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Effect of end of season water deficit on phenolic compounds in peanut genotypes with different levels of resistance to drought

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ABSTRACT

Terminal drought reduces pod yield and affected the phenolic content of leaves, stems and seed of peanut (*Arachis hypogaea* L.). The aim of this study was to investigate the effects of end of season water deficit on phenolic content in drought tolerant and sensitive genotypes of peanuts. Five peanut genotypes were planted under two water regimes, field capacity and 1/3 available water. Phenolic content was analyzed in seeds, leaves, and stems. The results revealed that terminal drought decreased phenolic content in seeds of both tolerant and sensitive genotypes. Phenolic content in leaves and stems increased under terminal drought stress in both years. This study provides basic information on changes in phenolic content in several parts of peanut plants when subjected to drought stress. Future studies to define the effect of terminal drought stress on specific phenolic compounds and antioxidant properties in peanut are warranted.

1. Introduction

Peanut (*Arachis hypogaea* L.) grown in arid and semi-arid areas is often subjected to drought during one or more stages of growth. Terminal (end of season) drought greatly reduces pod yield and also increases aflatoxin contaminations (Girdthai et al., 2010). Thus, terminal drought is considered to be a major problem in peanut production.

Peanut is a source of secondary metabolites beneficial to health. It is rich in nutrients such as protein, minerals, vitamins, fatty acid, fiber and phenolic compounds. Phenolic compounds are secondary metabolites in plants, which are derived from phenylpropanoid pathway (Vogt, 2010). Phenolic compounds have several health benefits including anti-oxidant, anti-inflammatory, anticancer and inhibition of cardiovascular disease (Chen & Blumberg, 2008). Wang, Yuan, Jin, Tian, and Song (2007) also reported that phenolic compounds from peanut skins at 500 μg/ml exhibited DPPH radical scavenging activity up to 97.1%.

The effect of drought stress on phenolic compounds has been reviewed by many researchers in many plant species and in different plant parts. Phenolic compounds in leaves of many crops such as in rice (Ayaz, Kadioglu, & Turgut, 1999), maize (Hura, Hura, & Grzesiak, 2008) and *Hypericum brasiliense* Choisy (Abreu & Mazzafera, 2005) generally increased under drought stress condition. However, drought stress reduced phenolic concentration in leaves of cotton (Yildiz-Aktas, Dagnon, Gurel, Gesheva, & Edreva, 2009), tea (Cheruiyot, Mumera, Ngetich, Hassanali, & Wachira, 2007) and cherry tomato (Sánchez-Rodríguez, Moreno, Ferreres, Rubio-Wilhelmi, & Ruiz, 2011). Drought also increased phenolic content in cumin seeds (Iness et al., 2012), but it reduced phenolic content in kernel oil of maize genotypes (Ali, Muhammad, & Farooq, 2010). Similar results were also found in rape seeds, where drought stress decreased phenolic compounds at early growth and flowering stages (Bouchereau, Clossais-Besnard, Bensaoud, Leport, & Renard, 1996).

Maize genotypes with resistance to drought stress increased the total phenolic content in leaves under water deficit treatment, whereas moderate and sensitive genotypes did not increase the total phenolic content (Hura et al., 2008). Drought reduced polyphenols in all cotton genotypes irrespective of drought resistance levels (Yildiz-Aktas et al., 2009).

Thus, the literature documents that plant phenolic content will differ in response to drought, and this response differs among species, plant parts and the drought tolerance of genotypes within a species. Yet, drought-induces changes in phenolic compounds in peanut have not been thoroughly investigated. The variation in phenolic compounds might depend on peanut genotypes and period of drought conditions. Peanuts with different levels of drought

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tolerance may respond differently to drought-induced changes in phenolic compounds. The objective of this study was to investigated the effect of terminal drought on phenolic compounds in peanut genotypes with different levels of drought resistance. The information obtained may help breeders to select peanut genotypes with high phenolic compounds in seeds and drought resistance.

2. Materials and methods

2.1. Experimental details

Peanut plants were grown under field conditions. The experiment was conducted at Khon Kaen University, Thailand during October 2011–February 2012 and repeated during October 2012–February 2013. A split-plot design with four replications was used in this experiment and the sub-plots were randomly arranged in the main plots. Main-plots were two water treatments [field capacity (FC) and 1/3 available water (1/3 AW) from R7 (beginning maturity with one pod showing visible natural coloration or blotching of inner pericarp or testa) (Boote, 1982) to harvest (two-thirds to three-fourths of all developing pods have testa or pericarp coloration) (Boote, 1982)]. One-third available water was used in this study because 1/4 AW was too severe and peanut genotypes were not clearly different when subjected to this level of stress (Vorasoot, Songsri, Akkasaeng, Jogloy, & Patanothai, 2003) and drought at 1/3 AW was more suitable for evaluation of root length density in deeper soil layer (Songsri et al., 2008).

The five peanut genotypes randomized within each subplot included ICGV 98348, ICGV 98324, ICGV 98308, Tifton 8, and Tainan 9. These genotypes were selected because of differences in their drought tolerant index (DTI) ratings (Girdthai et al., 2010). ICGV98348 and ICGV98324 are drought resistant genotypes with high DTI for mass and pod yield. Tifton 8 is a drought tolerant genotype with a large root system from the United States Department of Agriculture (USDA). ICGV 98308 and Tainan 9 are classified as drought susceptible genotypes with high reduction in total mass and pod yield under terminal drought stress. Details of crop management were described in a previous report (Koolachart et al., 2013).

2.2. Water management

A drip-irrigation system was set up before sowing and each sub-plot was supplied with sufficient water to get FC up to a depth of 60 cm. Soil moisture content was irrigated daily to FC from sowing to R5 growth stage (beginning seed defined as one fully-expanded pod in which seed cotyledon growth is visible) (Boote, 1982) for all treatments. After R5 growth stage, the non-stress treatment was maintained at FC until harvest. For the stress treatment, water was withheld at the R5 growth stage of each genotype and soil moisture was allowed to decrease gradually to meet the predetermined level of 1/3 AW at R7 stage. The times from R5 to R7 growth stage for each genotype were slightly different between years. However, they were different among peanut genotypes (Table 1). The level of 1/3 AW was determined from a model simulation which used 20 years of historical pan evaporation data. At any time the soil moisture obtained 1/3 AW at 60 cm soil depth, it was kept at the level of 1/3 AW up to harvest. Soil moisture content was checked by gravimetric methods. Crop water requirement was based on Doorenbos and Pruitt (1992) and surface evaporation was calculated as published by Singh and Russel (1981).

Table 1
Days from R5 to R7 growth stage of five peanut genotypes when grown under field capacity (FC) and terminal drought stress (1/3 AW) in 2011/2012 and 2012/2013.

Genotype	2011/2012		2012/2013	
	FC	Stress	FC	Stress
ICGV 98348	29	27	26	24
ICGV 98324	26	22	23	21
Tifton 8	36	27	33	29
ICGV 98308	27	22	23	21
Tainan 9	27	25	26	24

2.3. Data collections

2.3.1. Leaves relative water content (RWC)

Relative water content was measured from five leaflets (the second fully expanded leaf) of each plot during 10:00 AM to 2:00 PM. The leaves were harvested and put quickly in a plastic zip bag for preventing water loss from the leaves. All plastic bags were kept in the ice box and transported to the laboratory, leaf fresh weights were recorded. The leaf samples were then soaked in distilled water and placed in a dimly lit room at 26 °C for 8 h, before blotting the surface dry and measuring the water saturated leaf weight. The samples were then oven-dried at 80 °C until reaching a constant weight and leaf dry weight was determined. RWC was calculated based on the formula suggested by Gonzalez and Gonzalez-Vilar (2001).

2.3.2. Phenolic compounds analysis in seeds

Mature seeds from five plants of each subplot were collected and air dried to approximately 8% moisture content. Seeds were then oven dried at 80 °C until reaching a constant weight. Seeds with skins were ground using a grinding machine and sub samples of 10 g each were ground again using a mortar and pestle. Forty milliliters of methanol were added to each sample and stirred for 2 h at room temperature. Each beaker was covered with aluminum foil and after 2 h the samples were filtered through Whatman No. 4 paper. The methanol solutions were then evaporated at 60 °C using a rotary evaporator. The methanol extracts were collected and blown with nitrogen gas until 2 milliliters were obtained. The methanol extracts of peanut were kept at 4 °C until analyzed. Total phenolic content was determined by the Folin–Ciocalteu's assay (Torres, Mau-Lastovicka, & Rezaaiyan, 1987). The phenolic content was demonstrated as gallic acid equivalent (mg GAE/10 g dry weight of peanut seed).

2.3.3. Phenolic compounds analysis in leaves and stems

Phenolic compounds were analyzed from leaves and stems at R5 and R7 growth stages, and at harvest. Five plants were selected at random from each subplot and separated into leaf, stem and seed. Leaves and stems were oven dried at 80 °C for 48 h or until the weights were constant. Leave and stems were then ground using a Wiley Mill and sub-samples of two grams were taken and further ground using a mortar and pestle. The extraction and analysis processes were the same as with the seeds.

2.3.4. Yield and drought tolerance index (DTI)

At harvest, 5 plants of each subplot were used to determine pod yield. Pods were separated from the plant and air dried to approximately 8% moisture content and weighed. Pod dry weight per plant was measured and drought tolerance index (DTI) for pod yield was calculated as pod yield under 1/3 AW divided by pod yield under FC condition as suggested by Girdthai et al. (2010).

2.4. Statistical analysis

The statistical analysis was conducted using Statistic 8 analytical software (Statistix8, 2003). Error variance of two years was