

Relationships between Transpiration Efficiency and Carbon Isotope Discrimination in Chickpea (*C. arietinum* L.)

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Since major cultivation areas of chickpea (*Cicer arietinum* L.) are in the arid and semi-arid zones, terminal drought is one of the major constraints limiting its productivity. Simple analytical crop models can help in identifying key strategies to improve the chickpea productivity under drought. For example, Passioura (1977) had proposed that the yield is a function of transpiration, transpiration efficiency (TE) defined as the biomass production per unit of water transpired, and harvest index. As improvement of TE means maximization of crop production per unit of water use, it is one of the important components for improving the drought resistance (Turner et al. 2001). Although TE had been recognized as a highly relevant trait, so far very little research effort had been made towards field screening for it, especially due to the difficulties in measuring TE in any screening method. The method developed by Farquhar et al. (1982) for estimating TE through measuring the discrimination against ¹³C by leaves during photosynthesis, and establishment of a close relationship between the carbon isotope discrimination ($\delta^{13}\text{C}$) and TE in many legume crops such as bean, cowpea, groundnut, and soybean has provided an useful method of screening. This gave scope for using $\delta^{13}\text{C}$ as an indirect screening tool for TE. In chickpea, however, there is no information available on the relationship between $\delta^{13}\text{C}$ and TE. The major objectives of this study were to check if there are any variations available for $\delta^{13}\text{C}$, to investigate the relationship between $\delta^{13}\text{C}$ and TE, and to ascertain the possibility of using $\delta^{13}\text{C}$ as a surrogate for TE measurements.

Ten chickpea (*C. arietinum* L.) genotypes (Annigeri, ICC 10448, ICC 13219, ICC 14199, ICC 1882, ICC 283, ICC 4958, ICC 5337, ICC 5680 and ICC 8261) with contrasting growth duration, type (desi or kabuli), growth habits, and root systems were used. The pot experiments

were conducted in a randomized block design (RBD) with two irrigation schemes plus pre-irrigation treatment harvest set in 5 replications in a greenhouse facility at the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) in 2004. At 30 days after sowing (DAS), pre-irrigation treatment (five) plants were harvested inclusive of roots from each genotype. At the same time, the soil in pots of both irrigation treatments was saturated with water to bring it to field capacity. All pots were then covered with polyethylene bags, leaving the plants outside to avoid evaporation, and short straw pipes were inserted for further irrigations. The daily transpiration was estimated as the difference in pot weight between two subsequent days. In the well-watered pots (control), the water lost in a day was added back, whereas in the water stress-imposed pots the water, which is equivalent to 70–90% of daily transpiration, was given to avoid the rapid build up of soil water stress. To monitor the daily available soil moisture, the daily transpiration rates (TR) in the stress condition were normalized against the transpiration rates measured in control plants on each day. The experiment was terminated when the TR of water-stressed plants fell below 0.1 (less than 10% of transpiration of control), which is considered as the point where plants are no longer able to take up water from the soil, and where all the physiological processes contributing to growth are fully inhibited. At this time, the 4th and the 5th most fully expanded leaves from the top leaf on the main stem were collected in all plants for $\delta^{13}\text{C}$ estimations. At the same time the entire plant parts, including the roots, were harvested to estimate final plant biomass. The total transpiration was calculated as a sum of the daily transpiration from the initial day when plants were bagged to the day when plants were

Table 1. Analysis of variance and its significance for water schemes, genotypes, and their interaction for carbon isotope discrimination ($\delta^{13}\text{C}$), and transpiration efficiency (TE) in ten chickpea genotypes grown under well-watered (control) and drought stress conditions in a pot experiment.

Source of variation	Mean sum of squares and significance level ¹	
	$\delta^{13}\text{C}$	TE
Irrigation scheme	146.83***	20.78***
Genotype	1.25***	0.49***
Genotype × Irrigation scheme	1.56***	0.05 NS
Residual	0.16	0.03

1. Significant at *** = <0.001 level and NS = Not significant.

harvested. The TE, therefore, could be calculated as the plant biomass gained between the first and final plant sampling divided by total transpiration during that period.

Analysis of $\delta^{13}\text{C}$ was carried out at International Research Center for Agricultural Sciences (JIRCAS), Tsukuba, Japan with the use of an isotope ratio mass spectrometer (IRMS), ThermoFinnigan Delta XP^{plus}, Hamburg, Germany, connected with an element analyzer (EA), Carlo Erba EA Flash 1112, Milan, Italy. Total carbon in leaf samples was incinerated in a furnace of EA and separated as pure CO_2 gas. A small quantity of the gas was introduced to IRMS to measure the ratio of $^{13}\text{CO}_2/^{12}\text{CO}_2$ as the different mass weight of 45/44 to obtain $\delta^{13}\text{C}$ (‰).

There were significant differences in $\delta^{13}\text{C}$ among the ten genotypes, and the $\delta^{13}\text{C}$ in stress condition was significantly higher than that in the well-watered control (Table 1). Genotype ICC 5337 showed the highest $\delta^{13}\text{C}$ (-26.0‰) in the stress condition. ICC 4958, a well known drought resistant variety, had a superior $\delta^{13}\text{C}$ value than the other genotypes. Also ICC 4958 ranked second

(-27.2‰) under stress condition and the first (-28.4‰) in the well-watered control condition. The genotype by irrigation ($G \times I$) interaction was significant for $\delta^{13}\text{C}$.

Among the ten genotypes, significant difference in TE was observed in both irrigated and stress conditions (Table 1). Genotype ICC 5337 showed the highest TE irrespective of irrigations of 3.9 g kg^{-1} under stress and 2.8 g kg^{-1} under well-watered control. The TE under stress was significantly higher than TE under control. There was a significant correlation in TE between the stress and control conditions ($r = 0.881$, $p < 0.01$), and there was no $G \times I$ interaction observed. This is indicative of the genotypic difference in TE and their rankings would remain across different soil water environments.

A significant positive correlation between $\delta^{13}\text{C}$ and TE was observed ($r = 0.857$, $p < 0.01$) under the stress condition (Fig. 1). This relationship agrees with the theoretical relationship between $\delta^{13}\text{C}$ and TE as observed in several other legumes. However, no significant correlation was observed between $\delta^{13}\text{C}$ and TE when the

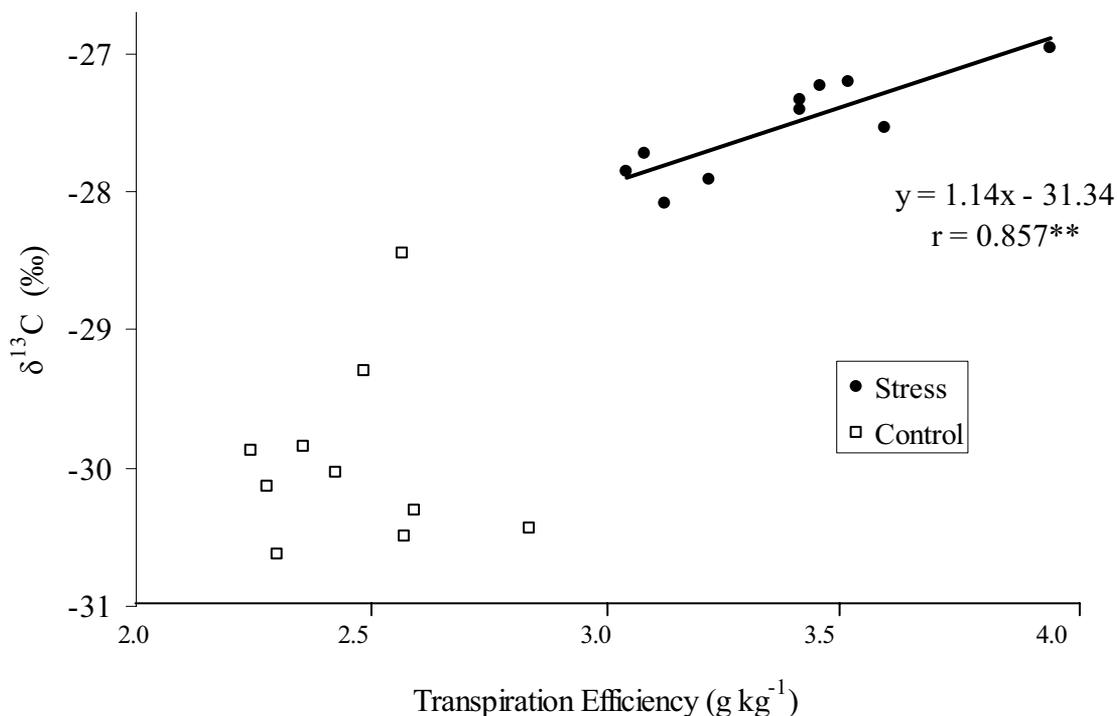


Figure 1. Relationship between transpiration efficiency (TE) and carbon isotope discrimination ($\delta^{13}\text{C}$) in ten chickpea genotypes grown under the well-watered (control) and drought stress conditions in a pot experiment.

plants were grown under well-watered conditions. A similar result has been obtained in sunflower (Virgona et al. 1990). This would indicate that the ^{13}C discrimination ability manifests into TE under water-limited conditions whereas under well-watered conditions the stomatal closure-led CO_2 limitation no longer becomes a constraint to C sequestration in plants. Our results in chickpea may indicate that the differences in TE are brought about by changes in stomatal conductance rather than by changes in mesophyll efficiency.

This is the first report to show the existence of a clear relationship between $\delta^{13}\text{C}$ and TE in chickpea. This result shows that TE of chickpea grown under drought conditions could be estimated through $\delta^{13}\text{C}$ measurement. Further evaluation of these chickpea genotypes for TE in field grown conditions is being carried out during 2006/07 to confirm the results obtained. Any queries related to this study may be directed to Dr J Kashiwagi, Associate Scientist, Crop Physiology, ICRISAT.

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