International Journal of Agriculture Sciences
ISSN: 0975-3710&E-ISSN: 0975-9107, Volume 9, Issue 43, 2017, pp.-4689-4692.
Available online at http://www.bioinfopublication.org/jouarchive.php?opt=&jouid=BPJ0000217

Research Article

GENOTYPIC VARIABILITY IN PHOTOSYNTHETIC PERFORMANCE AND GAS EXCHANGE INDEXES OF GUAVA CULTIVARS

SHIVA B.*, NAGARAJA A., SRIVASTAVA M. AND GOSWAMI A.K.

Division of Fruits and Horticultural Technology, ICAR-Indian Agricultural Research Institute, New Delhi, 110 012, India
*Corresponding Author: Email-banothshivaiari@gmail.com

Received: September 01, 2017; Revised: September 15, 2017; Accepted: September 16, 2017; Published: September 18, 2017

Abstract- Photosynthesis is a central metabolic process in plants and photosynthetic traits can be used as indicators to judge the adaptability and resistance of plants. Besides the classical measurements of photosynthesis by gas exchange analysis, light harvesting pigment measurements have become a widely used method to study the functioning of the photosynthetic apparatus and are a powerful tool to study the plant's response to environmental stress. In this study, the photosynthetic performance and gas exchange characteristics among 22 cultivars of *Psidium guajava* were investigated under hot arid conditions of India for identification of promising genotypes. Significant differences were noticed in the rate of photosynthesis (P_N), stomatal conductance (gs), transpiration rate (E), internal CO_2 concentration (C_i), water use efficiency (WUE) and light harvesting pigments. This study indicates that it could provide a useful target for breeding programs and will lead to more efficient use of guava cultivars with better adaptation to the limiting agro-climatic conditions of India.

Keywords- Photosynthesis, Water-use efficiency, Chlorophyll, Carotenoid content.

Citation: Shiva B., et al., (2017) Genotypic Variability in Photosynthetic Performance and Gas Exchange Indexes of Guava Cultivars. International Journal of Agriculture Sciences, ISSN: 0975-3710 & E-ISSN: 0975-9107, Volume 9, Issue 43, pp.-4689-4692.

Academic Editor / Reviewer: Vanitha S

Introduction

Guava (*Psidium guajava* L.) is an evergreen fruit species belongs to the Myrtaceae family. It is believed to have originated from an area extending from southern Mexico into or through Central America [1]. Due to its ability to grow in tropical and subtropical climates, the guava tree has been introduced to many countries; it is well adapted to a wide range of soils [2]. Furthermore, the guava tree requires an annual water supply of 1000–2000 m^3/ha-year [3]. The best temperature for guava cultivation ranges from 15 to 30.8°C, with an annual average temperature of 18.8°C [4]. However, plant experiences high temperature stress of the North, where temperatures often exceed 40°C. Presumably this has major consequences on fundamental plant processes such as physiological and biochemical functions of the plant and concurrent may alter plant growth, agronomic yields and quality [5]. It disrupts photosynthetic pigments and reduces the gas exchange leading to a reduction in plant growth and productivity. Crops sense and respond directly to contrasting environmental circumstances/conditions through changes in photosynthetic activity and stomatal conductance (gs) [6, 7]. Because photosynthesis is the basic crucial process that plants employ to fix energy; so, a plant's yield and survival had appeared to be depend on their photosynthetic capacity [8]. As the key process for neogenesis of biological material, photosynthesis plays a central metabolic role in plant performance under abiotic stresses, and the net photosynthesis rate (PN) and transpiration rate (E) in most fruit crops could be reduced with a rapid closure of stomata, with the reduction of stomata conductance (gs) as well [9, 10]. At higher levels,' it is difficult to distinguish differences in photosynthesis from differences in canopy structure that alter light interception. The photosynthetic gas exchanges of C_3 plants have been widely explored and the dependence of CO_2 assimilation to environmental parameters like atmospheric CO_2 concentration and irradiance has been mathematically formulated [11]. Photosynthetic traits can help identify suitable growth conditions and plant adaptation strategies to different environments [12]. Moreover, expanding knowledge about the magnitude of genotypic variation in stomatal behavior would be of considerable importance because photosynthesis is one of the key characteristic processes underpinning dry matter production and ultimately yield [13]. Nevertheless, well focused research work to examine apparent genotypic variation for more precise information on physiological parameters have been received less attention in this crop.

Thus, the principal objective of the present study was to assess genotypic variation in photosynthetic performance and physiological attributes in a set of 22 germplasm comprising commercial varieties of guava

Material and Methods

The experimental materials utilized for the present investigation 6 year old uniformly growing plants of 22 guava cultivars, *viz.,* Allahabad Safeda, Arka Amulya, Arka Mridula, Behat Coconut, Black Guava, Hafsi Red, Hissar Safeda, Hissar Surkha, Lalit, Lucknow-49, Pant Prabhat, Punjab Pink, Red Peel, Red type, Sasni Collection, Sasri Selection, Shweta, Snow White, Sour type, TN Selection, Thai guava, Yellow type were selected from the experimental orchard of the Division of Fruits and Horticultural Technology IARI, New Delhi, India. Experiment was laid out in randomized block design, planted in square system at a distance of 6 m × 6 m, with cultivars as four treatments, and four replications. The experimental site falls under trans-gangetic plains of agro-climatic zones of India located at 77◦12 'E' longitude, 28◦40 'N' latitude and an altitude of 228.6 m above mean sea level. It has typical subtropical climatic conditions characterized by hot and dry summer (41–44◦C) followed by cold winter (3–7◦C). The average annual rainfall of the experimental site was 613 mm and more than 60% rainfall received during July, August and September Sunshine hour varied from 1.2 h/day in January to 10.9 h/day in June. Soil type was a sandy loam with a pH of 7.10 and

EC (1:2) of 0.35 dS m−1, acation exchange capacity (CEC) 7.54–10.72 Cmol kg−1, organic carbon 4.8 g kg−1. The experimental plants were supplied 200 g N, 50 g P and 400 g K/tree/year along with 30 kg well-rotted farm yard manure. Other cultural operations were carried out uniformly. Foliar micro-nutrients application and pest and disease management were in accordance with normal commercial practices.

Gas exchange characters and related traits

Net photosynthesis rate (PN: mol CO_2 m-2s^{-1}), stomatal conductance (gs: mol $m^{-2}s^{-1}$), internal CO_2 concentration (Ci: µmol CO_2mol air $^{-1}$) and transpiration rate (E: mmol H_2O $m^{-2}s^{-1}$) were measured using a portable infrared gas analyzer system (IRGA) (Li-Cor 6200, Li-Cor Biosciences, Lincoln, NE, USA). The gas exchange rates were determined at each step after maintaining the leaf for 5–10 min at the new CO_2 concentration. Five top most fully expanded leaves from each treatment were selected randomly for the measurements. The leaf was enclosed in the assimilation chamber and the Pn was monitored while CO_2 concentration changed over a definite time interval. The system automatically calculated the rate of photosynthesis on the basis of preloaded flow rate and leaf area. Transpiration and stomatal conductance were also recorded simultaneously by infrared gas analyzer (Li-Cor 6200, Li-Cor Biosciences, Lincoln, NE, USA) on the same leaf. All these measurements were taken at 10:00 to 11:00 h (Indian standard time) when relative humidity, temperature, photosynthetic photon flux density and CO_2 concentration ranged from 50–60 %, 30–35 C, 1200 µmol (photon) m^{-2} $s^{-1}$and 350–360 µmol mol^{-1}, respectively. Water use efficiency (WUE) was calculated by taking the ratio of photosynthetic rate and transpiration rate (PN/E) and internal CO_2 concentration of the leaf (PN/Ci) was calculated as suggested by Silva-Marcelo de Almeida *et al*. (2013) [14].

Light harvesting pigments

The leaf chlorophyll contents (chlorophyll a, b, and total chlorophyll) and total carotenoids were estimated using the method suggested by Hiscox and Israelstam (1979) [15]. Accurately weighed 100 mg of clean, fully matured leaves were immersed in 10 ml of dimethylsulfoxide (DMSO) (AR grade, SRL Chem. Co., Mumbai, India). The sample was incubated at 700C for 4 h in an incubator (TH 7004, Sanco Co., New Delhi). After incubation the sample was removed and 1 ml of the solution was diluted to 5 ml with pure DMSO and the sample was read on a UV–VIS spectrophotometer (UV–VIS 5704SS, E. C. India Limited, Hyderabad, India) at 645, 663 nm and 665 using pure DMSO as a blank. During the assay, samples were protected against light to prevent pigment degradation and pigment contents were calculated from the equations proposed by Lichtenthaler and Buschmann (2001) [16].

The data obtained were statistically analyzed through one-way analysis of variance (ANOVA) using SPSS 16 software and significance was determined at $P<0.05$. The data are presented as mean + SD of three replicates.

Results and Discussion

Gas exchange traits

Gas exchange characters and related traits are highly important for the growth and yield in plants. Variations in content of light pigments can directly influence the leaf gas exchange [17].

Significant genotypic variation was found for PN among the cultivars [Table-1]. Net photosynthesis was recorded maximum in Shewta (10.24 µmol $m^{-2}s^{-1}$) and minimum is in Arka Amulya (2.36 µmol $m^{-2}s^{-1}$).Photosynthesis, the unique biological process responsible for the conversion of light energy to chemical forms, is the ultimate basis of crop growth and productivity [18].Certainly, larger leaf area must have resulted in more synthesis of photosynthates and their accumulation, which might be responsible for better growth.

Genotypic variation for transpiration rate was observed in studied cultivars [Table-1]. The transpiration rate was highest in Lucknow-49 (0.72mmol $m^{-2}s^{-1}$) and lowest in Arka Amulya(0.06mmol $m^{-2}s^{-1}$). The stomatal control of transpiration rate is an important component of the leaf energy balance and can be of great importance for maintaining an optimal or appropriate leaf temperature for photosynthesis particularly under conditions of increasing or highlight intensity that are observed over a typical diurnal period. Transpiration often is seen as a cost for carbon fixation at the leaf level, but it is important to take into consideration its roles in the transport of solutes in the different parts of the plant or for leaf cooling [19].

Table-1 *Variation in gas exchange and water use efficiency parameters of guava cultivars*

S.No.	Genotype	P_N (µmol m^{-2} s^{-1})	gs (mol m^{-2} s^{-1})	E (mmol m^{-2} s^{-1})	C_i (ppm)	WUE (µmol mol^{-1})
1	Allahabad Safeda	4.23	0.05	1.28	298.65	2.89
2	ArkaAmulya	2.36	0.05	0.30	327.00	2.17
3	ArkaMridula	6.17	0.12	2.64	247.13	2.53
4	Behat coconut	4.07	0.09	1.45	301.75	2.71
5	Black guava	5.35	0.08	2.38	261.54	2.94
6	Hafsi Red	8.57	0.13	2.96	201.50	2.57
7	Hissar Safeda	7.78	0.05	2.73	224.27	2.29
8	HissarSurkha	5.37	0.09	1.49	260.93	2.80
9	Lalit	4.93	0.01	1.55	295.64	2.60
10	Lucknow-49	7.46	0.14	2.72	226.58	2.59
11	Pant Prabhat	3.99	0.10	1.09	308.00	2.00
12	Punjab Pink	7.59	0.21	2.79	227.39	2.48
13	Red peel	5.35	0.08	1.19	262.15	2.53
14	Red type	7.78	0.05	2.72	224.37	2.68
15	Sasni collection	4.33	0.06	1.14	296.78	2.87
16	Sasri selection	6.20	0.09	2.20	247.20	2.81
17	Shweta	10.24	0.24	4.25	190.15	2.39
18	Snow White	7.46	0.14	2.72	226.00	2.28
19	Sour Type	4.93	0.05	1.27	295.64	2.53
20	T.N selection	7.46	0.08	2.66	226.27	2.40
21	Thai guava	5.35	0.01	2.45	262.15	2.00
22	Yellow type	6.17	0.13	2.19	248.54	2.74
	SEm±	0.576	0.001	0.012	6.93	0.026
	$CD_{0.05}$	1.793	0.003	0.036	25.47	0.059

P_N, photosynthetic rate; g_s, stomatal conductance; E, transpiration rate; C_i, internal leaf CO_2; WUE, water use efficiency.

Stomatal conductance is an important biological determinate of carbon accumulation and transpiration by plants, because the flow of CO_2 into the leaf is controlled by stomatal regulatory processes. Stomatal conductance of guava cultivars significantly varied within the genotypes. Differences in Stomatal conductance among cultivars were observed [Table-1]. The maximum stomatal conductance was recorded in Shewta (0.24mol $m^{-2}s^{-1}$) and minimum in Thai guava (0.01mol $m^{-2}s^{-1}$). In annual plants stomatal conductance was found to be related to yield in some instances. Ulloa *et al*. [20] confirmed that high stomatal conductance was associated with high cotton lint yields at supra-optimal temperatures for the cotton crop under irrigated environments. However, unlike cotton, fruit trees are perennial plants with more complex physiological features that are difficult to be related. In apples the stomatal behaviour of the leaves appears to be correlated with photosynthetic rate [21]. Several factors are known to control stomatal conductance, such as light, soil water potential, internal CO_2 concentration as well as sink strength in trees. But, genotypical differences in stomatal conductance were often neglected. The results of this study showed that stomatal conductance values of the guava genotypes grown in the same conditions greatly varied. Also, a low stomatal sensitivity to drought in benefit of an increase in growth would probably be a more successful strategy under the competitive conditions during tree establishment [22]. The stomatal conductance seems to be independent of stomatal frequency or stomata size, since there was no significant relation between them in all genotypes tested (data not shown). The conductance values can be taken into consideration as possible selection criteria for apricot genotypes to regions with higher summer temperatures.

Internal CO_2 concentration of leaves has a profound effect on CO_2 assimilation rate, though in the present study significant variations were observed in tested cultivars [Table-1]. The lowest Internal CO_2 concentration was found to be in Shweta (190.15 ppm) and Hafsi red (201.50 ppm) and the highest in Arka Amulya(327.00 ppm). In many species leaf stomatal CO_2 concentration tends to remain constant over a range of environmental conditions [23]. For many years, internal CO_2 concentration (Ci) was considered to link stomatal responses to photosynthetic demands for CO_2 [24]. For example, when A increases due to an increase in irradiance, Ci is reduced and stomata respond to the increased demand for CO_2 by increasing aperture; conversely, when the demand for CO_2 decreases, high Ci results in stomatal closure. However, relatively recent research from several laboratories has suggested that Ci is not the only determinant of the coordination between A and gs. von Caemmerer *et al*. [25] suggested that guard cells may not sense Ci but instead may sense external (CO_2), while other reports have suggested that stomatal responses to Ci are too small to account for the observed change in gs in response to light [26]. More recent studies on transgenic plants have shown that gs increases with photosynthetic photon flux density even in plants with reduced A and higher Ci values [25, 27], which agrees with reports that gs responds to various stimuli even when Ci is held constant [28].

Plant gas exchange is a key process shaping global hydrological and carbon cycles and is often characterized by plant water use efficiency (WUE - the ratio of CO_2 gain to water vapor loss). The results presented in [Table-1] showed that Water use efficiency is highest in Allahabad Safeda (2.89 µmol mol^{-1}) and lowest in Black guava (2.00 µmol mol^{-1}). Water use efficiency provides information regarding the carbon fixed by photosynthesis per unit of water lost by stomatal conductance and transpiration or capacity of plants to preserve water and maximizing carbon fixation [29]. The variation among cultivars for these characters might be due to unique genetic features of individual cultivars under the same environmental condition.

Light harvesting pigments

The light harvesting pigments are involved in light capture and photosynthesis in leaves. Hence, changes in pigment content in leaves can affect photosynthesis of plants [17]. They also reported that variations in leaf morphological characters and chlorophyll content directly influence the leaf gas exchange. In guava, the pigment content is influenced by different seasons, cultivars, growth and maturity stages of leaves [30]. In particular, with regards to the total chlorophyll content [Table-2]' Black guava' had the lowest value (0.16 mg g^{-1} FW) and 'L-49' the highest (1.74 mg g^{-1} FW), whereas, maximum carotenoid was observed in 'Black guava' cultivar (2.94 mg g^{-1} FW) and minimum in 'Hissar safeda' (1.07 mg g^{-1} FW). The cultivars with high chlorophyll content can produce higher biomass and increase photosynthesis. In higher plats, increase in PN with increasing chlorophyll content has been reported [31].

Table-2 *Variation in leaf harvesting pigments of guava cultivars*

S.No.	Genotype	Total chlorophyll (mgg^{-1} FW)	Total carotenoids (mgg^{-1} FW)
1	Allahabad Safeda	1.57	1.17
2	ArkaAmulya	1.03	1.23
3	ArkaMridula	1.27	1.71
4	Behat coconut	0.77	1.23
5	Black guava	0.16	2.94
6	Hafsi Red	0.63	1.52
7	HissarSafeda	1.17	1.07
8	HissarSurkha	0.33	1.86
9	Lalit	0.21	2.14
10	Lucknow-49	1.74	1.33
11	Pant Prabhat	1.65	1.11
12	Punjab Pink	0.84	1.79
13	Red peel	0.57	1.81
14	Red type	0.59	1.87
15	Sasni collection	1.37	1.37
16	Sasri selection	1.43	1.13
17	Shweta	1.63	1.42
18	Snow White	1.67	1.25
19	Sour Type	0.33	1.55
20	T.N selection	0.97	1.85
21	Thai guava	1.30	1.37
22	Yellow type	1.30	1.16
	S.E.m.±	0.31	0.06
	$CD_{0.05}$	0.89	0.18

Conclusion

In conclusion, our results demonstrated that the differences in the functioning of the photosynthetic apparatus and gas-exchange characteristics among the studied genotypes. The study presented herein also provides useful information on photosynthetic traits and gas exchange parameters of guava cultivars. Basic information thus obtained would help chalk out a potentially successful breeding programme. Thus, new strategies could be created for new breeding programs of guava cultivars with better adaptation to the limiting agro-climatic conditions of India.

Acknowledgement

I express my sincere thanks to the Head, Division of Fruits and Horticultural Technology, ICAR- IARI, New Delhi, for providing me support and technical guidance in all course of my field and lab work.

Author Contribution: All author equally contributed

Ethical approval: This article does not contain any studies with human participants or animals performed by any of the authors.

Abbreviations: P_N, photosynthetic rate; g_s, stomatal conductance; E, transpiration rate; C_i, internal leaf CO_2; WUE, water use efficiency.

Conflict of Interest: None declared

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