0.706	0.205	0.974(**)	-0.857(*)	-0.983(**)	0.934(**)	-0.890(**)	
PFD	0.962(**)	0.264	-0.858(*)	0.980(**)	0.793(*)	-0.790(*)	0.660	-0.784(*)

The abbreviations were same as aforementioned. *, Significant differences at 5 % level; **, significant differences at 1% level.

**Figure 2.** Interaction effect of all factors relating to photosynthesis in *L. japonica* Thunb.

The peaks occurred at 9:00 and 14:0, respectively. There was a significant midday depression. Environmental factors, plant growth situation and their interaction result in the depression. On May 17, new leaves grew only two months, the mean temperature was 30.4° on the day. At midday, light intensity was the largest, but air relative humidity, stomatal conductance and intercellular CO₂ concentration were at minimum, and hence photosynthetic midday depression occurred. In other months, the temperature was relatively low on the day of the experiment, the plants grew strong and had good stress tolerance, and the photosynthetic depression at midday was not obvious. In the bloom as a whole, the biomass decreased considerably between June and August, the

flowers were passing through a senescent phase during this time. However, the increasing photosynthetic activities indicated an active turnover of the flowers with a subsequent renovation by mid-August. So the important thing is that there was not a significant midday depression from June to August.

Correlation analysis

The correlation analyses were made between Pn and 8 independent traits (Table 1, n = 45; Figure 2). It was obvious that the correlation matrix showed that the Tr and PFD were significantly correlated with Pn. This means

Table 2. Quadratic regression equation for calculation the effect of different factor to Pn in *L. japonica* Thunb.

No.	Factor	Correlation	Significant	Binominal regression equation
1	C _i	-0.697	0.004	$Y=12.462+0.069X-0.025X^2$
2	Tr	0.795	0.002	$Y=-2.477+4.512X-0.263X^2$
3	Ta	0.584	0.022	$Y=-33.673+2.626X-0.025X^2$
4	TI	0.564	0.035	$Y=-294.644+3.418X-0.041X^2$
5	Ca	-0.602	0.036	$Y=3.769+0.225X-0.003X^2$
6	PFD	0.875	0.001	$Y=0.458+0.024X-0.006X^2$

X represented factor, Y represented Pn; the abbreviations was same as aforementioned.

Table 3. Results of factor analysis for for all characters.

Factor	Component	
	1	2
Sc	-0.115	0.984
C _i	-0.987	0.061
Tr	0.927	0.375
TI	0.980	-0.183
Ca	-0.953	0.222
Ta	0.883	-0.275
RH	-0.972	0.115
PFD	0.944	0.297

The abbreviations was same as aforementioned.

that transpiration rate and photo flux density brought about considerable influence to photosynthesis in *L. japonica* Thunb. The correlation was not significant between Sc, TI, Ca, Ta, RH and Pn but one of them was moderately correlated with Pn. The C_i showed significant negative correlation coefficients with Pn, Tr, TI, Ta, PFD, but C_i showed significant positive correlation coefficients with Ca. The significant negative correlation between C_i and Pn suggested the importance of CO₂ at the photosynthesis in *L. japonica* Thunb. There was no correlation between Sc and other factors, it was possible the photosynthetic variation of *L. japonica* Thunb. was not relative to plant stomata. Tr showed significant positive correlation coefficients with TI and PFD, and Tr showed significant negative correlation coefficients with C_i, Ca and RH. TI showed significant negative correlation coefficients with Ca and RH, while it showed significant positive correlation coefficients with Ta and PFD. PFD showed significant negative correlation coefficients with Ca ($r = -0.79$), indicating an indirect relation between photo flux density and air CO₂ concentration.

Linear regression analysis

The characters which made the largest single contribution to Pn was the PFD. C_i, Ta, TI, Tr, made important contributions to Pn. There were also indications

that Sc and RH were not important components of Pn. Using X₁, X₂, X₃, X₄ and X₅ as independent variable, represented the PFD, C_i, Ta, TI and Tr, respectively, Pn(y) as dependent variable. The linear model was set up, $Y = 149.66 + 0.436X_1 - 0.379X_2 - 0.101X_3 + 0.118X_4 + 0.58X_5$. The multiple correlation coefficient of regression equation was 0.947, and R² was 0.897. The validity of equation was tested by analysis of variance (ANOVA). F value was 37.97, P = 0.002. These results showed that the model had high relative coefficients, good stability and good predictability. We proposed a model-based approach, step down quadratic regression for Pn and different environment factors. It was performed by initially fitting a quadratic regression model to each factor (Table 2). The model would not be built if the quadratic term was determined to have no statistically significant relationship with Pn.

Factor analysis

At first, we conducted Kaiser-Meyer-Olkin (KMO) and Bartlett's test for all factors. The value of KMO was 0.676 ($0.5 < KMO < 0.1$: suitable). Value of Bartlett was 159.44, P < 0.01. The factor analysis was appropriate to all factors in relation to photosynthesis. This form of analysis divided the 8 causes into 2 groups (Table 3). The Factor 1 which made the largest contribution to Pn and accounted for 78.11% of the total variation was composed

of Ci, Tr, TI, Ca, Ta, RH and PFD. The Sc was accounted for 21.89% of the total variation. The 2 principal components of Pn, internal character and external character, were most closely associated with Pn. The external character was reflected by Ci, Tr, TI, Ca, Ta, RH and PFD, was also an important part of Factor 1. The only internal character included in Factor 2 was Sc. The result indicated the importance of Tr, PFD, Ci in relation to Pn. Increasing Tr and PFD, accelerating the hot air flow, would be the most effective way of increasing Pn.

DISCUSSION AND CONCLUSION

In the study, an attempt was usually made to investigate a complex pattern which was the outcome of many causes by using multiple regressions and multiple correlations. The resultant factors are mutually orthogonal and the loadings indicate the contribution of the variables. The factor analysis indicates both groupings and percentage contribution. The different methods of analysis provided complementary ways of studying the data. For example the correlation analysis showed the initial importance of RH which diminished when its regression analysis were introduced into the equation. Here the factor analysis also indicated the importance of the components RH. Factor analysis indicated the relationship between internal character and external character, but this was not evident from the regression analyses. Determination coefficients (R^2) are valuable indicators of the appropriateness of a model to describe the relation between variables. A high R^2 coefficient suggests that much of the variability was described by the model (Compton, 1994). After the equation was obtained, a numerical procedure was applied to find Determination coefficients. To this point, regression analysis seemed to be an excellent way when several quantitative treatment levels are compared.

Photosynthetic productivity of plant was the primary power to drive and sustain the whole ecological system. Ninety percent of dry matter in plant was produced by plant photosynthesis, which was the basis of dry matter accumulation and yield formation in crops (Tian et al., 2005). Among other factors, the impact of water and nitrogen stress on photosynthesis of crops was one of the main causes leading to yield loss and quality variation. To determine the photosynthetic parameters of plant by remote sensing technique was helpful for establishing simulation models for evaluating biomass production and estimating crop yield. Thus, a nondestructive, quantitative, and rapid method for assessment of plant leaf photosynthetic characteristics under varied growing conditions would evidently contribute to monitoring growth status and estimating yield and quality characters of plants. We determined the quantitative relationships between leaf photosynthetic characteristics and different factors. The modeling could be used to monitor leaf photosynthetic characteristics at different growth stages

of *L. japonica* Thunb. under diverse growing conditions, with proper shading, irrigating and increasing CO₂ concentration or under weak light with ventilating, heating, reducing shading and relative humidity, the medicinal herb grower could effectively improve output and quality of flower. The photosynthetic capacity of *L. japonica* Thunb. varied significantly. Because the environmental changes were relatively big during the growth period, the differences in photosynthetic capacity of *L. japonica* Thunb. would be mainly caused by the changes of environmental. The results suggested that the different effect of various environmental on photosynthesis should be taken into consideration growth of *L. japonica* Thunb. were predicted. The results suggested that there was no ecotypic differentiation with respect to environments from which the individuals of *L. japonica* Thunb. were collected. Also external character was important to obtain high photosynthetic capacity. In general high PFD, high TI and low RH should result in a high Pn.

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