Effect of Soil Temperature on the Seed Composition of Three Spanish Cultivars of Groundnut (*Arachis hypogaea* L.)

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The influence of the soil temperature regimes 20/14 (T1), 26/20 (T2), 32/26 (T3), and 38/32 °C (T4) (day/night) on seed composition of three Spanish genotypes of groundnut (Arachis hypogaea L.) was investigated. From T1 to T2 the oil concentration increased and the starch concentration decreased remarkably, but only slightly to T3. The protein concentration was higher in the two warmer soil temperatures than in the colder temperatures. There was no difference between T3 and T4 for oil, starch, and protein concentration. The total soluble sugar concentration was higher in both extreme temperature regimes than in the intermediate treatments. The oleic/linoleic acid ratio increased from T1 to T3. The results show that soil temperature has a marked effect on the proximate composition and fatty acid profile of these groundnut cultivars.

Keywords: Groundnut; seed composition; soil temperature; oil; fatty acid composition; starch; protein; total soluble sugar

INTRODUCTION

Although groundnut (Arachis hypogaea L.) seeds mature in the soil and groundnut is grown over a wide range of soil temperatures, there is little information about the effect of soil temperature on seed composition. Even slight variations in mean soil temperature can result in pod maturation, yield, and seed size differences (Ono et al., 1974; Dreyer et al., 1981; Sanders et al., 1984). Because groundnut seeds mature 3-10 cm below the soil surface, soil temperature rather than air temperature is probably the governing factor in groundnut seed lipid biosynthesis (Brown et al., 1975). It has been reported that the fatty acid composition of seed oils of a number of plant species other than groundnut, such as sunflower, flax, and rape, varies according to the temperature at which the seeds have developed, with low temperatures favoring a more unsaturated oil (Canvin, 1965; Slack and Browse, 1984). McMeans et al. (1990) observed a significant difference in free carbohydrate concentrations in groundnut seeds grown at different soil temperatures.

More information about the influence of soil temperature on groundnut seed quality may be useful to guide the choice of planting location, sowing date, and crop management according to the purpose of the groundnut production and vice versa. Hence, the purpose of this study was to investigate the effect of soil temperature on seed composition of groundnut grown under controlled soil temperatures.

EXPERIMENTAL PROCEDURES

Materials. The experiment was conducted in a greenhouse at Patancheru, near Hyderabad, India (17° 30′ N, 78° 16′ E). The groundnut (A. hypogaea L.) cultivars used in this experiment belong to subsp. fastigiata var. vulgaris, Spanish type, and are TMV 2, AH 6179, and Comet. The Spanish type of groundnut was chosen for this experiment because of its short time to maturity combined with high yield. This is important in the semiarid tropics and in many other areas for fitting into

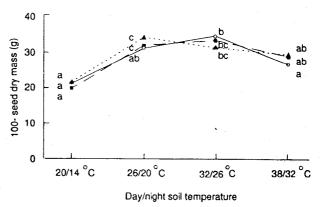


Figure 1. Effect of soil temperature and cultivar on the 100-seed dry mass: (O) Comet; (\blacktriangle) TMV 2; (\blacksquare) AH 6179. Means within one variety are significantly different at the $P \le 0.05$ level if followed by different letters.

crop rotations and to escape diseases and where growing seasons are short. The cultivars were selected for similar time to flowering and maturity, 100-seed mass, and number of seeds per pod according to tests in several locations. TMV 2 is commonly grown in India, AH 6179 performed relatively well in the hot environment of ICRISAT Sahelian Center, Niger (J. H. Williams, 1993, personal communication), and Comet produced a relatively high yield in comparison with other varieties grown in the relatively cool climate of Ontario, Canada (Roy et al., 1980; Court et al., 1984).

Individual plants were grown in 7 L containers with a 4:2:1 mixture of Alfisol soil, sand, and vermiculite. The rooting medium was inoculated with Rhizobium, and an application of 6.5 mg of single superphosphate/kg of mixture was given to optimize the phosphorus availability. All other nutrients were in the optimal range, as determined by soil analysis. Irrigation was applied twice daily so as to maintain the soil medium near field capacity (9.27%). Soil temperature treatments of 20/14 (T1), 26/20 (T2), 32/26 (T3), and 38/32 °C (T4) day/night with a 12 h "day" period and a 12 h "night" period were imposed by placing the pots in large temperature-controlled water baths. Per treatment (water bath) there were 12 containers, 4 containers with single plants of each of the three cultivars. The adjustment period between day and night soil temperature was about 2 h. This soil temperature course simulates natural conditions better than a sudden temperature change. The temperatures were measured with copper/constantan thermo-

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Table 1. Effect of Soil Temperature on Proximate Chemical Composition of Mature Groundnut Seeds^a (Percent Dry Mass)

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cultivar	day/night soil temp (°C)	oil	protein	starch	total soluble sugars	remaining
Comet	20/14	43.5a	21.8a	24.4c	4.3b	6.0a
	26/20	50.0^{b}	22.3^{a}	$17.7^{\rm b}$	3.8^{ab}	6.2ª
	32/26	51.8^{bc}	26.6^{b}	11.9^{a}	3.2^{a}	6.5ª
	38/32	53.4^{c}	23.7^{ab}	12.2^{a}	4.0^{ab}	6.7^{a}
TMV 2	20/14	44.3^{a}	21.2^{a}	24.6^{c}	4.5^{a}	5.4a
	26/20	49.4^{b}	23.1^{ab}	$17.4^{\rm b}$	3.9^{a}	6.2ª
	32/26	$50.4^{\rm b}$	$26.2^{\rm bc}$	13.8^{a}	3.7^{a}	5.9a
	38/32	49.4^{b}	27.9°	11.8a	4.5^{a}	6.4a
AH 6179	20/14	40.1^{a}	22.6^{ab}	27.4^{c}	$4.8^{\rm b}$	5.1a
	26/20	49.3^{b}	21.7^{a}	$18.5^{\rm b}$	3.4^{a}	$7.1^{\rm b}$
	32/26	51.6^{b}	25.7^{c}	11.9ª	3.2^{a}	7.6^{b}
	38/32	$51.4^{\rm b}$	25.3^{bc}	12.2a	4.5^{b}	6.6^{b}
mean	20/14	42.6^{a}	21.9^{a}	25.5^{c}	$4.5^{\rm b}$	5.5a
	26/20	$49.6^{\rm b}$	22.4^{a}	17.9^{b}	3.7^{a}	6.5 ^b
	32/26	51.2^{c}	26.2^{b}	12.5^{a}	3.4^{a}	$6.7^{\rm b}$
	38/32	51.4^{c}	$25.6^{\rm b}$	12.0^{a}	$4.3^{ m b}$	$6.6^{\rm b}$
	\mathbf{SE}	± 0.9	± 0.9	± 1.2	± 0.3	± 0.5

 $[^]a$ Means within one physiological parameter and variety are significantly different at the $P \leq 0.05$ level if followed by different letters.

couples and recorded with a datalogger. In the greenhouse the air temperature range was 24-35 °C during the day and 20-27 °C during the night.

The plants were grown outside the water baths in the greenhouse until the start of the treatments. Fifty percent germination was observed at 7 days after sowing (DAS). At the 75% flowering stage (53 DAS in all cultivars) plants were selected for uniformity and subjected to temperature treatments which continued until the final harvest. The plants were harvested at maturity, 109 days after sowing.

Maturation was determined by the hull-scrape method (Williams and Drexler, 1981). The 100-seed mass was calculated by extrapolating from the seed number and seed mass of each single plant. Mature peanut seeds were freeze-dried, ground into a meal using a Waring blender, defatted, and stored at 0–4 °C for further analysis. The 100-seed mass, chemical composition, and fatty acid composition were determined of the mature seeds of each single plant of the experiment.

Chemical Composition. Nitrogen concentration was determined using a Technicon autoanalyzer (Singh and Jambunathan, 1980). A factor of 5.46 was used for converting the nitrogen into crude protein concentration. Oil was determined by extracting the peanut meal with *n*-hexane in a Soxhlet apparatus (AOCS, 1980). Total soluble sugars in peanut meal

were extracted with hot 80% aqueous ethanol and determined according to the procedure of Dubois $et\ al.$ (1956). Starch was determined by hydrolyzing the meal with amyloglucosidase enzyme (Thivend $et\ al.$, 1972) and analyzing the sugars in the hydrolysate according to the procedure of Dubois $et\ al.$ (1956).

Fatty Acid Composition. Fatty acid methyl esters (FAME) of hexane extracts were prepared using 14% (w/v) boron trifluoride in methanol (Metcalfe et al., 1966) and analyzed according to the method of Mercer et al. (1990) with the following modifications. Gas chromatography analysis was performed on a Shimadzu Model GC-9A gas chromatograph (Shimadzu Corp., Tokyo, Japan) fitted with a flame ionization detector and a glass column (210 mm \times 3 mm i.d.) packed with Altech CS-10 W-AW (80-100 mesh) (Alltech Associates, Deerfield, IL). The carrier gas helium was maintained at a flow rate of 50 mL/min. The temperatures of the injector and detector were maintained at 250 and 300 °C, respectively. The column temperature was held at 190 °C for 4 min initially, programmed to increase from 190 to 250 °C at 10 °C/min, and held at 250 °C for 2 min. Peaks were identified by using standard FAME and quanitified by using methyl heptadecanoate (17:0) as internal standard.

Statistical Analysis. The experimental design was a split plot with four replications, in which the soil temperatures were the main plots and the genotypes the subplots. The statistical analysis was performed by standard ANOVA using the Genstat (1977) package and the comparison of means was conducted by using Duncan's multiple range test.

RESULTS AND DISCUSSION

Mature Seed Dry Mass and Number per Plant. The 100-seed mass (Figure 1) as well as the number of mature seeds per plant (data not shown) increased with a rise in soil temperature from 20/14 to 26/20 °C and decreased from 32/26 to 38/32 °C.

Proximate Chemical Composition. The oil concentration of the mature seeds increased remarkably in all cultivars from 20/14 to 26/20 °C, and slightly from 26/20 to 32/26 °C; up to 38/32 °C a further slight increase in Comet, but not in the other cultivars (Table 1), was seen. The protein concentration was higher in the two warmer treatments as compared to the lower temperature treatments (Table 1).

With an increase of the 100-seed mass of groundnut within the range of the values of our experiment, Mishra and Gaur (1982) observed an increase in oil concentration and Ponnuswamy and Ramakrishnan (1984) an increase in protein concentration of the seeds. Therefore, it cannot be excluded that the increase in oil as

Table 2. Effect of Soil Temperature on Fatty Acid Composition of Mature Groundnut Seeds^a (Percent of Total Fatty Acids)

ricius)						1.4				
cultivar	day/night soil temp (°C)	$16:0^{b}$	18:0	18:1	18:2	20:0	20:1	22:0	24:0	O/L ratio
Comet	20/14	12.8ª	2.4 ^a	36.6ª	38.9°	1.4ª	1.8°	4.7°	1.3ª	0.94ª
	26/20	12.7^{a}	$2.9^{ m ab}$	40.8^{b}	36.5°	1.4a	1.1^{b}	$3.5^{\rm b}$	1.0a	1.12^{b}
	32/26	13.2ª	3.1^{b}	44.4 ^c	$32.3^{\rm b}$	1.4^{a}	0.9^{a}	3.0^{a}	1.2a	1.38^{c}
	38/32	13.0^{a}	3.8^{c}	47.2^{c}	29.5^{a}	1.6^{a}	0.9a	2.8^{a}	1.1a	1.60^{d}
TMV 2	20/14	12.8^{ab}	2.5^{a}	36.6^{a}	38.8^{c}	1.4^{a}	$1.7^{\rm c}$	4.4^{c}	1.4a	0.94^{a}
	26/20	12.4^{a}	$3.1^{\rm b}$	$41.0^{ m ab}$	$36.0^{\rm b}$	1.5^{a}	$1.1^{\rm b}$	$3.8^{\rm b}$	1.0^{a}	$1.14^{ m b}$
	32/26	$13.0^{ m ab}$	3.4^{b}	44.2^{bc}	32.4^{a}	1.5ª	1.0^{a}	3.3^{a}	1.2^{a}	1.36^{c}
	38/32	$13.4^{ m b}$	$3.2^{\rm b}$	44.9°	31.6^{a}	1.5^{a}	0.9^{a}	3.1ª	1.4^{a}	1.42^{c}
AH 6179	20/14	12.9^{ab}	2.6^{a}	36.3ª	38.4^{c}	1.5^{a}	$1.8^{\rm b}$	5.1°	$1.3^{ m ab}$	0.95^{a}
	26/20	12.3a	3.9^{b}	40.5 ^b	$35.7^{\rm b}$	1.8^{b}	1.0^{a}	3.9^{b}	$0.9^{ m ab}$	1.13 ^b
	32/26	12.9^{ab}	3.4^{b}	45.4°	32.0^{a}	1.5^{a}	0.9ª	3.1ª	0.8ª	1.42a
	38/32	$13.3^{\rm b}$	3.3^{b}	43.9^{bc}	32.3^{a}	1.6^{ab}	0.9ª	3.3ª	$1.4^{ m b}$	1.36^{c}
mean	20/14	12.8^{b}	2.5^{a}	36.5ª	38.8^{c}	1.4^{a}	1.8^{c}	4.7^{c}	1.3^{c}	0.94^{a}
	26/20	12.5a	3.3^{b}	40.8^{b}	$36.1^{\rm b}$	1.6^{b}	1.1^{b}	$3.7^{\rm b}$	1.0^{a}	1.13^{b}
	32/26	13.0^{bc}	3.3^{b}	44.7^{c}	32.2^{a}	1.5^{ab}	0.9ª	3.1a	$1.1^{ m ab}$	1.39^{c}
	38/32	13.2^{c}	$3.4^{\rm b}$	45.3°	31.1^{a}	1.6^{b}	0.9^{a}	3.1^{a}	$1.3^{ m bc}$	1.46 ^d
SE		± 0.3	± 0.2	± 1.2	± 0.8	± 0.07	± 0.04	± 0.15	± 0.17	± 0.07

^a Means within one physiological parameter and variety are significantly different at the $P \le 0.05$ level if followed by different letters. ^b Figure before colon indicates the number of carbon atoms, and the figure after the colon is the number of double bonds in the fatty acid chain.

well as protein concentration from 20/14 to 26/20 °C was also due to a temperature effect on seed size, although with the decrease in 100-seed mass from 32/26 to 38/32 °C there was no change in oil and protein concentration. However, Dwivedi *et al.* (1990) found no significant association of seed mass with oil or protein contents among 64 genotypes.

The starch concentration decreased from 20/14 to 38/32 °C in all cultivars (Table 1). The sharp decrease in starch concentration from 20/14 to 26/20 °C was inversely related to the marked increase in oil concentration. From 26/20 to 32/26 °C the starch/oil ratio decreased further.

With increasing soil temperature the total soluble sugar concentration decreased from 20/14 to 26/20 °C and increased with a further rise in temperature from 32/26 to 38/32 °C (Table 1). McMeans et al. (1990) observed within each seed size category a decrease in soluble sugar concentration with an increase in soil temperature from about 22 to 29 °C, which is in conformity with our results. They also found a reduction in sugar concentration with increasing seed size. On the contrary, Pattee et al. (1981) observed an increase in soluble sugar concentration with increasing seed size. Therefore, in our experiment the influence of soil temperature on the sugar concentration may be also due to an effect on seed size.

Fatty Acid Composition. Soil temperature markedly affected the composition of the major fatty acids. Oleic acid (18:1) increased and linoleic acid (18:2) decreased in all cultivars with an increase in soil temperature from 20/14 to 32/26 °C (Table 2). Comet showed a further decrease in linoleic acid from 32/26 to 38/32 °C. Therefore, an increase in soil temperature from 20/14 to 32/26 °C led in all cultivars to an increase in oleic/linoleic acid ratio (O/L) from values of 0.95 to values between 1.36 and 1.42. The O/L ratio of Comet increased further up to 38/32 °C, to a value of 1.60. A high O/L ratio is desirable for better oil stability or longer shelf life of the groundnut products.

Mozingo et al. (1988) observed with increasing seed size an increase in oleic acid (percent of total fatty acids), a decrease of linoleic acid, and in consequence an increase in O/L ratio. Therefore, an indirect effect of soil temperature in the range from 20/14 to 26/20 °C on fatty acid composition through seed size cannot be excluded.

The major changes in seed oils in response to temperature occur between oleate and polyunsaturated fatty acids (linoleate or linolenate depending on species), indicating that the oleate desaturase is important in determining the temperature effect on the O/L ratio in oil seeds (Browse and Slack, 1983; Slack and Browse, 1984). Browse and Slack (1983) suggest that the effect of temperature on fatty acid composition may also be the result of a difference between the temperature response of fatty acid and triacylglycerol synthesis.

The concentration of behenic acid (22:0), a long-chain saturated fatty acid, was reduced by increasing temperature from 20/14 to 32/26 °C. Long-chain saturated fatty acids have been implicated in the elevated atherogenic effect of groundnut oil (Kritchevsky *et al.*, 1971).

Soil temperature has a marked effect on the proximate composition and fatty acid profile of groundnut seeds in the range from 20/14 to 32/26 °C. Besides the

direct soil temperature effect, an additional indirect effect through seed size on seed composition cannot be excluded.

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