INFLUENCES OF TEMPERATURE ON *ARACHIS HYPOGAEA* L.

WITH SPECIAL REFERENCE TO ITS POLLEN VIABILITY

J. F. DE BEER

STELLINGEN

I

Bij het vaststellen van de levensvatbaarheid van stuifmeel wordt onvoldoende rekening gehouden met de temperatuur waarbij de planten waarvan dit stuifmeel afkomstig is, zijn opgegroeid.

Dit proefschrift.

II

De gevolgtrekking van MOORE dat bij de aardnoot vegetatieve en reproductieve ontwikkeling complementen zijn, is aanvechtbaar.

Dit proefschrift.

III

Het is onjuist dat suikers in het kiemmedium alleen dienen om een gunstige osmotische toestand te scheppen bij de ontkieming en groei van de stuifmeelkorrel.

VISSER, T. *Meded. Landb. Hogesch. Wageningen*, *55* (1955) 1–68.

IV

De opvatting dat accumulatie van nitraatstikstof gedurende de droge seizoenen in de bovenste lagen van tropische gronden veroorzaakt wordt door photochemische of biologische processen, is niet voldoende gegrond.

V

Hulpverlening aan ontwikkelingslanden in tropisch Afrika met het doel produktie en uitvoer van traditionele landbouwprodukten te vergroten, zal de economie van deze landen weinig verbeteren.

VI

Subsidie ter stimulering van de produktie houdt veelal grote gevaren in voor het kleinbedrijf.

VII

De classificatie van tabak in Zuid-Afrika, die hoofdzakelijk berust op verschillen in kleur en lengte van het blad, is onvoldoende om met deze tabak op de wereldmarkt te kunnen concurreren.

VIII

Het biedt geen voordelen Orinoco 'flue cured' tabak, die op noriet gronden verbouwd wordt te bevloeien, alvorens ten minste 90% van het beschikbare bodemvocht verbruikt is.

IX

De dood van roofvijanden als gevolg van bespuitingen met DDT moet niet als voornaamste oorzaak van een snelle populatievergroting der phytophage Arthropoden beschouwd worden.

J. F. DE BEER

Wageningen, 1963

INFLUENCES OF TEMPERATURE ON *ARACHIS HYPOGAEA* L.
WITH SPECIAL REFERENCE TO ITS POLLEN VIABILITY

Dit proefschrift met stellingen van
JOHANNES FREDERIK DE BEER, M.Sc. (Agric.),
geboren te Klerksdorp, Zuid-Afrika, 20 februari 1933,
is goedgekeurd door de promotor
Dr. Ir. J. D. FERWERDA,
hoogleraar in de tropische landbouwplantenteelt.

De Rector Magnificus
van de Landbouwhogeschool
W. F. EIJSVOOGEL

Wageningen, 4 maart 1963

THESIS

SUBMITTED TO
THE STATE AGRICULTURAL UNIVERSITY
WAGENINGEN, THE NETHERLANDS, IN PARTIAL FULFILMENT FOR THE DEGREE OF
DOCTOR OF AGRICULTURAL SCIENCES
ON WEDNESDAY 1 MAY 1963

INFLUENCES OF TEMPERATURE ON *ARACHIS HYPOGAEA* L.

WITH SPECIAL REFERENCE TO ITS POLLEN VIABILITY

PROEFSCHRIFT

TER VERKRIJGING VAN DE GRAAD
VAN DOCTOR IN DE LANDBOUWKUNDE
OP GEZAG VAN DE RECTOR MAGNIFICUS, IR. W. F. EIJSVOOGEL,
HOOGLERAAR IN DE HYDRAULICA, DE BEVLOEIING,
DE WEG- EN WATERBOUWKUNDE EN DE
BOSBOUWARCHITECTUUR,
TE VERDEDIGEN TEGEN DE BEDENKINGEN
VAN EEN COMMISSIE UIT DE SENAAT
VAN DE LANDBOUWHOGESCHOOL TE WAGENINGEN
OP WOENSDAG 1 MEI 1963 TE 16 UUR

DOOR

J. F. DE BEER

CENTRUM VOOR LANDBOUWPUBLIKATIES EN LANDBOUWDOCUMENTATIE
WAGENINGEN, 1963

To my parents

PREFACE

I should like to express my grateful thanks to the committee of the State Agricultural University, Wageningen, The Netherlands, who offered me the opportunity to conduct this investigation.

The author further wishes to express his sincerest thanks to Prof. Dr. J. D. FERWERDA, head of the Laboratory of Tropical Agriculture, under whose keen guidance this investigation was carried out.

Particular mention should be made to Mrs. Dr. FRAHM-LELIVELD for her helpful suggestions, valuable help in preparing the manuscript as well as for writing the Dutch summary.

I am deeply indebted to Mr. G. G. BOLHUIS, who, due to his wide experience with the groundnut plant, made many valuable suggestions.

The help and friendship I received from my colleague Mr. J. F. WIENK, I will keep as a pleasant memory.

To the staff and members of the Laboratory of Tropical Agriculture who helped making my stay such a pleasant one, I extend my greatest thanks.

For her kind assistance in typing the manuscript, I greatfully acknowledge the help of Mrs. J. SNIJDERS. The neat way in which Mr. R. BOEKELMAN prepared the graphs is greatly appreciated.

The author gratefully acknowledges the efficiency and cooperation received from Mr. A. RUTGERS of the Centre for Agricultural Publication and Documentation.

Finally, I should like to thank my parents for their continuous encouragement and financial support.

DRUK: N.V. NOORD-NEDERLANDSE DRUKKERIJ — MEPPEL

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CONTENTS

I INTRODUCTION

Slightly over a century ago, the groundnut was still a food crop of minor importance. Today, however, although essentially a tropical crop, its cultivation extends over vast areas not only in the tropics but also in both sub-tropical and temperate regions. Although publications concerning the physiology of the groundnut have appeared from time to time, especially during the last 10 years, there still exists an urgent necessity to extend and co-ordinate our physiological knowledge with a view to resolve practical problems associated with groundnut cultivation. Too frequently in the past authors have dealt only with mere observational peculiarities without really touching the essentials of the problems. It is therefore not surprising and also with some justification that the groundnut earned the bogus name of '*the peanut, the unpredictable legume*' (30).

As far back as 1938 JODIDI (41) stated that: 'The average yields and efficiency of crop production in general are so far below the apparent possibilities that there is a pressing need for exhaustive fundamental studies of the plant in all stages of development and from numerous points of view'.

The effect of temperature on the development of the groundnut has been investigated by various workers; few, however, have studied the plant under controlled environmental conditions. FORTANIER (27), investigating the influence of some environmental factors on flowering, states that the initiation of flowers is independent of both photoperiod and thermoperiod. The same worker therefore found the groundnut not to respond to any day-night temperature fluctuations, but to be mainly dependent on the average temperature. On the basis of these findings, BOLHUIS and DE GROOT (9) studied the groundnut plant at constant temperatures and found temperature to have a great influence on the number of fruits formed. With the background knowledge that temperature may be regarded as a decisive factor in growth and development, and with increasing evidence that this plant exhibits no thermoperiodicity, we limited ourselves to study the groundnut plant only at different constant temperatures. By carrying out those investigations, we attempted to explain to what extent temperature influences vegetative and generative growth, first and foremost bearing in mind that temperature has a distinct effect on fruit production.

In view of the fact that no single environmental factor acts independently, we should like to stress that our results and conclusions are only valid for the circumstances under which the experiments were carried out.

II MATERIALS AND METHODS

2.1. Plant material

Arachis hypogaea L. belongs to the family of the *Papilionaceae*. The cultivated groundnut is an annual plant which, depending on the cultivar, grows erect (bunch) or prostrate (runner). As the plants had to be grown in pots, the choice of cultivars was restricted to the erect or bunch types.

The influence of temperature on the growth and development was the main object of study, therefore three cultivars from different climatic regions were employed:

(a) Schwarz 21, a cultivar from Indonesia 6–7° lat. S.

(b) A cultivar from Mallorca, 39° lat. N.

(c) A cultivar from the Ukraine, 51° lat. N.

The major part of the experiments was carried out with the Schwarz 21 cultivar, whereas the latter two were used as comparisons. The cultivar mainly used, Schwarz 21, was bred and selected in Java by Schwarz and others (28) some forty years ago and is known to have great resistance against slime disease (*Pseudomonas solanacearum*), the first major disease of groundnuts observed in the East Indies.

Regarding the morphological characteristics of the groundnut, in particular as far as pod shape and size and number of kernels per pod are concerned, the Schwarz 21 and Ukraine cultivars may be considered to belong to the Spanish type, whereas the Mallorca cultivar may be considered as a Valencia type.

2.2. Description of the experimental rooms

2.2.1. Controlled Environment Rooms

In these rooms light, temperature and humidity were controllable. Illumination was supplied by twenty-four 40 W Philips TLF 55 daylight fluorescent tubes fitted in a sliding frame which could be moved upwards and downwards. An intensity of 50,000 erg. $sec.^{-1}$ cm^{-2} was measured at plant level, 15 cm from the lamps. Temperature was controlled by a thermostat between 24° and 33° within 1°C. The humidity was also controllable and was kept at a relative humidity of 75%.

2.2.2. Thermostat Cabinets

In these six cabinets light as well as temperature were controllable. Temperature was controlled between 21° and 36° within 1°C. The cabinets were mainly used for

germination and pollen viability studies. Illumination, when applied, was given by 2 H.O. 450 Watt mercury lamps; a light intensity of 45,000 erg. $sec.^{-1}cm^{-2}$ was measured 20 cm from the glass screen which separated the cabinets from the light source. The relative humidity varied from 50 % to 80 %.

2.2.3. Thermostat Rooms

Also in these rooms light as well temperature were controllable. The light unit consisted of a three stage removable frame with sixteen 40 W Philips TLF 55 daylight florescent tubes. An intensity of 45,000 erg. $sec.^{-1}cm^{-2}$ was measured at plant level 15 cm from the lamps, which approximately equalled the intensity used in the environment rooms. By means of thermostats, 750 W heat elements and ventilators, the temperature was controllable within 2°C during winter. The relative humidity varied from 65% to 75% but was always kept at a high level by means of water baths and water absorbent rags.

(For more detailed information see FORTANIER (27) and SMILDE (79).)

2.2.4. Greenhouse

During the summer periods one greenhouse was used for growing control plants as well as plants later to be used for a more detailed study. Neither temperature nor humidity were controllable in the greenhouse, which made direct comparisons with treatments under controlled conditions somewhat difficult.

2.3. EXPERIMENTAL TECHNIQUE

2.3.1. Vegetative and Reproductive Development

Seeds were pregerminated at the same constant temperatures as in the main temperature treatments, *viz.* 24°, 28° and 33°C. In the case of the greenhouse control, seeds were pregerminated at 28°C. The germinated seeds were sown in earthen pots which were filled with a 1 : 1 clay-leaf mould mixture. To each pot 2 g of a 12–10–18 fertilizer mixture was added. Harvesting took place at the first signs of dying off. Plants were watered daily and surplus water could drain off through the central opening below the pots.

Observations were made on the following subjects and in the following ways:

(a) The main stem lengths were measured every other day from the cotyledons upwards, whereas lateral stems were measured from their place of branching.

(b) Leaf counts were made at weekly intervals.
(c) By means of leaf tracing, weighing and multiplication, thereby using the 10th leaf on the main stem (counted from the cotyledons upwards), a rough measure for the total leaf area was obtained.
(d) Due to the ephemeral behaviour of the groundnut flower, accurate daily flower counts were possible.
(e) The number of gynophores and fruits were counted at harvest time.
(f) Dry weights were obtained by oven drying at 105°C till a constant weight was reached.

2.3.2. Pollen Viability Tests

Plants were grown under the same conditions as described above. Pollen collections were made on different days as well as at different times during the day. The pollen germination media were the following:
(a) A culture medium composed of 100 ml of distilled water, 8 g of commercial cane sugar and 1 g of shredded agar.
(b) The same medium as mentioned under (a) to which was added 50 p.p.m. H_3BO_3.

Microscope slides were spread with a thin coating of the culture medium and the pollen was distributed by brushing the exposed anthers over the medium. The slides were then placed in petri-dishes which were lined with wetted filter paper and covered, thus forming moist germinating and growing chambers. A minimum of 1500 pollen grains were used in determining germination percentages, while pollen was allowed to germinate for at least 90 minutes before counts were made. When germination percentages were determined separately, after the 90 minute germination period, pollen was killed by blowing formaldehyde vapour over the medium.

Pollen tube lengths were determined with the aid of an ocular micrometer. In each case, only the ten longest, i.e. the most viable pollen tubes, were measured. Measurements took place over a period of 150 minutes at 30 minute intervals.

Germination and tube growth determinations were performed with material grown in the thermostat cabinets at temperatures ranging from 21° to 36°C.

2.3.3. Growth and Transport of Carbohydrates

(a) Top-root ratios were determined on plants grown at constant temperatures of 24°, 28° and 33°C. The texture of the glass-sand used in this experiment allowed accurate root weight determinations. In this case Mitscherlich pots, instead of earthen pots, were used. A modified HEWITT (33) nutrient solution was applied twice daily. Drainage took place through plastic tubes which were fitted to the drainage openings of the pots.

TABLE 1 Composition of the nutrient solution in mg per 100 litre

Macro elements		Micro elements	
KNO_3	20.2	(B) as H_3BO_3	210.2
$Ca(NO_3)_2 \cdot 4H_2O$	94.4	(Mn) as $MnSO_4 \cdot H_2O$	169.2
$MgSO_4 \cdot 7H_2O$	36.9	(Cu) as $CuSO_4 \cdot 5H_2O$	25.3
$NaH_2PO_4 \cdot H_2O$	20.3	(Zn) as $ZnSO_4 \cdot 7H_2O$	28.6
		(Mo) as $(NH_4)_2MoO_4$	3.9
		(Fe) as Sequestrene 10.7%	5200.0

(b) For the determination of the sugar content of the leaves, plants were grown in the same way as in 2.3 under greenhouse conditions for 30 days and thereafter they were transferred to the temperature cabinets. In the temperature cabinets different nyctotemperatures were applied before leaf sampling took place. Only the young fully developed leaves were clipped off for leaf analysis. In addition, some plants were kept under continuous illumination for 20 and 32 hours before leaf sampling took place.

The sugar analyses were carried out at the 'Bedrijfslaboratorium voor Grond- en Gewasonderzoek', Oosterbeek.

2.3.4. Additional Remark

Whenever necessary, a more detailed description of the technique will be given separately in the chapters concerned.

III VEGETATIVE AND REPRODUCTIVE DEVELOPMENT AS INFLUENCED BY TEMPERATURE

3.1. INTRODUCTION

The results obtained by various workers, amongst others WENT (103), CHOUARD (18) and BLAAUW (6, 7) make it evident 'that almost as many reaction types of plants to climate exist as plants have been investigated' (103). It remains true, however, that temperature may be regarded as a decisive factor in normal plant growth. The influence of temperature on the various growth processes is quite intricate, mainly due to the fact that no single environmental factor acts independently, but that the effects modify one another. Thus the effect of light intensity and photoperiod may vary with different temperature conditions, and *vice versa.*

WENT (98), working with the tomato plant, pointed out that the optimum night temperature was shifted from 26°C to 8°C, when the light intensity was decreased from full daylight to 200 foot candles (7827.9 erg. $sec.^{-1}cm^{-2}$).

WENT and HULL (101) and HEWITT and CURTIS (34), although contradicting one another, showed that temperature has a significant effect on the translocation of carbohydrates in plants.

The effect of temperature on respiration and photosynthesis must be regarded as being of considerable importance. WENT (102) reported that at lower temperatures the ratio of photosynthesis to respiration is over 10, but that this ratio decreases at high temperatures. According to the latter author, these lower photosynthesis–respiration ratios may indicate why many plants produce a more vigorous growth in the temperate regions than in the tropics.

The total growth of a plant may be considered as the product of growth of its component parts. Thus leaf growth may influence stem growth etc. It is also possible that the optimum temperature for stem growth may not necessarily be the same as that for fertilization.

The investigations of BLAAUW *et al.* (7) with the hyacinth clearly demonstrated the differential response of plants in their various growth stages. He found for example that root growth has an optimum temperature of 28°C, that the temperature for flower and leaf initiation is 35°C, preparation for elongation 13°C and that the optimum for actual stem elongation lies at 20°C.

The effect of diurnal differences between day and night temperatures on plant growth have also been studied and WENT (96, 100, 98) introduced the word 'thermoperiodicity' to explain the response of the plant to cyclic temperature variation. He found that the majority of species tested in greenhouses grow considerably better

when subjected to a daily change in temperature, with the phototemperature higher than the optimal nyctotemperature. The term thermoperiodism was used in a wider sense by CHOUARD (18), who distinguished between a seasonal and a daily thermoperiodicity in the temperature response of plants.

By studying the soybean, PARKER and BORTHWICK (65), on the other hand, could not find any beneficial effect of low night temperatures. More related to our study were the findings of FORTANIER (27), who also studied the groundnut at different temperatures. This writer came to the conclusion that the groundnut does not respond favourably to any day – night fluctuations, but is mainly dependent on the average temperature. A low temperature could be counteracted by a high day temperature and *vice versa*.

From the foregoing it is clear that no generalizations are possible. It is certain, however, that closely related plants usually react to variations in climatic factors in a similar way. Differences between cultivars of the same species are also possible, although such differences are usually quantitative rather than qualitative. Hence three groundnut cultivars of widely different origin were tested in relation to temperature.

3.2. VEGETATIVE DEVELOPMENT

3.2.1. Germination and Development of the Seedling

The groundnut seed is composed of two cotyledons, an upper stem axis and young foliage leaves (epicotyl), and a lower stem axis and primary root (radicle). The whole seed is covered by a thin brittle seed coat which may vary in colour according to the cultivar. All the leaves and aerial parts which the seedling will develop during the first 2 to 3 weeks of growth are already present in the dormant seed.

An investigation of the axillar bud of the seed pointed to the fact that floral initiation even exists in the earliest phases of development in the groundnut plant (70).

Experimental: Groundnut seeds of the Schwarz 21 cultivar were germinated in petri-dishes, which were lined with moist absorbent paper, at temperatures ranging from 24° to 33°C. One hundred seeds were tested at each temperature. Seeds showing no sign of germination after a time lapse of 96 hours were regarded as failures and on that assumption the germination percentages were subsequently determined.

In order to follow the development of the seedling, 50 seeds were allowed to germinate at each of four temperature treatments, (24°, 27°, 30° and 33°C). After germinating for about 48 hours, the first germinated seeds from each treatment were separated from the others in order to determine the elongation of the radicles.

Results and discussions: At 27°, 30° and 33°C the first radicles appeared after 24 to 48 hours, though at 24°C a time lapse of 48 to 72 hours was necessary for their

appearance. Table 2 shows that there was no striking difference in the final germination percentage recorded at temperatures ranging from 24° to 30°C, although 33°C gave

TABLE 2 The effect of four constant temperatures on germination

Germination in hours	Number of seeds germinated			
	24°C	27°C	30°C	33°C
0–24	0	0	0	0
24–48	0	6	6	6
48–72	33	92	89	71
72–96	99	100	95	84
Germination %	99	100	95	84

inferior results than the other temperatures. It appeared that temperature has a greater influence on the further development of the seedling than on the actual germination.

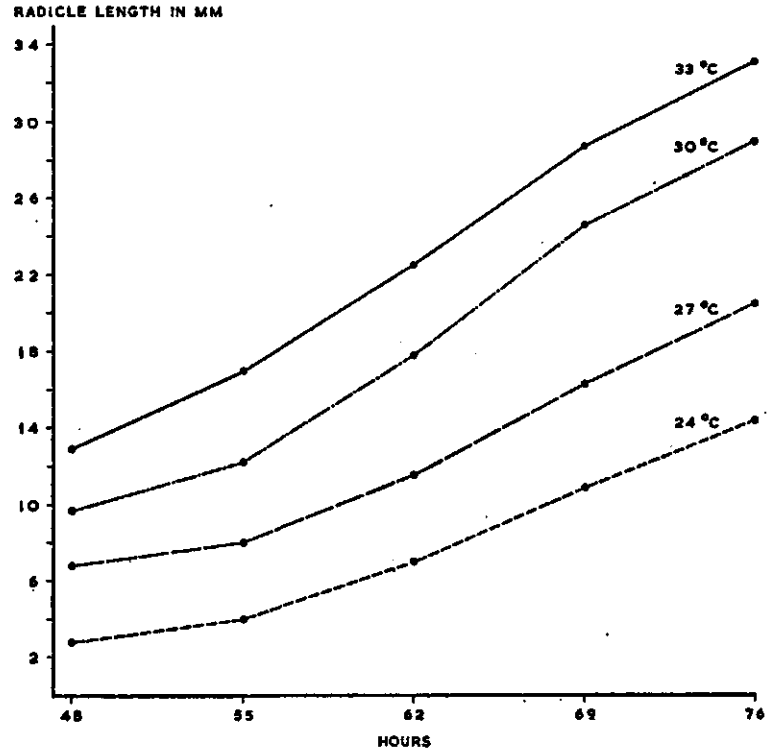

FIG. 1. The elongation of the radicles at 4 different temperatures

The results presented in fig. 1 indicate that the radicle growth was favoured at the high temperatures. It is interesting to note, however, that the rate at which the radicles grew in relation to time was approximately the same for seedlings at 33°, 30°, 27° and 24°C. Expressed in hours, one might say that the seedlings germinated at 33°C gained an advantage of 28 hours over the seedlings at 24°C, 16 hours over the seedlings at 27°C and 7 hours over the seedlings at 30°C. This retarded effect of growth at the lower temperatures is also noticeable when one studies the time necessary for the emergence of the cotyledons above ground, when the seeds were planted at the same depth at different temperatures.

TABLE 3 Number of days from sowing to emergence of the cotyledons as influenced by temperature

Temperature	Cultivars		
	Schwarz 21	Mallorca	Ukraine
24°C	7	7	6
28°C	5	5	5
33°C	5	5	4

The data presented in table 3, show that there was very little difference between 28° and 33°C, where 5 days were required for the emergence of the cotyledons of Schwarz 21 and Mallorca seeds, and 4 days in the case of the Ukraine cultivar. At 24°C the cotyledons of the Schwarz 21 and Mallorca cultivars took 7 days, but those of the Ukraine cultivar 6 days to emerge.

BOLHUIS and DE GROOT (9), who also made observations on the emergence of the cotyledons at different temperatures, obtained the same results. It is interesting to note from their experiments that the greatest difference occurred at 21°C, where 14 and 12 days were required respectively for the Mallorca and Ukraine cultivars before the emergence of the cotyledons. At 18°C they found that, although germination occurred, no subsequent development took place.

It is surprising that the root growth is so much more vigorous than the shoot growth. When the cotyledons appear above the ground, from 5 to 7 days after sowing, the shoot growth only equals more or less the length of the cotyledons. YARBROUGH (107) found an epicotyl – hypocotyl ratio of 1 to 8 at an age of 5 to 5½ days, which clearly demonstrates the discrepancy between the appearance of the young root and that of the shoot.

In general it may be concluded that germination is favoured at temperatures ranging from 24° to 30°C, with a sharp decline in germination at 33°C. Although germination was relatively poor at 33°C in this experiment, the further development of the seedlings was more energetic at 33°C.

BOLHUIS and DE GROOT (9) found that there was little difference in germination at

temperatures ranging from 27° to 33°C. FORTANIER (27), obtained an optimum at 30°C. MONTENEZ (57), on the other hand, found 33°C to be an optimum temperature for germination.

Bearing in mind the results of the other workers, it is obvious that the germination of the groundnut seed is not seriously influenced by temperature, provided the temperature does not exceed 33°C or fall below 24°C.

Our results show, however, that the rate of germination and also the further development of the seedlings are favoured at the higher temperature, in this case 33°C.

3.2.2. Stem Elongations

To obtain more information on the vegetative development of the groundnut plant, the influence of temperature on the main stem-, first lateral stem- and second lateral stem elongation was investigated. The investigation was carried out at 24°, 28° and 33°C except in the case of the Schwarz 21 cultivar, where an additional investigation was carried out in a greenhouse. Neither temperature nor humidity were controllable in the greenhouse, which made direct comparisons impossible. The daily maximum-, minimum- and average temperatures were, however, noted down.

With a view to obtain a better insight into the influence of temperature at various growth stages of the plant, plants grown at 24° and 33°C were transferred at three different developmental stages to 33° and 24°C respectively. The temperature combinations and stages were as follows:

33°C × 24°C (a)	24°C × 33°C (a)
33°C × 24°C (b) and	24°C × 33°C (b)
33°C × 24°C (c)	24°C × 33°C (c)

(a) Transferred at the beginning of flowering.
(b) Transferred 16 days after (a), i.e. at full flowering.
(c) Transferred 34 days after (a), i.e. more or less at the end of flowering.

In reality this scheme was only entirely applicable for the plants grown at 24°C because flowering continued uninterruptedly at 33°C.

As stated before, the main investigation was carried out with the Schwarz 21 cultivar and therefore the other two cultivars were merely investigated at the constant temperatures.

3.2.2.1. *Main Stem*

Results and discussions: In figs. 2, 3 and 4 it is shown that in all three cultivars 33°C proved to be an 'optimum' for main stem elongation, while 24°C may be regarded as a 'minimum' and the elongation at 28°C as a more or less 'normal'. The irregularity of the curve representing the elongation of the main stem under greenhouse conditions

(fig. 2) is easily explainable when the curve is divided into three parts, one for the first 28 days, one for the following 20 days and one for the last 44 days. The average temperatures during those time intervals were 24.1°, 21.7° and 25.4°C. This explains the first part of the curve, where elongation was practically the same as for the

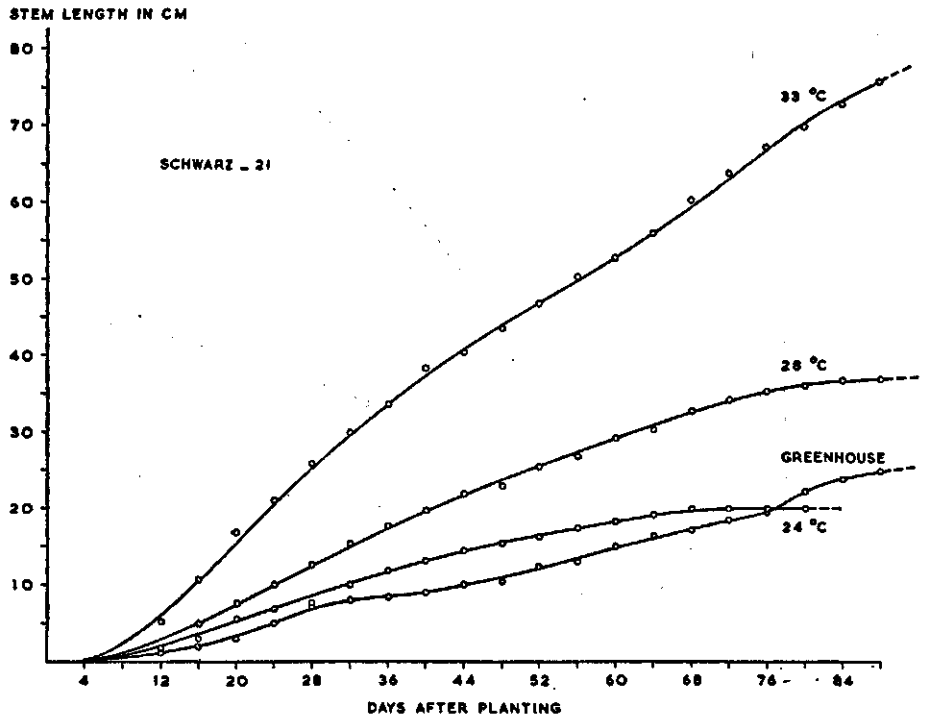

FIG. 2. Main stem elongation of the Schwarz 21 cultivar grown at constant temperatures of 24°, 28°, 33° and in a greenhouse as a control.

plants grown at an even temperature of 24°C. The second part of the curve, which indicates hardly any elongation, coincided with the period of low average temperatures; this period was followed by one with higher average temperatures and consequently a more energetic elongation.

3.2.2.2. *Lateral Stems*

Results and discussions: The elongation of the first lateral stem pairs developed in more or less the same way as did the main stems. Lateral stem elongation was equally favourable at the higher temperatures. In the case of the Ukraine cultivar, however, the first lateral stem pair did not develop in the expected way. Growth was retarded and instead of the first lateral stem pair, the second lateral stem pair developed. The

retarded growth of the first lateral stem pair was counteracted by the growth of the second lateral stem pair.

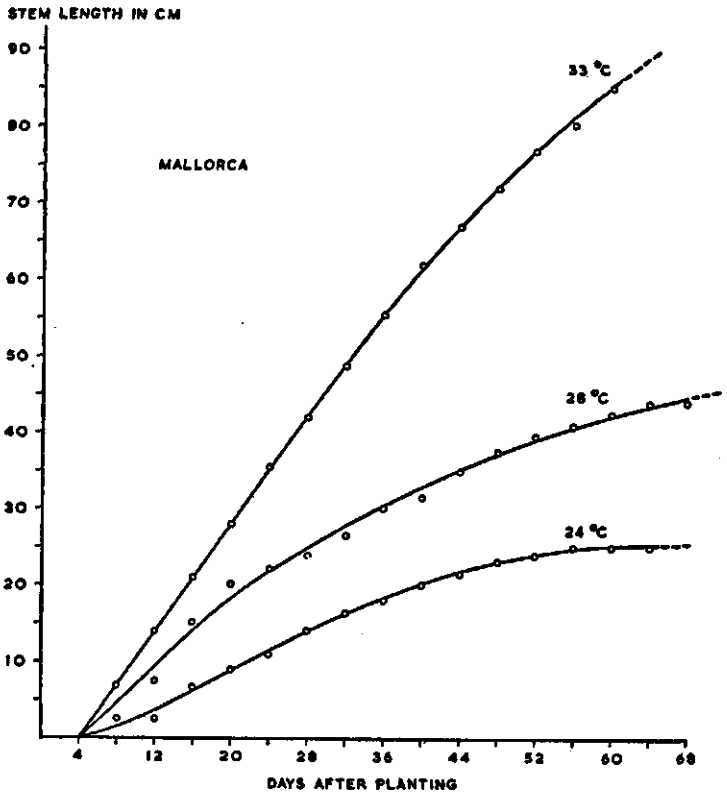

Fig. 3. Main stem elongation of the Mallorca cultivar grown at constant temperatures of 24°, 28° and 33°C.

When studying the data in table 4, it became clear that no striking differences in length between the first and second lateral stem pairs occurred at the lower temperatures. At 33°C, however, the ratio between the first and second lateral stem pairs, 69 days after sowing, was entirely out of proportion *viz.* 102.0 to 29.0 cm. for the Schwarz 21 cultivar, 89.3 to 23.3 for the Mallorca cultivar and 24.4 to 106.0 cm. for the Ukraine cultivar.

Extra lateral stems did not develop to such an extent as to play an important part at any of the above mentioned temperatures. In the case of plants growing in the greenhouse, however, the total average lengths were as follows:

First lateral stem pair = 60.0 cm.
Second lateral stem pair = 53.5 cm.
Third lateral stem pair = 46.5 cm.

TABLE 4 The influence of temperature on the elongation of the first lateral stem pair of three cultivars in cm

Days after sowing	33°C			28°C			24°C		
	Schwarz 21	Mallorca	Ukraine	Schwarz 21	Mallorca	Ukraine	Schwarz 21	Mallorca	Ukraine
21	17.6	13.6	0	4.6	5.2	0	2.8	0	0
33	29.6	32.0	2.2	10.8	18.2	3.4	8.6	10.8	3.0
45	52.0	52.0	14.0	19.8	34.8	10.8	14.2	26.6	8.6
57	73.6	70.8	21.8	26.6	50.0	19.2	18.6	38.0	12.6
69	102.0	89.3	24.4	33.8	60.2	28.0	22.2	43.0	15.2
69*	29.0	23.1	106.0	26.2	47.3	22.1	15.7	37.4	12.0

* Indicating the total length of the second lateral stem pair

As previously stated, the results obtained from plants grown under greenhouse conditions should not be compared with the results obtained from plants grown under artificial conditions; better proportional development of the greenhouse plants and the development of their third lateral stem pair, are most probably due to a higher light intensity and a longer photoperiod.

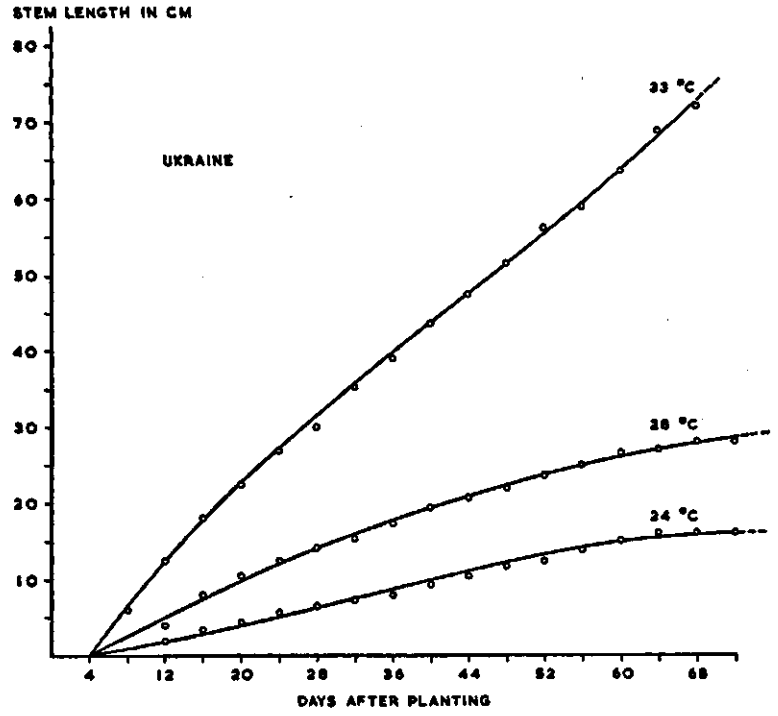

FIG. 4. Main stem elongation of the Ukraine cultivar grown at constant temperatures of 24°, 28° and 33°C.

From the results in table 4, it is evident, however, that the proportional development between the first and second lateral stem pair was hampered at 33°C but not at 24° and 28°C. The total stem length measured at 33°C was considerably greater than at the other temperatures. It is interesting to note that growth did not stop at 33°C; even after 120 days growth continued in the same way as indicated in figs. 2, 3 and 4.

3.2.2.3. *Effect of Temperature Combinations*

Results and discussions: Fig. 5 represents the development of the main stem at the two temperature combinations 33° × 24° and 24° × 33°C. At 33° × 24°C (a), (b) and (c) growth was arrested fairly soon after the plants had been transferred. A normal deduction liable to be made in this respect is that vegetative growth seems to be

retarded at 24°C. This is not true, however, because vegetative development continued for about 68 days when plants were grown at 24°C, whereas growth ceased after about 58 days at 33° × 24°C (c). The foregoing gives thus reason to believe that the duration of vegetative growth is more dependent on the physiological developmental stage of the plants than on their age. At the higher temperature, the physiological development of the plants most probably proceeds at a higher rate than at the lower temperature. When transferred from the high to the low temperature, in this case 33° × 24°C (c), the plants may be considered to be in a full stage of reproduction, but due to the high temperature, few or no fruits developed although flowering was abundant. When plants like the above, physiologically mature to produce fruit, but handicapped to do so at the high temperature, are moved to the lower 24°C, they almost immediately start their reproductive function. After the plants had been transferred, the reproductive activities may proceed at such a high rate, that almost all the energy produced by the plant is directed towards the reproductive organs,

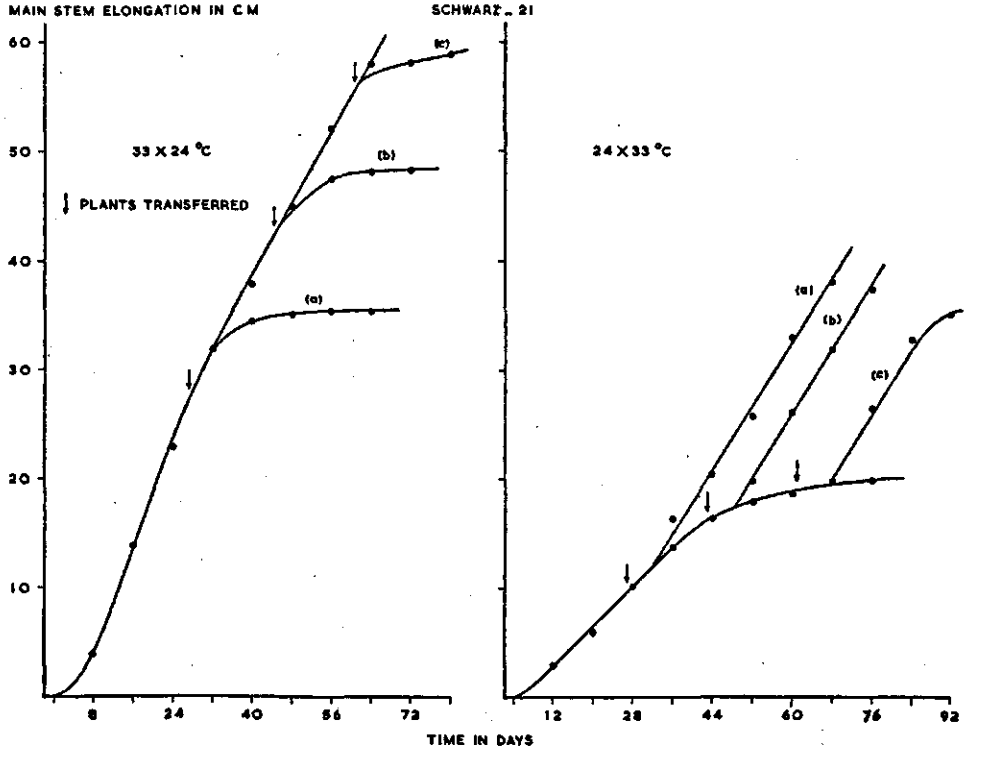

FIG. 5. The influence of a temperature change from 33° to 24° and from 24° to 33°C at three different stages of development on the elongation of the main stems.

(a) Transferred more or less at the beginning of flowering.
(b) Transferred 16 days after (a).
(c) Transferred 34 days after (a).

with the result that vegetative growth slows down and this may be the explanation for divergence of curves (a), (b) and (c) of fig. 5.

From the same figure it is evident that vegetative growth only continued for 48 days at 33° × 24°C (a), although continuation of growth lasted for about 70 days and 86 days at 33° × 24°C (b) and (c).

When plants were grown at 24° × 33°C, the opposite effect became apparent. As far as the vegetative growth of the 24° × 33°C plants was concerned, they reacted as if grown at a constant temperature of 33°C. The elongation of the main stems at 24° × 33°C (a), (b) and (c), almost appear as three parallel lines (fig. 5). It is of importance, however, that at 24° × 33°C (c), where growth had already stopped, i.e. where a balance between vegetative and reproductive growth had already been reached, the plants could again be stimulated to further vegetative growth when placed at the higher temperature. In this case growth continued for 16 days from the date of transfer and then diminished, although the plants remained green without any signs of dying off. The higher temperature, therefore, gave very much the same results as the removal of flowers and fruits had given. BOLHUIS (8) also found that inhibition of fructification is not only a stimulus for the intensity of flowering and the total number of flowers produced, but also resulted in a marked increase in the duration of the flowering period. The removal of fruits resulted in the same continuation of flowering and further development of leaves and stems (27). We believe, that it is not merely the number of mature fruits, but the number of mature fruits in relation to the vegetative mass that will counteract vegetative development. This may explain why plants at 24° × 33°C (c), although almost at the end of their

TABLE 5 The elongation of the first lateral stem pair as plants were moved from 24°C to 33°C and from 33°C to 24°C at three different stages of development. (Average length of 5 plants in cm)

Date	Schwarz 21					
	33°C × 24°C			24°C × 33°C		
	(a)	(b)	(c)	(a)	(b)	(c)
4/11	10.2	12.4	10.0	2.8	3.8	2.8
16/11	25.0	28.4	22.2	9.8	12.0	9.0
28/11	35.4	47.4	36.8	24.0	23.4	17.0
10/12	37.8	63.2	43.2	38.8	38.2	23.8
22/12	39.8	71.8	68.2	58.8	57.6	27.6
3/1	39.8	74.2	90.8	73.4	71.2	40.0
15/1	—	74.2	103.7	83.8	83.4	50.2
29/1	—	—	110.0	92.9	91.7	53.0
10/2	—	—	110.0		very slow	

(a) transferred on 16/11 (More or less at the beginning of flowering)
(b) transferred on 2/12
(c) transferred on 22/12

growing period, were once more stimulated to further vegetative growth. Though no further fruits developed, additional energy was needed to mature the number of fruits required for checking vegetative growth. Instead of diverging the greater part of the produced energy to the reproductive organs of the plant, the high temperature caused the opposite, which may have resulted in an accumulation of energy in some vegetative parts of the plant, and consequently in a further vegetative development for 16 days. The 'required' number of fruits therefore needed an extra 16 days to reach maturity, after which vegetative development was checked 24° × 33°C (c), (fig. 5).

Considering the results obtained by FORTANIER (27) and also those obtained by us, one might conclude that a temperature of 33°C had exactly the same influence as the removal of fruits. A temperature of 33°C must, therefore, either prevent fertilization or retard the development of the embryo.

The development of the lateral stems at different temperatures and temperature combinations followed more or less the same pattern as that of the main stems. The results on the development of the first lateral stem pair are presented in table 5.

3.2.3. Leaf Production

As in the case of stem elongation determinations, a similar investigation was carried out to study development of the leaves. The same temperatures were applied. An additional determination with the Schwarz 21 cultivar was carried out in a greenhouse. The total number of leaves per plant were counted at weekly intervals. The leaf area of the tenth leaf on the main stem, as well as the final number of leaves were determined at harvest time.

TABLE 6 The average number of leaves produced by three cultivars at three constant temperatures

weeks after sowing	Schwarz 21			Mallorca			Ukraine		
	33°C	28°C	24°C	33°C	28°C	24°C	33°C	28°C	24°C
1	4	—	—	4	—	—	4	—	—
2	13	6	4	7	6	4	7	5	4
4	21	19	16	14	20	12	18	14	9
6	43	30	28	33	32	22	32	22	22
7	58	43	36	52	47	34	42	33	30
10	72	50	36	67	60	36	55	45	32
12	80	55	36	74	65	36	64	63	32

Results and discussions: As can be seen from table 6, the development of the leaves was favoured by the high temperature. One week after planting, each of the three cultivars tested had produced 4 leaves, whereas none had been produced at the other two temperature treatments. The results in table 6 further indicate that the higher the

temperature, the higher the number of leaves produced. At 33°C, the Schwarz 21 cultivar produced more leaves than the Mallorca-or Ukraine cultivars, although both cultivars produced more leaves than Schwarz 21 at 28°C. At 24°C the leaf production of all three cultivars was more or less the same, the Ukraine cultivar giving slightly inferior results. The Ukraine cultivar produced more or less the same number of leaves at both 28° and 33°C. This unexpected result was most probably due to the fact that the first lateral stem pair did not develop in the expected way. (See page 11 and 12).

Leaf development in the groundnut proceeds very rapidly. Taking the leaf development from the first to the tenth week after planting, the Schwarz 21 cultivar produced a new leaf every 0.97 days at 33°C, every 1.40 days at 28°C and every 1.94 days at 24°C. A new leaf was produced every 1.05, 1.17 and 1.94 days in the case of the Mallorca cultivar while the Ukraine cultivar needed 1.27, 1.55 and 2.19 days at 33°, 28° and 24°C respectively.

Taken at the tenth week, the Q_{10} of leaf formation was 2.3 for the Schwarz 21 cultivar, 2.1 for the Mallorca cultivar and 1.9 for the Ukraine cultivar.

When studying the results obtained in figs. 2, 3 and 4, where stem elongation was plotted out, it becomes evident that stem growth and number of leaves are fairly well correlated: more leaves and longer stems occur together at the high than at the low temperatures.

The total leaf area is most probably of greater importance for photosynthesis than the total number of leaves produced. In order to obtain an indication of the total leaf area per plant, the area of the tenth leaf was carefully measured and then multiplied by the number of leaves produced. As no experiments were carried out to determine which leaf would give the best estimate for leaf area, these figures may only be considered as a rough approximation of the total leaf area.

The results presented in table 7, indicate that smaller leaves were formed at the high than at the low temperature. Of the three cultivars tested, the Ukraine cultivar was least affected by temperature, resulting in an average tenth leaf area of 30.65 cm^2 at 33°C and 39.89 cm^2 at 24°C. The greatest response to temperature was found with the Mallorca cultivar, where the area of the tenth leaf on the main stem at 24°C was over three times the size of that formed at 33°C, with the result that a greater total leaf area was obtained at 24°C than at 33°C. This, however, only holds true for the Mallorca cultivar, because the greater number of leaves produced at 33°C, counteracted the smaller leaf size, so that the total leaf area of the Schwarz 21 and the Ukraine cultivars was considerably greater at 33° than at 24°C.

3.2.4. Dry Weights

Results and discussions: The dry weights of the aerial parts of the plant, obtained at different temperatures and temperature combinations, are shown in tables 8a and 8b.

TABLE 7 The influence of temperature on the total leaf area (cm^2) of three cultivars as obtained from the 10th leaf on the main stem at harvest time

	Schwarz 21			Mallorca			Ukraine		
	33°C	28°C	24°C	33°C	28°C	24°C	33°C	28°C	24°C
Total number of leaves	80	55	36	74	65	36	64	63	32
Area of 10th leaf	39.30	61.88	56.11	23.59	66.98	75.64	30.65	44.06	39.89
Total leaf area p. plant	3144.0	3403.4	2019.9	1745.7	4353.7	2723.0	1961.6	2775.8	1276.5

TABLE 8a The influence of temperature on the average dry weights of the aerial parts of three cultivars in g

Schwarz 21			Mallorca			Ukraine		
33°C	28°C	24°C	33°C	28°C	24°C	33°C	28°C	24°C
15.1	8.4	4.8	8.9	9.8	5.9	8.7	5.4	3.1

TABLE 8b The influence of a temperature combination of 24° × 33° and 33° × 24°C on the average dry weight of the aerial parts of the Schwarz 21 cultivar in g

33° × 24°C			24° × 33°C		
(a)	(b)	(c)	(a)	(b)	(c)
5.7	10.1	15.2	14.9	11.4	7.3

(a) Transference at beginning of flowering
(b) Transference 16 days after (a)
(c) Transference 34 days after (a)

When comparing these results with the results obtained for stem elongation, it is apparent that there is a fairly close correlation between stem length and dry weight. The higher the growth temperature, the longer the stems and the higher the dry weights. This was true in all cases, except one. For the Mallorca cultivar, 33°C was definitely too high for normal vegetative development, and its dry weights were less at 33° than at 28°C.

The results obtained at the temperature combinations show that the longer the period at 33°C, the higher the dry weights.

3.3. REPRODUCTIVE DEVELOPMENT

3.3.1. Flower Development

Although an investigation of the axillar bud of the groundnut cotyledons pointed to the fact that floral initiation even exists in the earliest phases of its development (70), flower initiation may be regarded as the transition from the vegetative to the reproductive phase in the life of the plant.

With a view to our field of study, only a few structural characteristics of the groundnut flower are of importance and need therefore some attention. For a more detailed description of the morphology of the groundnut we refer to (80, 30 and 44).

The groundnut flower possesses a long tubular hypanthium which usually gives

the wrong impression that the flowers are pedicellate. Actually, the base of the hypanthium is inserted at the end of a simple flowering branch, directly subtended by its cataphyllar bract (30). The ovary is thus far removed from the stigma and consequently the style has more or less the same length as the hypanthium. The longer the hypanthium, the farther the pollen tubes have to travel down the style in order to effect fertilization. This characteristic in combination with the fact that the groundnut flowers are ephemeral, may be a decisive factor in the fertilization process.

With the preceding in mind, an investigation was carried out to examine the influence of temperature on the length of the hypanthia as well as on the longevity of the flowers.

Experimental: The length of the hypanthia as well as the longevity of the flowers were studied at the same temperature and temperature combinations as described in the stem elongation experiments, *viz.*, 24°, 28°, 33°, 24° × 33° (a), (b), (c), 33° × 24°C (a), (b) and (c).

Results and discussions: The hypanthium structure of all three cultivars tested was affected by temperature in the same way. The results in table 9 indicate that the higher the temperature, the longer the hypanthia. Although the differences in hypanthium lengths were small at 24° and 28°C, the hypanthia were much shorter than those at 33°C. In most cases the hypanthia were twice as long at 33°C as at the other two constant temperatures. It is of interest to note that about 80% of the total hypanthium elongation occurs during the night. During the day bud elongation proceeds at a very slow rate but during the night elongation accelerates.

The longevity of the flowers is undoubtedly of the same importance as the length of the hypanthia. The longer the flowers stay fresh, the more time will be available for pollen tube growth. An inverse correlation between hypanthium length and longevity of the flowers was found. The higher the temperature, the longer the hypanthia and the sooner the flowers wilt. At 24°C the lifespan of the flowers was found to be about 12 hours, at 28°C 11 hours and at 33°C 9 hours. Corresponding results were obtained where the temperature combination effect on the hypanthia was investigated. Two days after the transfer to the second temperature had taken place, *viz.* 24° × 33°C and 33° × 24°C, the flower characteristics were the same as those when the plants were grown continuously at the second temperature. It may be considered that it is possible to alter the character of the flower, *i.e.* the lengths of the hypanthia as well as the longevity of flowers, by changing the temperature during a minimum of 48 hours.

3.3.2. Intensity of Flowering

Experimental: Since the groundnut flower is ephemeral and wilts within a couple of hours, an accurate account of flower production is possible. The influence of tempera-

TABLE 9 Hypanthia lengths (mm) of three cultivars as influenced by three constant temperatures

	Schwarz 21			Mallorca			Ukraine		
	33°C	28°C	24°C	33°C	28°C	24°C	33°C	28°C	24°C
Average of 20 longest hypanthia	66.3	39.7	31.0	63.4	46.4	32.0	63.1	42.4	33.7
Average of 20 shortest hypanthia	42.5	17.8	17.5	40.0	23.9	13.7	33.7	21.0	16.9
Mean length	53.5 ± 7.2*	28.1 ± 7.1	25.5 ± 3.5	53.7 ± 6.3	36.5 ± 6.3	25.0 ± 5.3	48.3 ± 8.3	33.3 ± 6.3	26.7 ± 4.5

* Standard error of the mean

ture on the intensity of flowering as well as the number of days between date of sowing and commencement of flowering was investigated. The same temperature and temperature combinations were applied as described before.

Results: The results as to the appearance of the first flowers, *i.e.* the number of days between date of sowing and the commencement of flowering, are presented in table 10.

Except for the Mallorca cultivar, the number of days from sowing till flowering, decreases as the temperature is raised. A temperature of 33°C was obviously harmful to flowering in the Mallorca cultivar. Flower initiation, however, took place although the flowers did not develop in the expected way. Flower initiation was thus not so much hampered by the high temperature as was the actual development of the flowers.

TABLE 10 The number of days from sowing to flowering of three cultivars as influenced by temperature

	Schwarz 21	Ukraine	Mallorca
24°C	35	34	32
28°C	28	26	25
33°C	25	25	43

The observations made on the intensity of flowering are presented in figs. 6 and 7. The curves are cumulative, expressing the daily average number of flowers per plant.

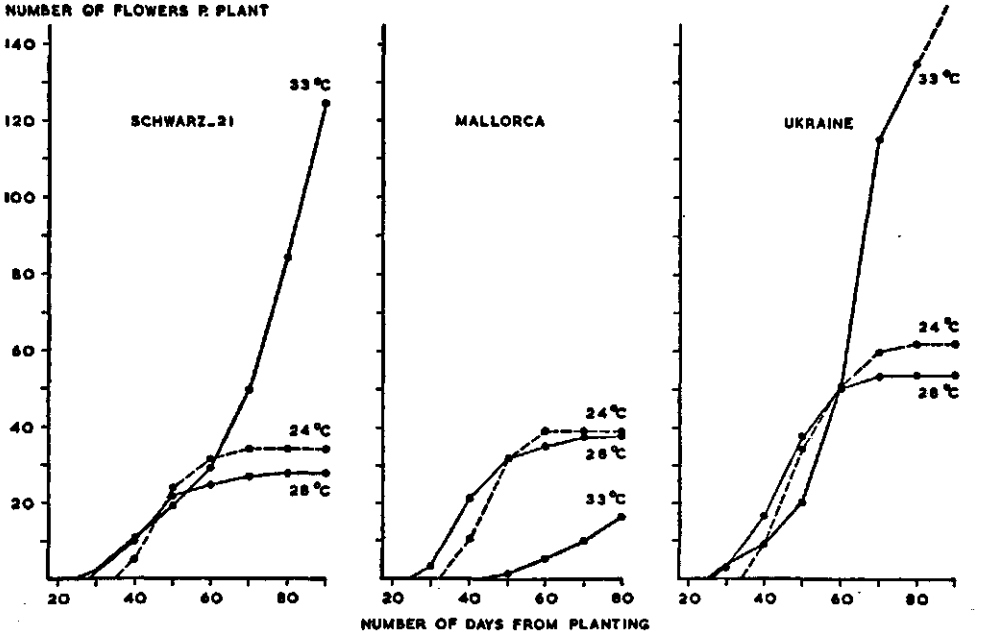

FIG. 6. Cumulative curves representing the number of flowers per plant per day, when plants of the Schwarz 21, Mallorca and Ukraine cultivars were grown at three different constant temperatures.

Flowering is sometimes rather slow during the first 5 days, whereafter it accelerates to stop again rather quickly. In the case of plants grown at 33°C, however, flowering continues at a high rate without any sign of stopping. As already stated, the high temperature has a depressing influence on flowering in the Mallorca cultivar. Although the intensity of flowering was low and the appearance of the first flowers late, flowering did not stop during the course of the experiment. The Mallorca cultivar therefore exhibited the same flowering characteristics as the other two cultivars at 33°C. The flowering curves of the Schwarz 21- and Ukraine cultivars were almost identical. At 33°C flowering went on uninterruptedly, while slightly more flowers were produced at 24° than at 28°C.

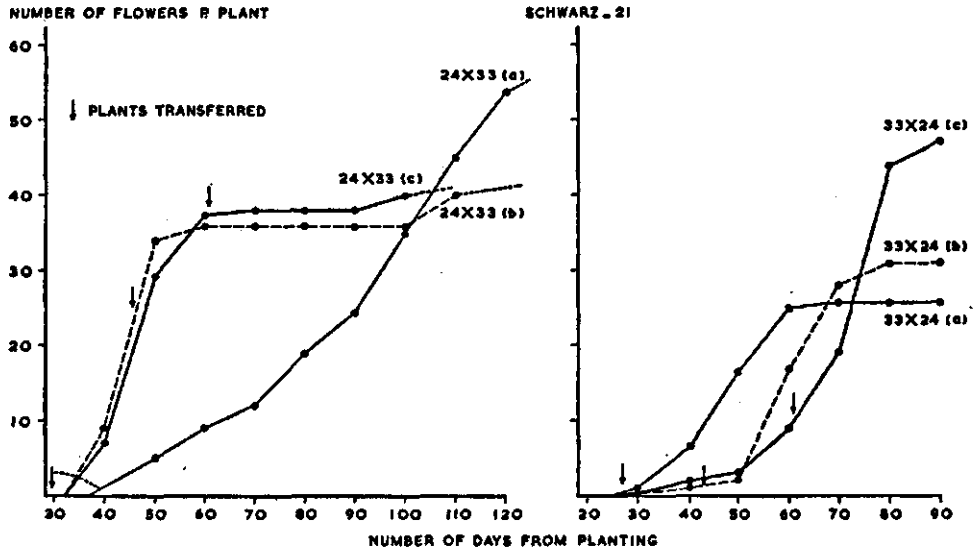

Fig. 7. Cumulative curves representing the number of flowers per plant per day, when plants which were grown at 24° and 33°C were respectively transferred to 33° and 24°C at three stages of development.

(a) Transferred more or less at the beginning of flowering.
(b) Transferred 16 days after (a).
(c) Transferred 34 days after (a).

After considering the results shown in fig. 7 with plants grown at 24° and then transferred to 33°C, it is clear that in the case of 24° × 33°C (a), the plants reacted as if grown at a constant temperature of 33°C, as far as their flowering habit was concerned. (Compare with curves representing flowering at a constant temperature of 33°C fig. 6).

When plants were transferred at a late stage, even at the end of flowering, i.e. 24° × 33°C (c), the plants could once again be stimulated to further flowering, although this occurred at a low rate.

Fig. 7 also presents the data on plants grown at a constant temperature of 33°C and then shifted to 24°C at three different stages. When transferred at the beginning of flowering, *i.e.* 33° × 24°C (a), flowering was the same as when the plants were grown at a constant temperature of 24°C. The same was found at 33° × 24°C (b) and (c) when plants were transferred at full flowering. After the plants had been transferred, flowering usually stopped after about 30 days, whereas flowering continued when plants were kept at a constant temperature of 33°C.

Discussion: The production of flowers in the groundnut was investigated by various workers including BOUFFIL (12), SMITH (80), FORTANIER (27), BOLHUIS and DE GROOT (9), just to mention a few. Only in the last two studies the plants were under controlled environmental conditions. BOUFFIL (12), working in Senegal, kept record of temperature, rainfall, hours of sunlight *etc.*, and came to the conclusion that the pattern of flowering is not influenced by meteorological conditions. The same worker concluded further that, although flowering may be reduced in some instances, the general pattern of flowering remains the same. This, in our opinion, is not quite correct although the pattern of flowering was more or less the same for all three cultivars tested at 24° and 28°C. At 33°C, as may be seen from fig. 6, the pattern of flowering changes completely and plants can produce flowers 'indefinitely'.

In his summary FORTANIER (27) stated that the opening of the flower buds as well as the total number of flowers formed, are highly dependent on the external conditions. Our results indicate, however, that the flowering pattern did not change in the temperature range from 24° to 28°, but at 33°C it changed to such an extent that flowering never stopped throughout the experiment. At 21°C, BOLHUIS and DE GROOT (9), working with the same cultivars as the present author, could only obtain flowering 81 days after sowing with the Mallorca cultivar, whereas no flowers were produced by the other two cultivars.

The foregoing suggests, therefore, that a temperature of 33°C as well as temperatures lower than 24°C are liable to change the flowering pattern considerably.

The results further indicate that temperature during the vegetative phase has little or no influence on the later development of flowering, but that flowering is mainly dependent on the reigning temperature during the generative phase. It was even possible to stimulate plants to further flowering at the end of their normal flowering period by transferring them from 24° to 33°C (c). After no flowers had developed for 20 days at 24° × 33°C (c) and for about 30 days at 24° × 33°C (b), the plants again started to produce flowers, although at a low rate, as can be seen from fig. 7.

3.3.3. Fertilization and Fruit Development

The success of fertilization is dependent on various flower and plant characteristics. It is well known to farmers and plant breeders that various environmental factors play an important role in fertilization.

Directly after fertilization the growth of the peg starts. The peg as defined by SMITH (80) is 'the young fruit during the stalk-like phase of development which intervenes between syngamy and fruit enlargement'. This stalk-like organ is commonly known as the gynophore, but in reality, according to SMITH (80), is the ovary itself, which is elongated due to growth of an intercalary meristem in its base. The development of the gynophore and the maintenance of vascular continuity in the intercalary meristem was investigated by JACOBS (40).

Due to the fact that most plant physiologists and morphologists know the stalk on which the ovary rests as a gynophore, this name or term, although wrong, is also used by the author.

The gynophore is positively geotropic (77, 4) and for normal development of the fruit it must penetrate the soil although in exceptional circumstances, ovule enlargement can take place in the aerial gynophores (30). SHIBUYA (77, 78) and also YASUDA (108) made a study of the morphology and physiology of fructification as well as a physiological analysis of the mechanism of fruit development in the groundnut, in which they describe conditions essential for the development of the fruit.

Since the success of fertilization is vitally important, some understanding and knowledge of the fertilization process is an absolute necessity.

The causes of unfruitfulness may be considered as being of two general types:

(a) Those which are inherent to the plant such as genetic inhibition etc.

(b) Those which are external to the plant such as environmental factors.

With temperature as the variable environmental factor, different experiments were carried out with a view to study fertilization and fruit development. Although not entirely correct (see p. 31 and 32) fertilization coefficients were obtained by counting the number of flowers producing gynophores.

3.3.3.1. *Temperature*

Experimental: The influence of three constant temperatures, *viz.* 24°, 28° and 33°C on fertilization was studied using the three cultivars already mentioned. Where the effect of the temperature combinations 24° × 33° and 33° × 24°C was investigated, the Schwarz 21 cultivar was used. Plants were transferred from one temperature to the other at the three stages already described on page 10.

Results: The results summarised in table 11 indicate that a temperature of 33°C was detrimental to fertilization, the fertilization percentages being only 3.6 for the Schwarz 21 cultivar, 1.6 for the Ukraine cultivar and nil for the Mallorca cultivar. At the other two temperatures the fertilization percentages were much higher, the results being slightly better at 28° than at 24°C.

When studying the results of the different cultivars separately, it is interesting to note that the higher the number of flowers, the lower the fertility coefficient. The Mallorca cultivar, producing only 16.7 flowers per plant at 33°C was, however, an

TABLE 11 The influence of three constant temperatures on the fertility coefficients of three cultivars (7 plants per treatment – number of flowers etc. given as average p. plant)

	Schwarz 21			Mallorca			Ukraine		
	33°C	28°C	24°C	33°C	28°C	24°C	33°C	28°C	24°C
Number of flowers	94.0	28.1	34.5	16.7	37.5	39.0	188.4	53.5	61.5
Immature pods	1.8	3.6	7.3	0	19.0	19.0	2.9	23.3	20.8
Mature pods	1.6	8.5	5.0	0	7.5	5.3	0	10.6	7.5
Dry weight of seeds + pods (g)	7.8	42.1	17.5	0	55.4	20.5	0	27.7	14.4
Total number of gynophores	3.4	12.1	12.3	0	26.5	24.3	2.9	33.9	28.3
Fertilization %	3.6	43.2	35.5	0	70.7	62.3	1.6	63.5	45.9

exception, as already previously described. In the case of the Mallorca cultivar the development of the flowers was hampered, whereas flower bud initiation occurred in the normal way.

TABLE 12 The influence of a temperature change at three different stages during the development of the plant on the fertility coefficient of the Schwarz 21 cultivar (5 plants per treatment – number of flowers etc. given as average p. plant)

	33° × 24°C			24° × 33°C		
	(a)	(b)	(c)	(a)	(b)	(c)
Number of flowers	25.2	31.2	47.4	79.9	42.6	37.0
Immature pods	8.8	9.6	18.2	2.8	2.8	5.2
Mature pods	5.4	7.0	6.0	1.8	5.4	5.2
Dry weight of seeds + pods (g)	17.7	17.4	11.6	5.5	18.8	21.1
Total number of gynophores	14.2	16.6	24.7	4.6	8.2	10.4
Fertilization %	55.0	53.0	51.0	5.8	19.2	28.1

(a) Transference at beginning of flowering
(b) Transference 16 days after (a)
(c) Transference 34 days after (a)

The data in table 12 show that the earlier the switch over from 24° to 33°C, the lower the fertility coefficient was. At 24° × 33°C (a) the fertility coefficient was 5.8 while at 24° × 33°C (b) and (c) it was 10.2 and 28.1 respectively. At the other temperature combinations, *viz.* 33° × 24°C (a), (b) and (c), the fertility coefficients were strikingly higher than at 24° × 33°C. Although the fertility coefficients were more or less the same at 33° × 24°C, slightly higher values were obtained at (a) than at (b) which in its turn yielded a slightly higher value than (c).

3.3.3.2. *Borium Sprays and Flower Removal*

Experimental: The first part of the experiment was conducted at 24° and 33°C as well as in a greenhouse. During flowering, weekly sprays of H_3BO_3 (200 p.p.m.) plus a wetting agent were applied to the leaves and flowers.

This experiment was mainly designed to establish whether borium sprays could cause the pollen to become more active, eventually resulting in a higher fertility coefficient. (See page 56 and 57).

At the beginning of flowering, the plants were divided into two equal groups, one group being treated with H_3BO_3, while the other was left untreated to serve as the control.

In the second part of the experiment, plants were grown under the same conditions as described above. During the first three weeks of flowering, flowers were removed daily. The flowers were clipped before fertilization could have taken place. After the flower removal treatment, the plants were left to flower and to develop in the normal way. The same control was kept as in the previous part of the experiment.

Results: From the results summarized in table 13 it is evident that the application of H_3BO_3 had no effect on the fertility coefficient. Where H_3BO_3 was applied at 24°C, the fertility coefficient was even lower than in the control. This small difference can hardly be important, so that it would seem justified to conclude that an application of H_3BO_3 had little or no influence on fertilization.

When the flowers were removed during the first three weeks of flowering, the fertilization percentages were even higher at 24°C than in the control, although less gynophores and fruits were produced. These higher fertilization percentages are most probably due to the smaller number of flowers produced after flower removal had taken place.

The results in table 13 make it clear that the removal of flowers during the first three weeks of flowering as well as the application of H_3BO_3 to the leaves and flowers, had no effect on fertilization and very little effect on the production of pods.

3.3.3.3. *Discussion*

The results indicate that much more gynophores were produced at 28° and 24° than at 33°C. Treatment at 33°C never resulted in a fertility percentage exceeding 3.6.

A higher temperature during the vegetative phase had little or no influence on the reproductive development, nor could a low temperature during the vegetative phase counteract the detrimental influence of a high temperature during the reproductive phase.

For all three cultivars tested, the highest dry weight of seed and pods was obtained at 28°C. Although the fertility coefficient at 24° and 28°C did not differ much, the weights of seed and pods were considerably higher at 28° than at 24°C. At 33° the failure of fertilization prevented fruit development. Taking into consideration that

TABLE 13 The effect of H_3BO_3 sprays (200 p.p.m.) during flowering and the influence of flower removal during the first three weeks of blooming on the fertility coefficient at three different temperatures (totals of 5 plants)

Treatments	Flowers removed during first 3 weeks			Weekly H_3BO_3 sprays on leaves+flowers			Control		
Temperatures °C	33°	24°	greenhouse	33°	24°	greenhouse	33°	24°	greenhouse
Number of flowers	513	123	465	608	155	424	635	165	452
Number of gynophores + fruits	8	63	125	10	61	113	11	70	120
% flowers fertilized	1.6	51.2	26.9	1.6	39.3	26.7	1.7	42.4	26.5

the vegetative mass was much higher at 28° than at 24°C, the difference in fruit weight is easily explainable. It is necessary to state, however, that the size of the pots was relatively small and that some gynophores could not enter the soil, due to the fact that they developed at the higher nodes. This did not really alter the results, for some gynophores, although in the soil, never came to maturity. The number of gynophores that will reach maturity is mostly dependent on the photosynthetic ability of the plant. Under identical conditions the plant with the greatest vegetative mass will mature the greatest number of fruits. It is true, however, that one specific temperature may be favourable for vegetative development but not for reproductive development.

While vegetative development seems to be limited at 24°C, fertilization or perhaps the development of the gynophore seems to be limited at 33°C.

With three main temperature treatments, *viz.* 27°, 29° and 32°C, applied for different durations and at different stages of development, LAMBERT and LINCK (50) working with pea plants, concluded that a temperature of 32°C reduced yields more than one of 29°C, a temperature of 29°C, on the other hand, more than one of 27°C. In other words, the higher the temperature, the lower the yields. BOSWELL (11), also working with the garden pea, reported that the closest inverse relation between high temperature and yield was the period from blooming to harvest, although he could not establish the period during which high temperatures were most detrimental. LAMBERT and LINCK (50) believed that the high temperature may have reduced pea yields, by causing an increase in respiration, decreasing the concentration of nutrients needed for ovule development, or by reducing translocation of materials into the pods and peas.

The results summarized in tables 11 and 13, make it once again clear that many more flowers than fruits were produced. It was in connection with this occurrence that JODIDI (41) stated that: 'The average yields and efficiency of crop production in general are so far below the apparent possibilities that there is a pressing need for exhaustive fundamental studies of the plant in all stages of development and from numerous points of view'.

It was anticipated that boron sprays would increase the activity of pollen from plants grown at 33°C and thus result in a higher fertility coefficient. The influence of boron sprays on the groundnut was previously investigated by HARRIS and GILMAN (31). These workers came to the conclusion that a boron application not only increases yield but also improves the quality of the nuts. In our experiments, however, boron had no such beneficial effects. More in agreement with our results were the findings of COLLINS and MORRIS (19) and KILLINGER, *et al.* (46). These workers found that where boron had been applied for years in field experimentation, no effect could be obtained either on yield or on quality. ALEXANDER (2) also reported that four weekly foliar sprays of various nutrients at the flowering stage had no significant effect on the yield or quality of the groundnuts.

In a study on the influence of temperature on fertilization and fruit development of the groundnut, it is important that the following should be kept in mind:

(a) That the groundnut flower is ephemeral and therefore wilts within a couple of hours.

(b) That the length of the hypanthia and consequently also of the styles is strongly influenced by temperature. The higher the temperature, the longer the styles, and therefore most probably effective fertilization will take a longer time.

(c) That some of the pollen tubes may reach the ovary without fertilization taking place.

(d) That some ovaries may remain dormant although fertilization has taken place.

3.4. Dormancy of ovaries

Experimental: An experiment was conducted to ascertain whether some ovaries at 33°C may remain dormant in their inflorescences after fertilization has taken place.

Plants of the Ukraine cultivar were grown from 14/10/61 till 22/12/61 at 33°C, a period during which flowering was abundant. On 23/12/61 half the plants were moved to 24°C where they were kept till 26/1/62 when harvesting took place. With a view to prevent fertilization at 24°C, early each morning all newly developed flowers were removed. In order to give all plants an equal chance, the flowers of the control plants were also removed every day.

Results and discussions: The results compiled in table 14 indicate that at least 5% of the flowers were fertilized at 33°C but were prevented to develop any further owing to the high temperature. This is clearly demonstrated by the control plants which were kept uninterruptedly at 33°C. In this case only plant number 8 developed any gynophores. These results suggest, therefore, that although fertilization may take place to some degree at 33°C, further development of the ovary is hampered by the high

Table 14 The influence of temperature on the occurrence of ovary dormancy when plants were allowed to flower at 33°C and then transferred to 24°C for further development. (All newly developed flowers being removed after transference)

Treatment	33° × 24°C				Control 33°C		
Plant no.	Number of flowers	Number of gynophores	Fer. %	Plant no.	Number of flowers	Number of gynophores	Fer. %
1	160	13	8.1	6	159	0	0
2	162	7	4.3	7	168	0	0
3	154	8	5.2	8	151	2	1.3
4	163	9	5.5	9	154	0	0
5	158	7	5.1	10	163	0	0

temperature. The ovary may thus stay dormant in its inflorescence when conditions are unfavourable for its further development.

SMITH (81) has also pointed out that certain ovaries can remain dormant while others may develop in the normal way. 'This demonstrated that following syngamy certain ovaries remained dormant in their inflorescences for several weeks without losing their ability to develop pegs and eventually to produce mature pods and seeds'.

All these results demonstrate that 33°C is either directly or indirectly responsible for creating an unfavourable condition for ovary development.

3.5. THE RELATION BETWEEN VEGETATIVE AND REPRODUCTIVE DEVELOPMENT

In those cases where the stem elongation at different temperature combinations, 24° × 33° (a, b, c) and 33° × 24°C (a, b and c) was investigated, the results revealed that as long as the plant can be stimulated to vegetative growth, it can also be stimulated to further flowering. (Compare flowering curves fig. 7, with stem elongation curves, fig. 5). This may partly be in accord with the statement made by MOORE (58), that in the case of the groundnut, vegetation and reproduction are not opposing tendencies but are complements in the normal course of plant development. This statement may, however, be considered as partly true as its implication only concerns flowering and not further reproductive growth such as fertilization, fructification etc. When gynophores are removed, or as in our case, when plants are grown at 33°C, *i.e.* where normal fruit development is impossible, flowering as well as vegetative growth are stimulated. It is true to say that vegetative growth and flowering are not opposing tendencies, but are complements in their development. Vegetative growth and flowering combined, however, constitute an opposing tendency to fruit development. Thus, fruit growth and vegetative growth are opposing tendencies.

3.6. SUMMARY

The results from germination up till the maturation of the seeds and pods irrefutably showed that the temperature had a decisive influence on the growth and development of the groundnut plant.

The growth responses at the different temperature treatments indicated that 24°C was too low and 33°C, on the other hand, too high for normal growth and development. The three cultivars studied reacted in more or less the same way to the temperature treatments. The Schwarz 21 cultivar was found to show the greatest temperature tolerance, while the Ukraine cultivar showed a somewhat greater tolerance to high temperature than the Mallorca cultivar.

A constant temperature of 33°C usually leads to an increase in vegetative growth

connected with a low pod production, whereas at 24°C vegetative growth was retarded but pod production was considerably higher than at 33°C.

If the growth and development of the plants at 28°C are taken as normal, the growth response of plants at 24° and 33°C may be tabulated in a relative way as in table 15.

If growth and development were:

(1) Better than when plants were grown at 28°C, they were considered as being *excellent.* (××××).

(2) The same as at 28°C, they were considered as being *good* (×××).

(3) Distinctly less than in (2), they were considered as being *fair* (××).

(4) Zero or almost zero, they were considered as being *poor* (×).

Germination percentages above 90 were considered as being *excellent* whilst percentages below 90 were classified as *good.*

TABLE 15 The relative influence of three constant temperatures on the different growth processes of the groundnut plant

	33°C	28°C	24°C
Germination	×××	××××	××××
Stem elongation	××××	×××	××
Leaf development	××××	×××	××
Flowering	××××	×××	××
Fertilization	×	×××	×××
Production of gynophores	×	×××	×××
Production of seeds and pods	×	×××	××

× Poor
×× Fair
××× Good
×××× Excellent

At 24°C the retarded vegetative growth was undoubtedly the limiting factor in the development of the plant. At 33°C the vegetative growth and further development of the plant proceeded in a more or less normal way up to the fertilization process, after which no or few gynophores developed. In some previous experiments we also determined that at high temperatures some gynophores failed to develop because a number of ovaries remained dormant in their inflorescences. This phenomenon is undoubtedly not the only limiting factor in the development of the gynophores and fruits.

In the following chapters some of the experiments conducted in relation to this problem of unfruitfulness at high temperatures will be described.

IV THE DEVELOPMENT OF POLLEN AS INFLUENCED BY TEMPERATURE

4.1. INTRODUCTION

According to the literature AMICI (see, 42), examining the papillate stigma of *Portulaca oleracea*, saw a 'hair' growing out of a pollen grain. He later described the pollen tube, which elongated bit by bit, disappearing in the stigmatic tissue of the style. The findings of AMICI may thus be regarded as the discovery of the pollen tube. About 10 years later VON MOHL (see 42) also made some observations on pollen tube growth and stated that germination and pollen tube elongation may take place if the humidity of the air is high enough.

Since 1924, when BRINK (13, 14, 15, 16, 17) made elaborate studies on pollen physiology and the requirements for pollen tube growth, a number of publications, unfortunately not always in agreement with one another, occasionally appeared.

4.1.1. The germination medium

4.1.1.1. *Water*

VAN TIEGHEM (89), LIDFORS (52, 53) and JOST (43) stated that *Dactylis* and *Hippeastrum* pollen grains germinated readily in water. PATON (66) even claimed to have obtained optimal germination with pollen of a *Lilium* species in ordinary tap water.

In 1951, EHLERS (24) stated that pollen tubes of 14 angiosperm species were found to develop to a length sufficient for fertilization in artificial media containing only distilled water and traces of boric acid. From his observation on pollen tube growth in artificial media as well as in *Tradescantia* and *Amaryllis* styles he concluded that it is most probable that the reserve materials of the pollen grain are the only source of nutrients for tube development. On the other hand, the majority of workers such as SCHWARZENBACH (75), VASIL (91, 92), and the present author, could not obtain any or in some cases only very slight germination or pollen tube growth in water. Though germination is possible in water, the germination as well as the pollen tube growth may generally be regarded as poor. When germination is possible in water, however, the pollen grain should at least contain sufficient reserve food to germinate and in some cases even to produce fairly long tubes.

It is well known that the pollen grain stores some reserve food in the form of starch, sucrose, glucose or fructose (39). IWANAMI (39) points out that at the time of germination, most of the starch in the pollen grains is converted into sugars. By analysing

the pollen of apples, pears and species of *Prunus*, OSTAPENKO (64) concluded that although the composition of the pollen varies considerably, they were generally characterised by a low pH and a high activity of oxidising and reducing sugars. The grains also contained some polyphenols, sulphydryl groups, carbohydrates and in some cases a little fat.

From the foregoing it becomes obvious that in some exceptional cases the pollen grains contain sufficient reserve nutrients to produce pollen tubes long enough to reach the ovules. In most other cases, however, it is doubtful whether adequate food reserves are present in the pollen grains to reach a length enabling fertilization.

4.1.1.2. *Sugar*

It was found that when sugar was added to the germination medium, with or without agar or gelatine, most pollen grains germinate successfully. The optimal sugar concentration may vary from 10 to 80 %, depending on the species. In a short summary VISSER (95) gave various sugar concentrations, which have been employed successfully in germinating pollen. Those workers who believe that the pollen and tubes do not need external food for their germination and development (55, 3, 24, 95), maintain that although pollen grains only develop fully grown tubes if sugar and other 'nutrients' are added, these 'nutrients' or sugar should serve only to change the growth medium in such a way as to promote tube growth without serving as a real nutrient to the pollen. VISSER (95) also states that the 'presence of sugar is only essential for creating favourable conditions for germination', and that 'the pollen tube is exclusively built up from the reserves of the pollen grains'.

As to the discrepancy between the two main views, *viz.* whether nutrients are supplied from an endogenous or an exogenous source, some later experiments convincingly point in favour of the theory of exogenous nutrition. Though various early workers detected reserve materials in the pollen grain (20, 36, 32), they did not consider these materials as present in sufficient quantities to produce pollen tubes long enough to reach the ovules. It was assumed, therefore, that the pollen tube derives its food from the conducting tissue in the style. LINSKENS (54), analysing the styles of the *Petunia* before pollination and again after fertilization, found that the sugar content dropped to half its original quantity after fertilization. When *Pinus densiflora* pollen grains were placed in a sugar solution, an abundant quantity of starch was detected afterwards (87, 88). VASIL (91) also observed that *Typha latifolia* pollen tubes, which were grown on sucrose solutions, accumulate oil globules.

By using C^{14}-labelled sugars, O'KELLEY (61) conclusively proved that pollen tubes respire during growth and absorb externally supplied sucrose, fructose or glucose. The foregoing therefore clearly suggests that sugars, apart from being a controlling agent in the osmotic pressure of the germinating and growing medium, also serve as an external food supply.

4.1.1.3. *Germinating and Growth Promoting Substances*

SCHMUCKER (73) made the important discovery that *Nymphaea* pollen germinated satisfactorily in a 1% glucose solution, to which stigmatic extract was added, but less readily in the same glucose solution without the extract. After an analysis of the stigmatic extract it was discovered that it contained appreciable quantities of boric acid. Three years later, after studying the pollen of 40 plant species, SCHMUCKER (74) concluded that the boron plays a vital role in the germination of pollen. The boric acid did not only improve germination, it also stimulated the development of the pollen tubes. Since then numerous workers like GONDO and TAKAHASHI (29), MOEWUS (56), BATJER and THOMPSON (5), VISSER (95), SCEPTOJEV and POBEGAILO (72), O'KELLY (62), ESQUIVEL (26), VASIL (92) and MÜNZNER (59) also found the stimulating effect of boric acid upon pollen germination and tube growth.

As to the action of boron on the pollen germination and pollen tube growth, several theories have been put forward which sometimes contradict one another. The promoting qualities of borium in pollen germination and pollen tube growth was accordingly attributed to the following possibilities:

(a) Boric acid plays a part in the wall structure of the tube membrane (82, 59, 92).

(b) It accelerates sugar absorption, translocation and metabolism (62, 90, 92).

(c) It increases oxygen uptake (62).

For a more detailed review on the literature concerning the possible functions of borium in pollen germination and pollen tube growth see (JOHRI and VASIL 42).

Even some of the earliest workers pointed out that as the number of pollen grains increases in a constant quantity of growth medium, the germination percentage as well as the tube lengths proportionately increase. KWACK and BREWBACKER (49) suspected that the population effect resulted from the equilibration of a water-soluble pollen growth constituent in the grains and surrounding medium. As a growth medium these workers used 10% sucrose and 100 p.p.m. H_3BO_3. When cell free extracts from large pollen populations were used in the growth medium, these resulted in overcoming completely the 'population effect'. In a series of biochemical studies on pollen and plant extracts and on conventional growth constituents, these writers found the calcium ion to be the pollen growth constituent. They further concluded that, though calcium was not replaceable by ions such as strontium, other cations (Mg^{++}, K^+ or Na^+) were essential for the activating effect of the calcium.

RAGHAVAN and BARUAK (69) found that the addition of indole acetic acid, indole butyric acid, H_3BO_3, $MnSO_4$, $(NH4)_2MoO_4$, $ZnSO_4$ and $AuCl_3$ in non-lethal concentrations to a basal medium of sucrose increased pollen germination and pollen tube growth within one hour.

BOSE (10), treating germinating pollen of *Pisum sativum* with 0.05 mg/l. gibberellic acid, increased pollen tube growth to 7 times that of the control; at higher concentrations the effect was less.

SAWADA (71) found that *Paris hexaphylla* pollen failed to germinate in an ordinary

sugar – agar medium, but was stimulated to germinate when aspartic acid, glutamic acid, histidine or cysteine was added to the germinating medium.

POTANINA and SMIGEL (67) even found that by subjecting apple and pear pollen to electric fields of 2,4, and 8 kv/cm for 10, 30 or 60 seconds, they could improve pollen germination, the length of the pollen tubes and longevity of storage.

Apart from sugar and boron, various other substances have been used to stimulate pollen germination and pollen tube growth. Of all the substances tested, as can be seen from the above, none has given such satisfactory results as boron.

4.2. VIABILITY OF POLLEN

4.2.1. Germination and Speed of Pollen Tubes

Experimental: Pollen of the Schwarz 21 cultivar, grown at a constant temperature of 28°C was sampled at the beginning of the light period. The standard germinating medium + H_3BO_3 (50 p.p.m.) was used.

In this case a microscope was installed in a temperature room, where the temperature and relative humidity were kept at 33°C and 75%. Under these conditions it was possible to observe germination and to follow the growth rate of the pollen tubes uninterruptedly. The germinating time as well as the pollen tube speed were obtained by taking the time necessary for the first 5 pollen grains to germinate and to follow their elongation minute by minute.

Results and discussions: Under these circumstances pollen tube growth continued for about 35 minutes. No further growth in any of the other pollen tubes was observed after 50 minutes and none of the pollen tubes reached a length of more than 1362 μ. The abrupt stop was most probably caused by the low humidity in the temperature room. In some later experiments, however, when the glass slides with pollen were put in petri-dishes, a higher humidity was obtained and pollen tube growth continued for much longer periods.

Because of the high humidity required and the availability of only two temperature rooms, which were usually fully occupied by plants, the influence of only one temperature could be studied.

The first sign of germination, i.e. the bulging of the tubes, was observed after 9 minutes. After the first pollen tubes had appeared, the elongation was very rapid and was almost directly proportional to time.

The pollen tube elongation during the first 5 minutes after germination was 132 μ, for the second-, third-, fourth- and fifth five minute intervals the pollen tube elongations were respectively 199 μ, 232 μ, 199 μ and 166 μ.

It was thus clearly shown that on an artificial medium, the early growth rates of the pollen tubes can reach speeds up to an average of 39 μ per minute.

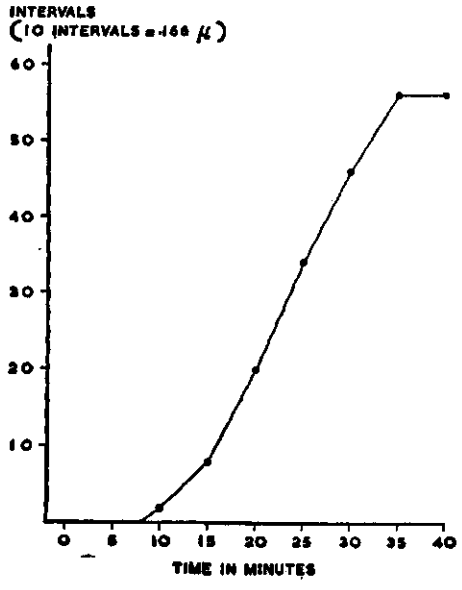

Fig. 8. Elongation of pollen tubes when pollen was collected from plants grown at 28°C and germinated at 33°C. (Standard germination medium + H_3BO_3 – 50 p.p.m.)

From the foregoing it may be deducted that when the environment is favourable and remains favourable, the growth rate of the pollen tubes will be more or less constant.

4.2.2. Sampling Date

Experimental: Pollen from the Schwarz 21 cultivar was sampled during the first week (5th day) and third week (16th day) from plants which were grown at constant temperatures of 24° and 33°C as well as from a control in a greenhouse. The methods for obtaining the pollen germination percentages and maximum pollen tube lengths were the same as described in the chapter dealing with materials and methods. No borium was added to the germination medium, which consisted of 1 g agar, 8 g cane sugar + 100 ml water.

Results and discussions: (a) Germination.

With regard to the pollen sampled from the plants grown at 24° and 33°C, it can be seen in fig. 9 that the pollen germination was higher during the first than during the third week. The curves representing the pollen germination of the plants grown at 24°C indicate, however, that these differences were relatively small, being never higher than 5 % at any germination temperature except at a germination temperature of 33°C, where the germination percentage dropped from 16% during the first week

to 0.1 % when sampled during the third week. This high germination percentage must, however, be regarded as exceptional because all the other pollen samples investigated showed definitely low germination at 33C°, pollen sampled during the third week from plants grown at 33°C even yielded very low germination percentages from the beginning.

The curves representing the pollen germination of plants grown at 33°C were quite striking as can be seen in fig. 9. The germination percentage was relatively high during the first week but diminished to such an extent during the third week that no germination whatsoever could be obtained at germination temperatures of 21°, 24° and 36°C. The germination obtained at the other temperatures, 27°, 30° and 33°C was, however, much lower during the third week than in the samples taken during the first week. Striking is the fact that 33°C, representing a minimum germination

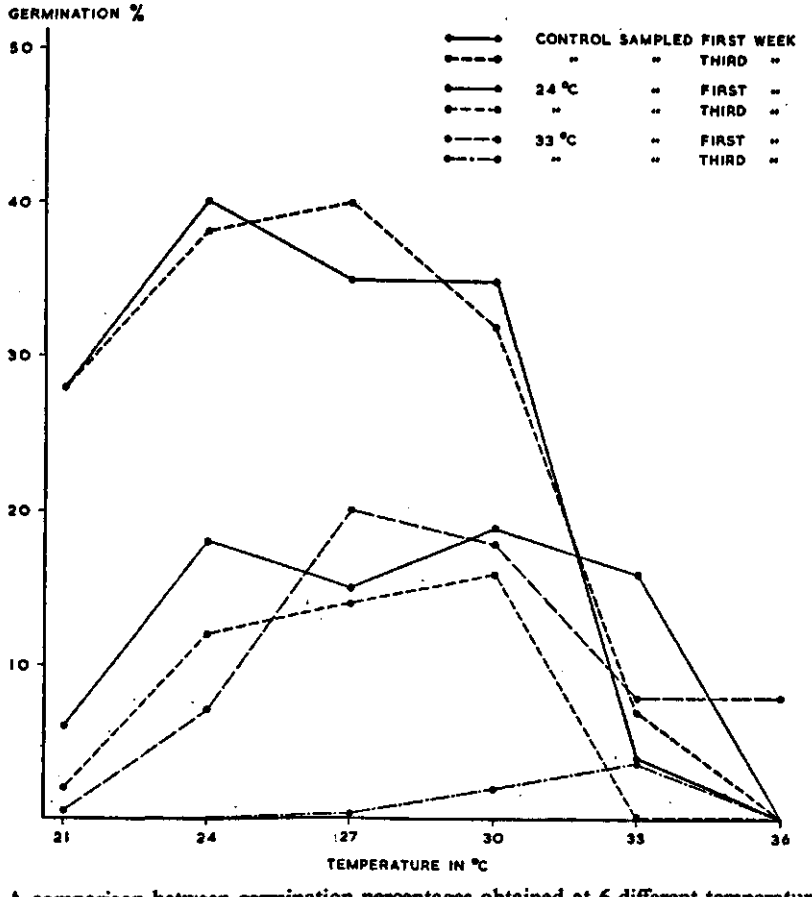

FIG. 9. A comparison between germination percentages obtained at 6 different temperatures when pollen was collected during the first and third week of flowering. (Standard germination medium without H_3BO_3.)

temperature during the first week, effectuated a maximum result during the third week. This maximum is, however, lower than the minimum obtained during the first week, suggesting that the entire character of the pollen, produced at 33°C, had changed from the first to the third week. This change of character was not observed when plants were grown at 24°C or in the greenhouse control. Disregarding pollen produced at 33°C, the germination percentages recorded at the various temperatures followed a more or less fixed pattern. As the curves indicate, germination is relatively low at 21°C, it reaches an optimum at germination temperatures of 24°, 27° and 30° and drops to zero and in some instances to nearly zero at 33° and 36° C.

From the foregoing it follows that:

(1) The germination of pollen, sampled from plants grown as a control in a greenhouse was approximately the same when sampled during the first and third week of flowering.

(2) The germination of pollen sampled from plants grown at 24°C was slightly higher when sampled during the first week than during the third week of flowering.

(3) The germination of pollen sampled from plants which were grown at 33°C was much higher when sampled during the first week than during the third week.

(4) Apart from the pollen produced at 33°C, the germination percentages represent an optimum curve with a rather broad optimum between the germination temperatures of 24° and 30°C.

The pollen produced by the plants grown at 33°C was not as seriously affected by the high temperature during the first phase of flowering as during the later phase. Although undoubtedly partly affected by the high temperature, it is rather strange that the pollen sampled during the third week was so much less viable than that sampled during the first week. An explanation for this phenomenon may be deducted from the findings of VAN ROSSEM and BOLHUIS (70) who stated that: 'Investigations on the axillar buds of the cotyledons pointed to the fact that in the earliest phases of development there exists floral initiation in the peanut plant'. FORTANIER (27) also mentions that the first flowers are already present in the buds of the cotyledons. From this it may be inferred that the behaviour of the pollen, sampled during the first phase of flowering, was already partly determined in the seed.

(b) Pollen tube growth.

Considering the results presented in table 16, it becomes evident that in the cases where the germination was affected, fig. 9, the pollen tube growth was affected accordingly. The pollen tube lengths, obtained from pollen of plants grown as the control, were the same whether sampled during the first or third week of flowering.

The pollen tubes obtained from pollen of plants grown at 24°C were much longer when sampled during the first than during the third week of flowering. At a germination temperature of 30°C and a germination period of 150 minutes for example, the pollen tubes attained an average maximum length of 1128 μ when sampled the first week of flowering and only 581 μ when sampling took place during the third week of flowering.

TABLE 16 A comparison between the maximum pollen tube lengths (μ) obtained after 150 min. when pollen was collected on the 5th and 16th day of flowering. Pollen was germinated at 6 successive temperatures and plants were grown at 24°, 33°C and in a greenhouse as a control. (Standard germination medium without H_3BO_3)

Plant growth temperatures	Pollen collected	Pollen germination and tube growth temperatures					
		21°C	24°C	27°C	30°C	33°C	36°C
24°C	5th day	116	996	996	1128	498	0
	16th day	83	415	747	581	149	83
33°C	5th day	32	498	547	498	929	664
	16th day	0	0	83	166	249	0
Control	5th day	946	1958	1958	1660	1510	0
	16th day	962	1925	1842	1660	1494	0

4.2.3. Sampling Time during the Day

The exchange of stored pollen between plant breeders has been common practice during recent years. It is important to know, therefore, at what time pollen should be collected to obtain the highest viability of the pollen. In germination determinations it is also important to know whether time of sampling has any influence on viability of pollen.

In the beginning of our pollen investigation it was found that the germination percentages recorded, as well as the final pollen tube lengths recorded varied considerably during the day, although the same plants, grown under the same environmental conditions were chosen for the pollen collections. This was especially true in the case of plants grown at the high temperature. The foregoing gave reason to believe that time of sampling must have had a greater influence on germination than was anticipated.

Experimental: Plants of the Schwarz 21 cultivar, which were grown at 24°, 33°C and as a control in a greenhouse, were used for the pollen collection. Samples were taken at a ½ of an hour, 4 hours, 7 hours and 10 hours after the light was switched on. Pollen from the control plants was collected at 6, 10, 1 and 4 o'clock. All the pollen samples were germinated at 30°C. All further methods were the same as previously described, except that no H_3BO_3 was used in the germination medium.

Results and discussions: Our results clearly suggest that there must be a specific period in the maturation of pollen when maximum germination will result and reversely also a period when the contrary will be in force.

Table 17 gives the germination percentages as well as the pollen tube lengths

TABLE 17 Pollen germination and pollen tube growth (μ) of pollen originating from plants grown at 24°C, 33°C and as a control in a greenhouse when sampled at different times during the day. Germination percentages obtained after 150 min. while pollen tube lengths were determined at 30 min. intervals. (Germination temperature 30°C; standard germination medium without H_3BO_3)

		Sampled in hours after illumination started							
		½		4		7		10	
	Intervals in min.	Ger. %	Length	Ger. %	Length	Ger. %	Length	Ger. %	Length
Plants grown at 24°C	30		282		166		166		83
	60		381		166		415		249
	90		747		415		498		298
	120		1128		581		498		298
	150	19.0	1128	12.0	581	8.0	498	5.0	298
Plants grown at 33°C	30		265		166		83		83
	60		398		198		166		166
	90		498		198		198		198
	120		498		198		198		198
	150	18.0	498	1.0	198	1.0	198	1.0	198
Plants grown in greenhouse as control	30		415		378		83		83
	60		996		498		265		83
	90		1577		747		348		83
	120		1660		996		498		83
	150	35.0	1660	21.0	996	7.0	498	3.0	83

determined for plants grown at 24°, 33°C and for the control after periods of 30, 60, 90, 120 and 150 minutes.

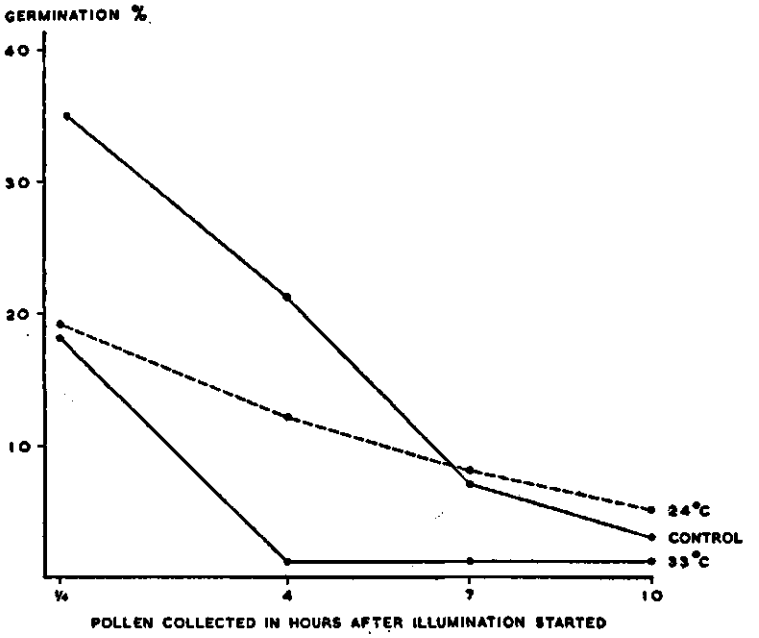

FIG. 10. Pollen germination as influenced by time of sampling. (Germination temp. 30°C; germination medium without H_3BO_3).

In the light of the data expressed in fig. 10, one can see that germination decreases from a ½ hour after illumination began till the final observation 10 hours after illumination had started. The above holds true for all three instances *i.e.* for plants grown at 24°, 33°C as well as for those grown as the control except that the observations made at a ½, 4, 7 and 10 hours in the case of the first mentioned plants, must be compared with the observations made at 6, 10, 1 and 4 o'clock in the case of the control plants. In the control the germination gradually decreases from 6 to 10 o'clock. From 10 to 1 o'clock the fall in germination is much steeper than from 1 to 4 o'clock when a gradual decline in germination was observed. For plants grown at a constant temperature of 33°C, the decline in pollen viability is quite striking. The germination percentage drops from 18 to 1 in a period of four hours; from then on it remains at the same low level for more or less the rest of the period investigated. The pollen viability of the plants grown at a constant temperature of 24°C shows a more gradual decline. From a germination percentage of 19 the moment the light was switched on, it falls to 5 percent 10 hours afterwards. This means that the pollen sampled from plants grown at 24°C retains its viability longer than in the other two treatments. Although the actual germination percentage for plants grown at 24°C is much lower

than the percentage recorded for the control plants, the viability or germination percentages are the same after 7 and 10 hours light or perhaps even slightly higher.

From the foregoing the obvious deduction to be made is that the higher the temperature, the quicker drops the viability of the pollen.

As far as the groundnut and especially the Schwarz 21 cultivar is concerned, it will most probably be true to say that pollen retains its viability longer at low than at high temperatures.

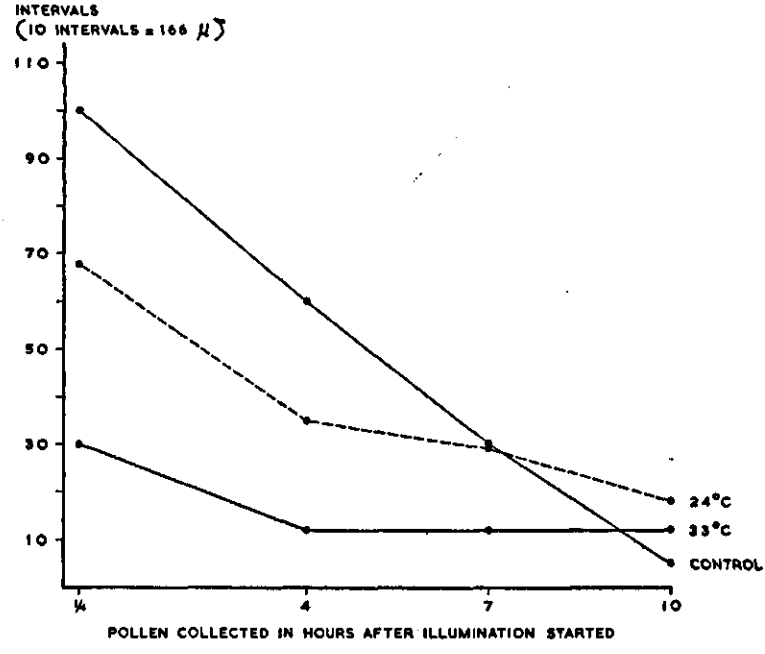

FIG. 11. Maximum pollen tube lengths as influenced by time of sampling. (Growth time 150 minutes at 30°C; germination medium without H_3BO_3).

The pollen tube lengths obtained at the different treatments are shown in fig. 11. When germination takes place shortly after illumination started, the maximum lengths of the pollen tubes are much higher than when germination occurs 4, 7 or 10 hours after illumination began. These results hold true for all three treatments investigated.

When examining each treatment separately, one observes that the maximum pollen tube lengths for the control plants are 1660 μ, 996 μ, 498 μ and 83 μ when germination took place respectively at 6, 10, 1 and 4 o'clock. For the plants grown at a constant temperature of 24°C, the pollen tube lengths are the following; 1128 μ after a ¼ of an hour illumination, 581.0 μ after 4 hours illumination, 498.0 μ after 7 hours illumination and 298.8 μ after 10 hours illumination.

The results, therefore, show that both the germination and the final pollen tube lengths obtained after 150 minutes, decreased considerably from a ¼ of an hour after illumination started to 10 hours after illumination. It may thus be assumed that germination and tube growth of pollen are high at the time of anthesis (± at the beginning of the light period) and then decrease rapidly to a very low value 10 hours after the illumination began.

To our knowledge the only reference to date regarding viability of groundnut pollen was made by OAKS (60), who found significant differences in pollen viability as pollen collections were made at different stages of physiological development. INONE and SHIBUYA (38), working with the Kentucky Wonder and the dwarf Masterpiece bean, found that the percentage of pollen germination increased towards the time of anther-dehiscence and then decreased rapidly, the grains losing their vitality about 5 to 6 hours after anthesis. Our results would seem to support this view and show in addition that the higher the temperature, the sooner the loss of vitality. WERFFT (104), studying the viability of pollen in the atmosphere, found that pollen germination of *Adonis vernalis*, *Tulipa* spp., *Pirus communis*, *Betula pendula etc.*, was more or less impaired by 8 hours exposure to solar radiation. With *Carpinus betulus*, *Deutzia scabra* and *Digitalis purpurea*, for instance, over 90% of the pollen grains lost their germination capacity. From his experiments with ultra-violet light he concluded that the action of the sun on the pollen is due to the ultra-violet rays. It is certain, however, that although the drop in pollen viability may greatly be attributed to the action of light, high temperatures definitely activate this detrimental process as can be seen from our experiments.

(1) The later in the day pollen sampling takes place, the lower will be the pollen viability.

(2) The higher the temperature, the sooner drops the pollen viability.

(3) In order to obtain the highest viable pollen, pollen collection should take place as early as possible while the temperature is still low.

(4) For comparative pollen studies, sampling should be done at the same time of the day, preferably as early in the morning as possible.

4.3. VIABILITY OF POLLEN AS INFLUENCED BY TEMPERATURE

4.3.1. Constant Temperature Conditions

The influence of temperature on pollen germination and pollen tube elongation was investigated by workers such as VISSER (95), KING and JOHNSON (47) and VASIL and BOSE (93), just to mention a few. Very few workers, however, investigated the effect of plant growth temperatures on the viability of pollen. An exception in this case is perhaps the work of SEMENUIK (76), who studied the effect of different temperatures

on the seed production of *Matthiola incana* and also included the effect of plant growth temperature on pollen viability. YAMADA and HASEGAWA (106), working with the rice plant at different soil temperatures, tried to clarify and establish a relationship between pollen germination and grain fertility.

Due to lack of information on the influence of growth temperatures on pollen viability and the fact that temperature has a great influence on seed production in the groundnut plant, various experiments were carried out by the author in order to determine whether any relationship between temperature, pollen viability and seed production is existing.

Before discussing the influence of temperature on the characteristics of pollen, it seems important to draw attention to the fact that pollen germination and pollen tube elongation experiments took place in *vitro*. It may be questioned, therefore, whether results, obtained on an artificial medium, can serve as an indication of pollen vitality and its ability to set fruit under natural conditions. It is well known that, although pollen sometimes fails to germinate in *vitro*, satisfactory fruit set may occur under natural conditions (83, 63). It may, however, be assumed that when pollen germinates well in vitro and responds to climatic variations, the chances of fruit set under natural conditions will be greater than when germination is poor or nil in *vitro*.

When pollen, *e.g.* that of apple, pear and groundnut, is known to germinate in vitro, the germination percentages on artificial media seem to be well correlated with its ability to set fruit.

From VISSER's (95) apple and pear pollination experiments the following relationship between percentage germination in vitro and fruit set was obtained:

germination <20% — — — — — fruit set poor to nil;
germination 20 to 40% — — — — — fruit set poor to moderate;
germination 40 to 60% — — — — — fruit set moderate to normal;
germination >60% — — — — — fruit set normal.

The above suggests, therefore, that when pollen is able to germinate in vitro, this germination vigour may be considered as a reliable standard as to its ability and potentialities in fruit set.

Experimental: Pollen from the Schwarz 21 cultivar, grown at 24°, 33°C and at a control was sampled during the first week of flowering as well as on the 16th day of flowering. Sampling of pollen took place at the beginning of the light period, whereas the pollen of the control plants was sampled about 1 hour after day break.

Pollen was germinated and grown at 5 different temperatures *viz.* 21°, 24°, 27°, 30° and 33°C and in some cases an additional determination was carried out at 36°C.

Determinations were first made without and later with H_3BO_3 in the germination medium.

Results and discussions: (a) Pollen tube elongation: When plants were grown at 24°C, pollen tube growth was detrimentally affected by 21°C where pollen tube growth

was practically nil. At 24°, 27° and 30°C pollen tube elongation was more or less equal but strikingly better than at 21°C, while 33°C was obviously too high a temperature for 'normal' pollen tube elongation.

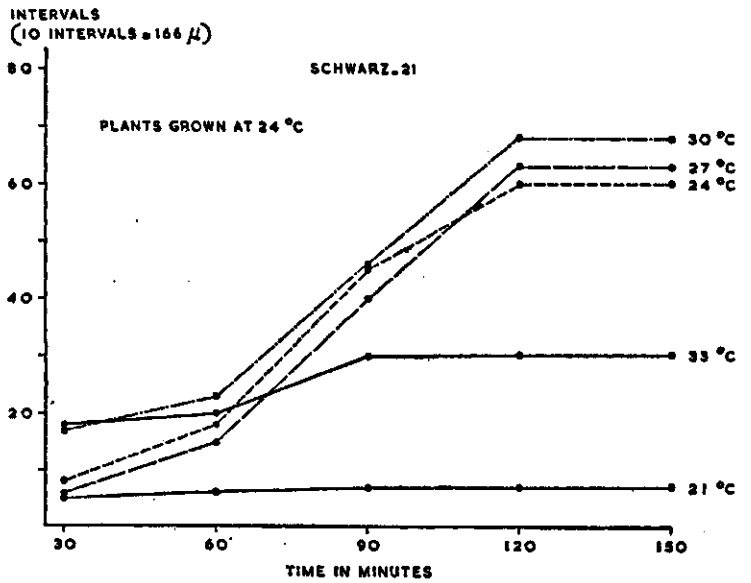

FIG. 12. Pollen tube elongation at 5 different temperatures when plants were grown at 24°C and pollen collection took place during the first week of flowering. (Germination medium without H_3BO_3).

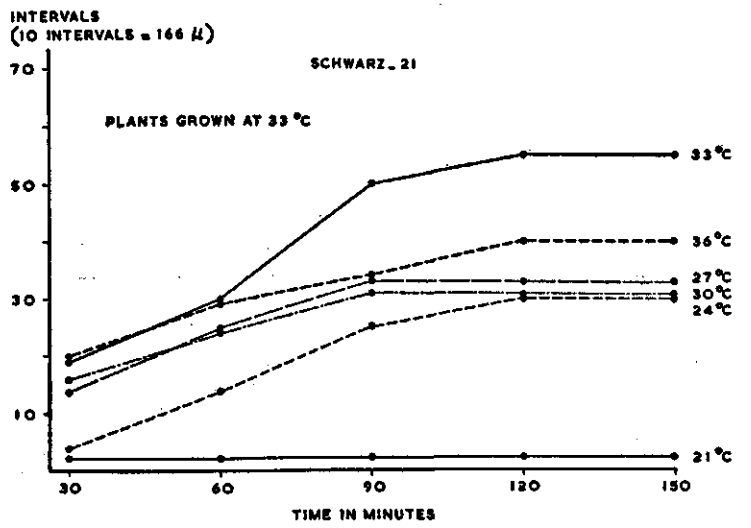

FIG. 13. Pollen tube elongation at 6 different temperatures when plants were grown at 33°C and pollen collection took place during the first week of flowering. (Germination medium without H_3BO_3).

Plants which were grown at 33°C produced pollen which showed a low activity. No pollen tube elongation occurred at 21°C, while the best results were obtained at the high germination temperatures, 33° and 36°C. The results suggest, therefore, that when plant growth temperatures are high, pollen tube elongation seems to be favoured by the same high germination temperatures.

The pollen produced by the control plants was much more viable than pollen obtained from plants grown at either 24° or 33°C. In this case the growth rate of the pollen tubes increased as the temperature was raised from 21° to 27°C and then gradually decreased as the temperature was further raised from 27° to 33°C.

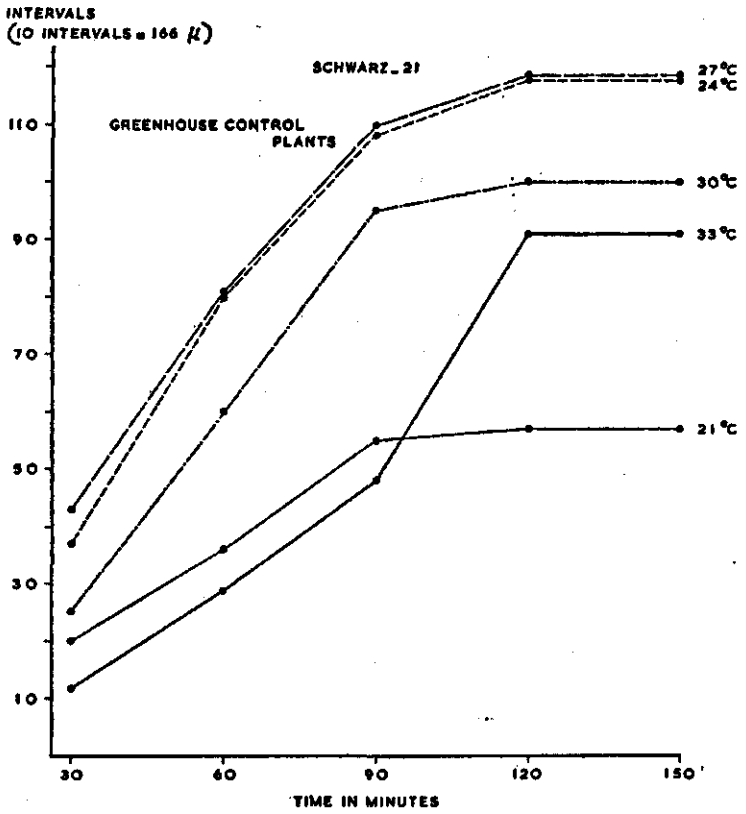

FIG. 14. Pollen tube elongation at 5 different temperatures when plants were grown in a greenhouse as a control and pollen collection took place during the first week of flowering. (Germination medium without H_3BO_3).

From these results, where germination and pollen tube growth took place in the absence of H_3BO_3, it was shown that plant growth temperature had a marked effect on pollen tube growth. The activity of pollen was highest when plants were grown as a control in a greenhouse, somewhat lower when plants were grown at a constant

temperature of 24°C and exceedingly low when plants were grown at a constant temperature of 33°C.

The addition of H_3BO_3 to the germination medium greatly improved the activity of the pollen from plants grown at 24°C. In this case the same level was obtained as with the pollen of the control plants. Although the maximum pollen tube lengths obtained after 150 minutes in the temperature cabinets were greater at 24°, 27° and 30° in the control pollen, the pollen from plants grown at 24°C resulted in longer tubes at 33° and 36°C.

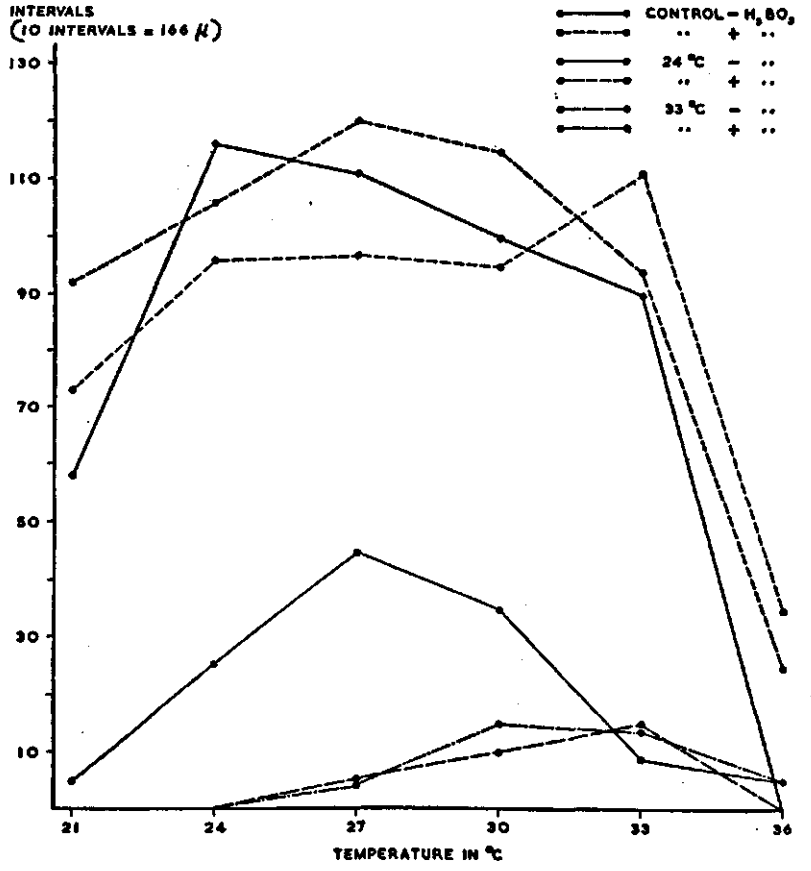

FIG. 15. Maximum pollen tube lengths obtained at 6 different temperatures, with and without H_3BO_3 (50 p.p.m.) in the germination medium. (Pollen collection took place during the third week of flowering; germination and growth time 150 minutes).

It is evident that the addition of H_3BO_3 to the germination medium could not induce a higher pollen activity when pollen was produced at 33°C. The same low activity, *i.e.* short pollen tubes, (fig. 15) was obtained in both the germination mediums with and without H_3BO_3.

From the foregoing it seems that when the activity or viability of pollen drops below a certain level, as is the case with pollen produced at 33°C, boron does not seem to be able to stimulate the pollen to a higher activity.

(b) Pollen germination: Fig. 16 shows clearly that pollen germination was affected in a similar way by plant growth temperatures as were pollen tube elongations.

When H_3BO_3 was omitted from the germination medium, germination percentages were relatively high in the control, whereas they were much lower for plants grown at 24°C and very low for plants grown at 33°C, where germination was only slight at 27°, 30° and 33°C and nil at the other temperatures.

When H_3BO_3 was added to the germination medium, the germination percentages

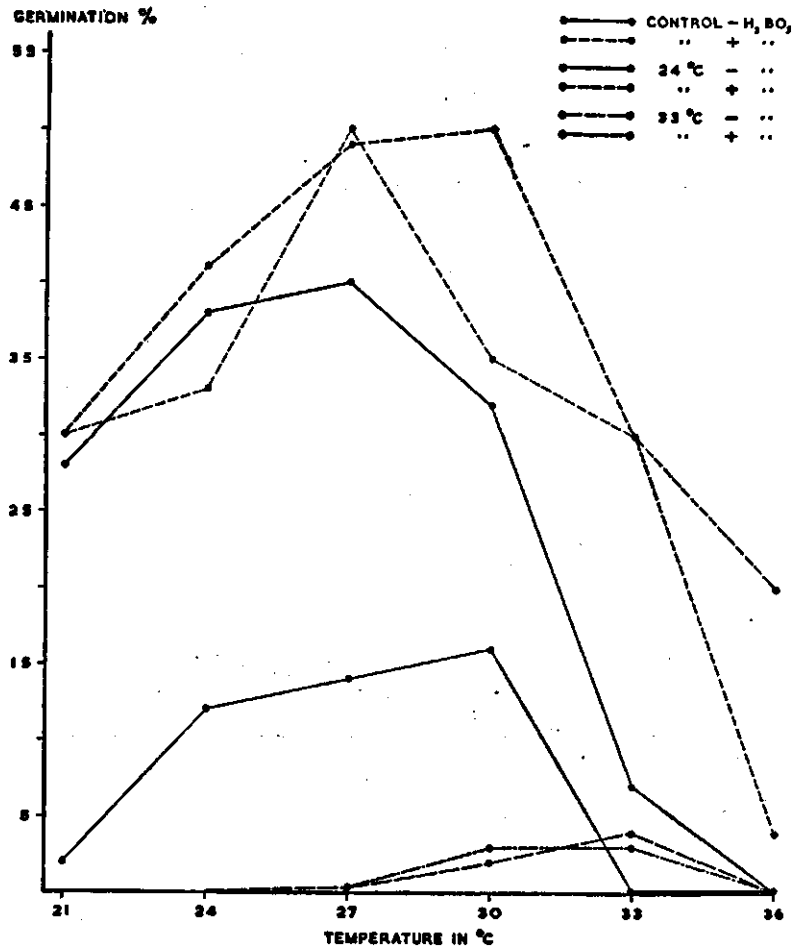

FIG. 16. Germination percentages obtained at 6 different temperatures, with and without H_3BO_3 (50 p.p.m.) in the germination medium. (Pollen collection took place during the third week of flowering; germination time 90 minutes).

were more or less the same when pollen was sampled from the control plants and from plants grown at 24°C. This was also the case with pollen tube elongation; here pollen germination was stimulated by an addition of boric acid to the germination medium. However, the addition of H_3BO_3 did not stimulate pollen from plants grown at 33°C, where the same low germination percentages were recorded as when H_3BO_3 was omitted from the germination medium.

From the foregoing it is logical to conclude that an addition of borium to the germination medium can stimulate pollen produced at 24°C as well as at the control but not when produced at a temperature as high as 33°C. The viability of pollen produced at 33°C was apparently too low to be activated by an addition of borium to the germination medium.

Table 18 summarises the results obtained at different temperatures with and without H_3BO_3 in the germination medium.

4.3.2. Effect of a change in temperature during flowering

Temperatures may fluctuate considerably during the growing season. An investigation was carried out to determine whether a temperature change during the flowering period has any influence on pollen viability.

Experimental: 5 Plants from each of the two main temperature treatments, *i.e.* plants grown at 24°C and plants grown at 33°C, were transferred and subjected to the opposite temperatures. In both cases the plants were already three weeks in bloom.

As a result of the foregoing method, pollen from plants grown at 24°C and then subjected to a temperature of 33°C and *vice versa* could be studied (24° × 33° and 33° × 24°C).

Pollen was sampled 36 and 96 hours after the plants had been subjected to the higher and lower temperatures.

16 Plants were kept at constant temperatures of 24° and 33°C to serve as controls. Germination percentages and pollen tube elongations were determined as already described.

Results: From the data recorded in table 19 it may be seen that the change in temperature did not affect the plants grown at a constant temperature of 24°C and then transferred to a temperature of 33°C. After 36 hours in the new environment, the pollen continued to behave in the same way as that from plants kept at a constant temperature of 24°C, although the germination percentages dropped slightly in both the germination media without and with H_3BO_3.

The plants grown at 33°C showed low germination percentages even before the plants were transferred. When they were subjected to the lower temperature of 24°C, the pollen germination percentages further decreased to zero in both media.

When the plants had been subjected to both the higher and lower temperatures

TABLE 18 Germination and pollen tube growth in (μ) of pollen originating from plants grown at 54°C, 33°C and as a control in a greenhouse at 6 different germination temperatures with and without 20 p.p.m. H_3BO_3 in the germination medium. Germination percentages were obtained after a germination period of 150 min. and the pollen tube lengths at 30 min. intervals

	Intervals in min.	—H_3BO_3												+H_3BO_3											
		21°		24°		27°		30°		33°		36°		21°		24°		27°		30°		33°		36°	
		G*	L**	G	L	G	L	G	L	G	L	G	L	G	L	G	L	G	L	G	L	G	L	G	L
Plants grown at 33°C	30		—		—		83		83		99		—		—		—		66		83		83		83
	60		—		—		83		166		166		—		—		—		66		166		166		83
	90		—		—		83		166		249		—		—		—		66		249		232		83
	120		—		—		83		166		249		—		—		—		66		249		232		83
	150	0	—	0	—	0.3	83	2	166	4	249	0	—	0	—	0	—	0.3	66	3	249	3	232	0.1	83
Plants grown at 24°C	30		49		166		166		166		179		83		332		382		830		581		630		415
	60		83		365		315		199		179		83		664		830		1162		1079		1261		581
	90		83		415		498		415		179		83		1211		1162		1510		1494		1643		581
	120		83		415		747		581		179		83		1211		1593		1577		1577		1842		581
	150	2	83	12	415	14	747	16	581	0.1	179	0.1	83	30	1211	33	1593	50	1610	35	1577	30	1842	20	581
Control plants	30		332		664		747		415		166		—		398		664		913		830		664		332
	60		597		1328		1328		996		498		—		747		1411		1494		1245		996		415
	90		913		1925		1743		1577		796		—		1460		1660		1992		1909		1477		415
	120		962		1925		1842		1660		1449		—		1527		1759		1992		1909		1560		415
	150	28	962	38	1925	40	1842	32	1660	7	1449	0	—	30	1527	41	1759	49	1992	50	1909	30	1560	4	415

* G – Germination % ** L – Length

for 96 hours before pollen was sampled, the complex of the results changed somewhat. In the case of the 24° × 33°C plants, the pollen germination percentages as well as the pollen tube lengths recorded after 36 hours dropped after 96 hours from 12 and 33% to 0.5 and 6% for the treatments without and with H_3BO_3 respectively. In correlation with the germination percentages recorded, the pollen tube lengths diminished from 664 and 1441 μ to 116 and 365 μ. The influence of the change in temperature was inversely the same when plants were taken from the high to the low temperature. Both the germination percentage and the pollen tube lengths improved strikingly. From a germination percentage of 0 and no pollen tube growth after 36 hours, germination percentages of 2 and 30 were recorded for the treatments without and with H_3BO_3, after 96 hours at the lower temperature. The maximum pollen tube lengths after a germination time of 150 minutes were 415 and 1743 μ for the treatments without and with H_3BO_3 respectively.

After studying the foregoing results, they may be grouped as follows:

24°C Control $\approx$ 24° × 33°C after 36 hours.

24°C Control $\approx$ 33° × 24°C after 96 hours.

33°C Control $>$ 33° × 24°C after 36 hours.

33°C Control $\ll$ 33° × 24°C after 96 hours.

Discussions: Although no pollen germination was obtained after 90 minutes with the 33° × 24°C plants, their pollen showed some amount of germination after 48 hours

in the germinator. Approximately 4% of the pollen germinated in this specific case and the pollen tube lengths were about 581.0 μ for both the treatments without and with H_3BO_3. As previously stated, however, pollen grains that showed no signs of germination during a time lapse of 90 minutes were regarded as complete failures. From this phenomenon it became evident that if the time allowed for germination is long enough, in this case 48 hours, even pollen showing no signs of germination after 90 minutes may prove to be viable. It may safely be assumed, however, that pollen which does not germinate before 48 hours have elapsed, is unlikely to play an important role in the fertilization process and may therefore be disregarded.

The results compiled in table 19 indicate that the activity of pollen, except when already low as in the 33°C control, was not seriously affected by a change in temperature for 36 hours. The germination as well as the pollen tube lengths were only affected where the plants had been subjected to the higher and lower temperatures for more than 36 hours, *i.e.* for 96 hours. The pollen sampled from plants which were transferred from 33° to 24°C and kept there for 96 hours, reacted in a similar fashion as the plants which were grown at a constant temperature of 24°C. The opposite is also true, because the pollen sampled from the 24° × 33°C plants reacted after a subjection for 96 hours as when grown continuously at 33°C.

With respect to the results obtained in the foregoing experiments, the following deductions may most probably be made:

TABLE 19 Pollen germination and pollen tube growth (μ) when pollen was collected 36 and 96 hours after plants grown at 24° and 33°C were transferred to 33° and 24°C respectively. (Germination % obtained after 150 minutes and tube lengths at 30 minute intervals. Germination medium with and without H_3BO_3)

	Intervals in min.	24°C				24° × 33°C				33°C				33° × 24°C			
		$-H_3BO_3$		$+H_3BO_3$		$-H_3BO_3$		$+H_3BO_3$		$-H_3BO_3$		$+H_3BO_3$		$-H_3BO_3$		$+H_3BO_3$	
		G*	L**	G	L	G	L	G	L	G	L	G	L	G	L	G	L
36 hours after transfer	30		166		581		166		249		83		83		0		0
	60		199		1079		398		747		166		249		0		0
	90		448		1494		664		1162		166		332		0		0
	120		747		1593		664		1411		166		332		0		0
	150	20	747	35	1593	12	644	33	1411	2	166	3	332	0	0	0	0
96 hours after transfer	30		83		664		83		132		0		32		83		747
	60		249		1294		116		315		0		66		265		1477
	90		415		1693		116		365		0		66		415		1743
	120		597		1693		116		365		0		66		415		1743
	150	16	597	37	1693	0.5	116	6	365	0	0	2	66	2	415	30	1743

* G – Germination %
** L – Length

(1) If the time allowed is long enough (48 hours), germination may sometimes result, although no germination may take place in a germination time of 90 minutes.
(2) The growth temperature has no permanent influence on the behaviour of the pollen.
(3) The growth temperature during the vegetative phase of the plant (± the first 25 days) has little or no influence on the behaviour of pollen.
(4) The behaviour of pollen may be altered according to the temperature applied during the flowering period.
(5) The character of pollen at flowering is not seriously influenced by the reigning temperature (within limits), but is influenced by the temperature 36 to 96 hours preceding the actual opening of the flowers. This may serve to explain why the germination percentages of pollen from control plants (greenhouse plants) are apt to vary so considerably, although sampled at more or less the same temperature, but not on the same day. HOLMAN and BRUBAKER (35) found that such irregularities may occur, 'when the pollen is collected from apparently equally mature flowers on different days or in different, though neighbouring, localities, and even when it is taken from flowers at the same age in a given locality and at about the same time'. Although various factors may influence pollen characteristics, we feel that, according to our results, some of these irregularities may be attributed not to a difference in temperature during sampling, but perhaps to a difference in temperature some time before the actual opening of the flowers.
(6) A sudden change from a high to a low temperature is more detrimental to the activity of the pollen already produced (pollen in a premature stage in flowerbud), than a change from a low to a high temperature.

4.3.3. Pollen Shed

When carrying out pollination experiments, several workers pointed out that variable quantities of pollen could affect fertilization considerably. It was usually found that large quantities of pollen gave higher fertilization percentages than small quantities. Conducting experiments with radishes, LI (51) for instance found that larger quantities of pollen produced more pods per plant as well as more seeds per pod. AIZENSTAT (1) even found that an addition of pollen unrelated to the pollen of peas and tomatoes increased the set and size of the fruit. KLIMENKO and KLIMENKO (48) showed that the pollination of some oranges, mandarins and pomeloes with mixed pollen was beneficial for fruitsetting and seed development. From his results with peas and tomatoes, EISENSTAT (25) concluded that the addition of alien pollen had a favourable effect on fertilization when either the quantity or quality, as the case may be, was unsatisfactory.

The results and conclusions of the above mentioned workers tend to suggest that the quantity of pollen produced may have a greater influence on fertilization than was previously suspected.

Experimental: Flowers of the Schwarz 21 cultivar, produced at 24°, 33°C and as a control in a greenhouse, were collected shortly after illumination started. The anthers of the flowers were exposed and then gently brushed on dry microscope slides, spreading the pollen evenly out so as to make counting as easy as possible.
Results and discussions: The average number of pollen grains set free by means of the method described, is presented in table 20.

It is shown that at 33°C the number of pollen grains obtained was far less and more irregular in number, ranging from 190 to 1762 per flower, than at the control or at 24°C. The average number of pollen grains set free at the control as well as at 24°C was more or less equal, both treatments yielding almost 4 times more pollen per flower than in the case of plants grown at 33°C.

TABLE 20 The number of pollen grains per flower when plants were grown at 24°C, 33°C and as a control in a greenhouse

Flower No.	24°C	33°C	Control
1	2438	190	2231
2	3549	1762	2832
3	4048	1162	2890
4	3360	181	2956
5	3515	1747	3415
6	3289	743	3959
7	2980	355	3966
8	3757	957	3792
9	3586	1405	3377
10	3359	1363	3372
Total	33881	9865	32790
Average	3388.1	986.5	3279.0

At 33°C a degeneration of some of the anthers was observed; they were brittle and tended to break off when brushed over the microscope slides.

The true cause of the low pollen shed at 33°C is yet unknown; among the possibities are hampering of the reduction division and retardation of the succeeding pollen development by the high temperature. Although temperature may influence pollen shed in many ways, no investigation as to its direct influence was carried out. A more detailed study therefore appears to be necessary.

To us the important fact remains that less pollen is set free at the high temperature. It is possible that the relatively small quantity of pollen, produced at 33°C, may have largely contributed to the low fertilization percentages obtained when plants were grown at 33°C.

4.4. VIABILITY OF POLLEN AS INFLUENCED BY BORON SPRAYS

The effect of boron added to the germination medium held the interest of numerous

workers. As already stated workers such as HUANG (36), BATJER and THOMPSON (5), VISSER (95), and VASIL (92), found borium to have a stimulating effect on the germination of pollen. A question which arises, however, is whether this phenomenon is a symptom of boron deficiency of the plant in relation to its pollen, or just a deficiency of the pollen itself. VISSER (95), who tried to answer this question by spraying separate branches from one tree with boric acid, because of the diversity of his results could not find a satisfying answer. He concluded: 'When the differences between all separate treatments and their controls are compared, irrespective of their significance, it is found that the treated branches give a higher yield than the control in 21 instances and a lower one also in 21 instances'. The results imply, therefore, that the effect of boron sprays on the total fruitset has been insignificant. The same worker investigated the effect of boron and sugar applications to the branches of pear trees on the germination of their pollen in water or 10% sugar solution with- and without 30 p.p.m. H_3BO_3. In his own words the results were as follows: 'These results imply that the supply of sugar, either alone or in combination with boron, to the branches, had no significant effect on the germinability of the pollen'. The negative results of this author are not surprising because he also stated that the fruitset was very good in all cases, which implies that the activity of the pollen must also have been high. He could hardly expect, therefore, to improve the viability of already highly active pollen, by means of boric acid sprays.

None of the above mentioned investigators studied pollen activity in relation to plant growth temperatures, and consequently no studies have been published on the effect of a boron spray on plants grown at relatively high and low temperatures. In our previous experiments it was shown that especially a high temperature at flowering lowers the activity of pollen, and that the activity of pollen produced at a low temperature of 24°C could be improved by supplying borium to the germination medium. This suggests that comparatively low, but perhaps even more specificly high temperatures, restrict the borium uptake of groundnuts or its translocation to the flowering parts of the plant. The influence of borium sprays on the activity of pollen produced at low, high and control temperatures was studied in a separate experiment.

Experimental: This experiment was carried out with 48 plants of the Schwarz 21 cultivar, grown at 24°C, 33°C and as a control in a greenhouse. From each treatment 8 plants were kept unsprayed as controls. The other 8 plants were sprayed twice with 200 p.p.m. H_3BO_3 to which a wetting agent was added. The spray was applied by means of an ordinary continuous spray apparatus. Plants were sprayed for the first time on 24/7/61 and the second time on 3/8/61. The pollen was collected for investigation 7 and 8 days respectively after the second spray had taken place.

Pollen was germinated in the culture medium usually applied. In the second instance, where pollen was sampled 8 days after the second spray had taken place, 50 p.p.m. H_3BO_3 was added to the germination medium.

Results and discussions: Considering the results presented in table 21, it is evident that the boron sprays had no effect on the pollen from plants grown at 24°C and from plants grown as the control. Furthermore, table 21 gives the impression as if the pollen from plants grown at 33°C improved slightly because the germination percentage was 1.0 with a final pollen tube length of 166 μ for the untreated, in comparison to 4% germination and a maximum pollen tube length of 448 μ for the sprayed plants.

TABLE 21 The germination and pollen tube growth (μ) of plants sprayed twice with 200 p.p.m. H_3BO_3 in comparison with that of unsprayed plants. (Germination temp. 30°C. Standard germination medium without H_3BO_3)

Intervals in min.	Plants grown at 24°C				Plants grown at 33°C				Greenhouse control			
	Treated		Untreated		Treated		Untreated		Treated		Untreated	
	G*	L**	G	L	G	L	G	L	G	L	G	L
30		232		249		166		83		332		249
60		547		498		315		166		614		531
90	13	647	16	680	4	448	1	166	24	846	26	796

* G–Germination %
** L–Length

The next day this presumed improved germination and tube growth of the pollen from plants grown at 33°C was followed up with H_3BO_3 in the germination medium. The results, as can be seen in table 22, show no differences between the sprayed and unsprayed treatments; even when borium was added to the germination medium no differences could be observed. Regarding the germination percentages obtained for the sprayed and unsprayed treatments, *viz.* 1 and 1% to 2 and 1% respectively, the unsprayed plants even gave slightly better results (table 22). The final pollen tube lengths obtained were 66 μ and 166 μ for the sprayed plants in a germination medium without and with H_3BO_3, and 182 μ and 149 μ respectively for the sprayed plants in a germination medium without and with H_3BO_3.

The deductions to be made from the foregoing results may be summarized and discussed as follows:

(1) The better pollen germination and pollen tube growth obtained in table 21 in case of the borium sprayed plants grown at 33°C can be regarded as ostensible.

(2) This experiment indicated that borium sprays could not induce a higher activity to pollen when retarded by a high growth temperature.

(3) Although the pollen from plants grown at 24°C and in some instances the pollen from plants grown at the control responds to borium in the germination medium, no such response could be obtained by spraying the flowers and leaves of the plants. This indicates that there must most probably be present sufficient borium or a

TABLE 22 The response of pollen from H_3BO_3-sprayed and unsprayed plants grown at 33°C, when 50 p.p.m. H_3BO_3 was added and omitted from the germination medium. (Pollen tube lengths in μ)

Intervals in min.	Spray treated				Unsprayed			
	Germination medium							
	—B		+B		—B		+B	
	G*	L**	G	L	G	L	G	L
30		66		83		83		83
60		66		166		182		149
90	1	66	1	166	2	182	1	149

* G – Germination %
** L – Length

complex of borium or a substance with the same effect of borium in the pollen or tissues surrounding the pollen of the plants grown at 24°C and at the control.
(4) The high growth temperature (33°C) may restrict the translocation of borium or borium complex from the vegetative parts to the flowering parts of the plant, for no response could be obtained either by spraying the plants or by adding borium to the germination medium.
(5) A substance such as borium or with the same effect of borium is primarily necessary for the production of active pollen, *i.e.* pollen which will give a high germination percentage as well as long pollen tubes. If this substance is absent or present in too low a concentration in the pollen and surrounding tissue, germination but especially pollen tube growth will be retarded or even be nil. Therefore when the 'substance' is present in too low a concentration, pollen cannot be activated by adding borium to the germination medium as with pollen produced at 33°C.

According to the foregoing hypothesis there may be sufficient of the above 'substance' present in the pollen of the control plants to ensure pollen tube growth to a length of 1660 μ (Control plants fig. 15, germination temperature 30°C). Further growth may then be stimulated by the presence of the 'substance' in the style. For the same reason, the pollen from plants grown at 24°C will grow to a length of 581 μ and will then be stimulated to further growth by the presence of the 'substance' in the style. As to the tube growth of the pollen produced at 33°C, one can see that the activity of the pollen was exceedingly low, the final pollen tube length was only 166 μ, and no further growth could be induced by adding borium to the germination medium. One cannot assume, therefore, that if the 'substance' is present in the style, it will induce further pollen tube growth, irrespective of other factors which are necessary for favourable growth.

4.5. DIFFERENCES BETWEEN CULTIVARS

In all previous experiments concerning pollen, only the main cultivar, Schwarz 21,

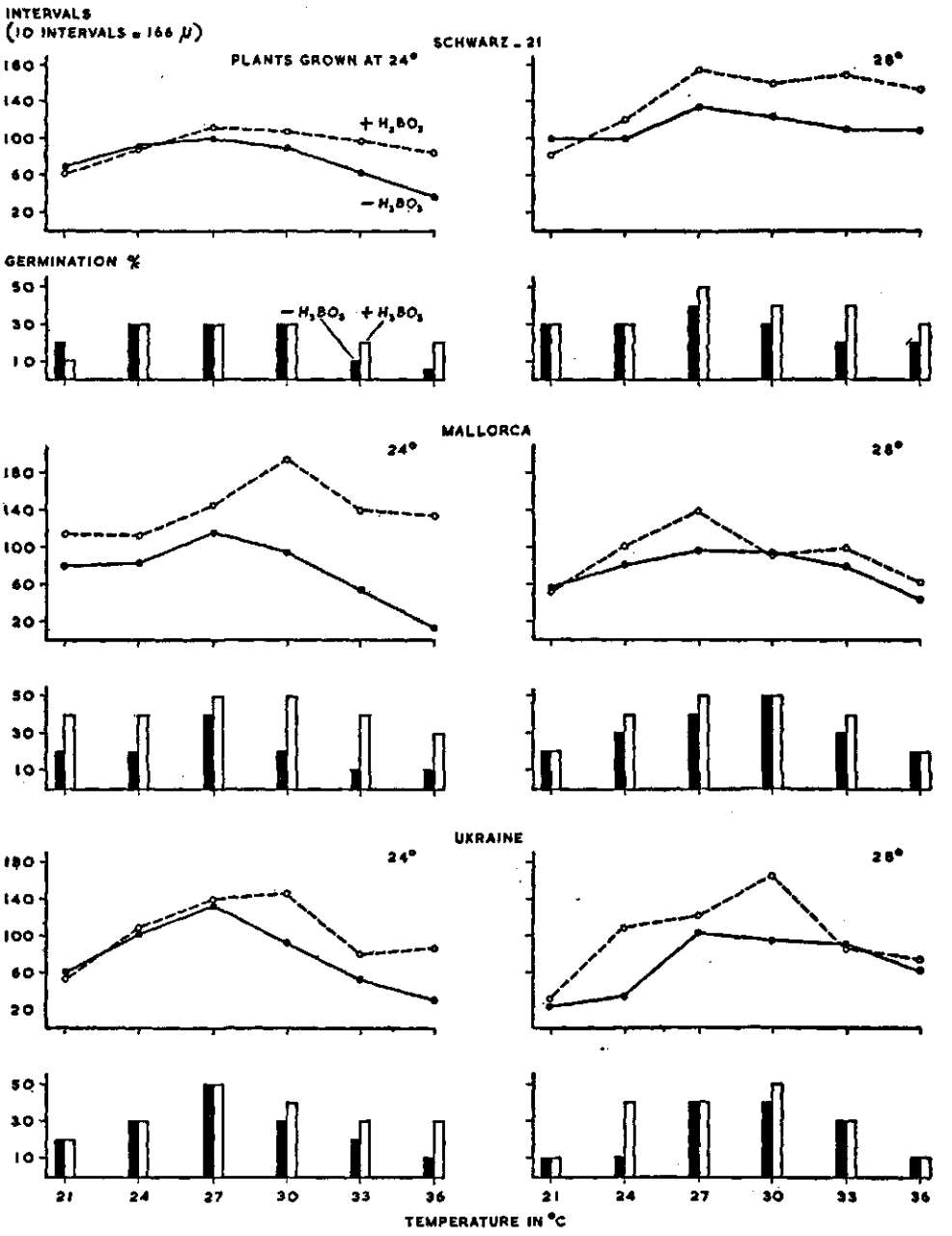
INTERVALS
(10 INTERVALS = 166 μ)
SCHWARZ - 21
PLANTS GROWN AT 24°
+ H3BO3
- H3BO3
28°
GERMINATION %
- H3BO3
+ H3BO3
MALLORCA
24°
28°
UKRAINE
24°
28°
160
140
100
60
20
50
30
10
21
24
27
30
33
36
TEMPERATURE IN °C

Fig. 17

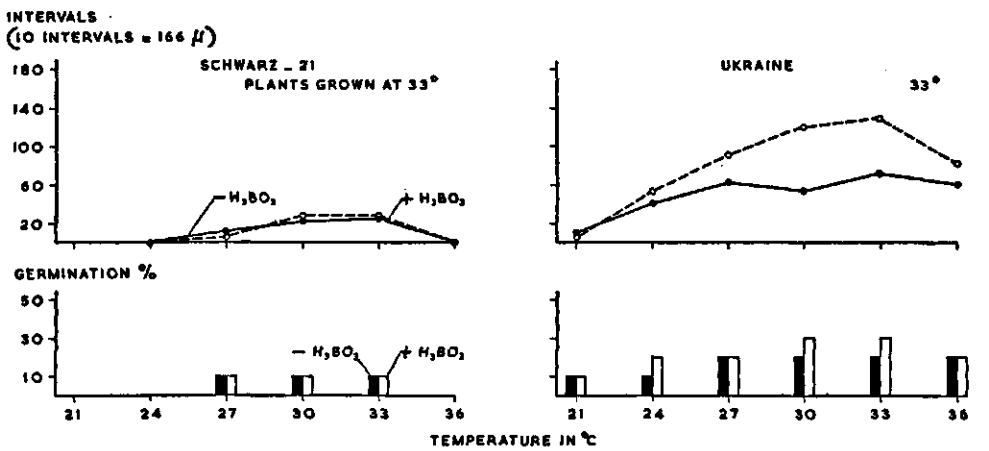

Fig. 17. A comparison between viability of pollen originating from three different cultivars, grown at three constant temperatures, with and without H_3BO_3 (50 p.p.m.) in the germination medium. (Germination percentages and pollen tube lengths determined after 90 and 150 minutes respectively).

was employed. The results obtained explicitly suggest that the viability of pollen was retarded when plants were grown at a temperature as high as 33°C. In order to determine whether the influence of temperature is applicable to other cultivars, a comparative study was conducted with the already described Ukraine-, Mallorca- and Schwarz 21 cultivars.

Experimental: Plants of the Schwarz 21-, Ukraine- and Mallorca cultivars were grown at constant temperatures of 24°, 28° and 33°C. As three cultivars were employed, 96 separate germination determinations were carried out. The comprehensiveness of the experiment made it peremptory to divide the germination percentages into 5 main divisions. Germination percentages between 1 and 10, 11 and 20, 21 and 30, 31 and 40, and 41 and above respectively were given one value each.

Germination percentages and pollen tube lengths were recorded both in the absence and presence of H_3BO_3 in the germination medium.

Results and discussions: The results are presented in fig. 17.

(1) Although the effect of high temperature was more pronounced in the Schwarz 21 and Mallorca cultivars, they were influenced in different ways. At a growth temperature of 33°C, the Mallorca cultivar did not produce sufficient flowers to conduct any comparative pollen viability tests. As in the case of previous tests, the Schwarz 21 cultivar yielded pollen of low viability when plants were grown at 33°C.

Of the three cultivars, however, the Ukraine was least affected by temperature, although a growth temperature of 33°C yielded less viable pollen than when the plants were grown either at 24° or 28°C. The harmful influence of the high temperature on the Ukraine pollen was more or less eliminated by an addition of H_3BO_3 to the

germination medium. This is in contrast to the behaviour of the Schwarz 21 pollen, produced at 33°C, where borium had no effect.
(2) When borium was omitted from the germination medium, all three cultivars at the low temperature yielded the lowest viability values at the highest germination temperature (36°C).
(3) The stimulative effect of boric acid was negligible at a germination temperature as low as 21°C, whereas it had a pronounced effect at the higher germination temperature and reached an optimum at about 30°C.
(4) Although not well defined, a positive correlation seems to exist between germination and pollen tube growth.
(5) The Mallorca cultivar produced its highest percentage of viable pollen at a plant growth temperature of 24°C, whereas the Schwarz 21 as well as Ukraine cultivars produced their highest percentage of viable pollen at 28°C.

Also in this case it was once again proved that the optimum temperature lies definitely lower for the Mallorca cultivar than for the other two cultivars. The same results were obtained where the influence of temperature was investigated as regards vegetative development.

The results suggest that the Ukraine cultivar can produce a higher percentage of viable pollen over a wider range of temperatures than the two other cultivars.

At this stage it appears important to mention that, although the Schwarz 21 cultivar generally produced pollen of low viability at 33°C, approximately one flower out of 25 produced viable pollen at this temperature.

4.6. Summary

The viability of pollen originating from plants grown at constant temperatures of 24°, 28°, 33°C as well as in a greenhouse, was studied. Pollen germination and growth studies were performed in media containing sucrose, agar and water, to which in some instances boric acid was added. All these studies were conducted in cabinets at temperatures ranging from 21° to 36°C.

Plant growth temperature was found to exert great influence on pollen viability. At a growth temperature as high as 33°C, the viability of pollen was much lower than at the other growth temperatures.

Addition of boron to the germination medium was usually found to stimulate both germination and pollen tube elongation. When pollen viability dropped below a certain level, however, the stimulating effect of boron was found to be negligible. While an addition of boron to the germination medium had definite germinating and growth promoting properties, the same effect was not obtainable by boric acid plant sprays.

Sampling date as well as the time of day when pollen was collected had a

distinct effect on pollen viability. In all cases it was found that the viability of pollen was more or less impaired after an illumination period of 10 hours. The higher the temperature during illumination, the quicker decreased the pollen viability.

Growth temperature during the vegetative phase had no influence on the behaviour of pollen, nor was the character of pollen seriously influenced by temperature during the day of flowering. Further results indicated, however, that the behaviour of pollen is mainly dependent on the temperature 36 to 96 hours preceding the actual opening of the flowers.

By studying the effect of growth temperature on pollen shed, the results brought to light that almost 4 times as many pollen grains were set free at 24°C and at the control than at 33°C.

The Ukraine cultivar was least affected by growth temperatures, although less viable pollen was produced at 33°C than at both 24° and 28°C.

V GROWTH AND TRANSPORT OF CARBOHYDRATES AS INFLUENCED BY TEMPERATURE

5.1. Introduction

Although the movement of materials in plants has been studied for the past 200 years (23), many questions still remain unanswered. As the way in which plants transport material is rather complex, it is not surprising that so many contradictory hypotheses have been put forward.

By means of C^{14} labelled compounds such as $C^{14}O_2$ (68, 86), 2,4-D (21, 22) and other labelled compounds such as P^{32} (105) there is increasing evidence that transport of assimilates, labelled indicators, hormones and viruses take place via the phloem (23). 'Thus the sieve tube system in plants may be considered as constituting a highly specialized system of conduits that serve as a functional part of the symplast continuum' (23). It may further be assumed that transport of carbohydrates usually takes place in the form of sucrose (109, 86).

The environment and especially the temperature may be considered to play an important part in the rate of translocation of food materials. Taking the groundnut as an example, our previous experiments indicate that stem growth is more energetic and the dry weights of the aerial parts greater at high than at low temperatures (figs. 2, 3, 4 and table 11). From some other results, it was also indicative that although a vigorous vegetative growth took place at 33°C, no or few fruits developed as compared with plants grown at 24° and 28° (table 11). Hence it might be assumed that at the high temperature a greater proportion of the food material was transported from the supplying regions of the plant to the growing points of the stems than to utilizing regions such as the fruits. Consequently there may be some form of competition between vegetative growth and fruit growth, thus pointing to the fact that the lower activity of pollen, produced at 33°C, is not the only limiting factor in fruit development. It is possible that energy is not evenly distributed throughout the plant at high temperatures, or that translocation decreases at high temperatures due to the fact that respiration is accelerated, thus depleting the reserves of the plant resulting in less carbohydrates being available for transport.

Went and Engelsburg (99), after analysing the leaves, stems and roots of tomato plants held at 8°, 17° and 26°C in the dark for 12 hours, found sucrose in the leaves and roots to be higher in the 8°C plants than in the 17°C plants. Also Went and Hull (101) reported that after chilling tomato stems, more sugar was observed in the roots of the chilled than in those of the unchilled treatment. By using C^{14} labelled glucose and sucrose, Hull (37) furthermore reported an inhibition of transport at

high temperatures. The results of the above mentioned workers suggest a distinct negative temperature coefficient for translocation. On the other hand, workers such as HEWITT and CURTIS (34), SWANSON and BÖHNING (84), VERNON and ARONOFF (94), KENDALL (45) and SWANSON and WHITNEY (85) demonstrated that translocation is hampered by low and accelerated by high temperatures. If the theory of WENT (97) is correct, one expects in spite of a greater rate of respiration, a higher sugar content in the leaves at high than at low temperatures. This was found by WENT (97) in his experiments and he concluded that less sugar was translocated at high than at low temperatures.

For normal fruit development it may be expected that the energy status of the plant will be at a high level, in other words, sufficient quantities of carbohydrates should be present, not only in the vegetative but also in the reproductive growing regions of the plant. If high temperatures cause a low energy status in the plant and a competition between vegetative and fruit growth arises, part of our results, *i.e.* the obtaining of low fruit yields at 33°C would be explainable.

As to obtain more information on the distribution of carbohydrates, top – root ratios as well as sugar analyses of leaves were determined at different temperatures.

5.2. TOP – ROOT RATIOS

Experimental: The first part of the experiment aimed at determining the top – root ratios of the Schwarz 21 cultivar when grown at constant temperatures of 24°, 28° and 33°C. Plants were grown in Mitscherlich pots filled with glass-sand to which a modified HEWITT nutrient solution (33) was supplied twice daily. The character of the glass-sand was such that accurate weights could be obtained due to a minimum loss of roots. After harvesting, both the roots and top parts were oven-dried and their weights determined.

The first group of plants was harvested 19 days and the second group 27 days after planting. Due to a defect which occurred in the ventilating system of one of the temperature rooms, the experiment had to be terminated at an early stage.

Results and discussions: The data summarized in table 23 clearly show that the higher the temperature, the higher the top – root ratio. In other words, the top or sprout development was favoured at the high temperature. The top – root ratios obtained when harvesting took place 27 days after planting, were higher than when harvested 19 days after planting. The earlier the harvest, the smaller the top – root ratio. This result was anticipated, however, because the greater part of the energy during the early phases of vegetative development is supplied by the cotyledons. It is well known that there is a striking difference between the initial growth of the root and that of the shoot. After germination the root grows very rapidly. According to BOUFFIL (12) it reaches a mean length of 46 mm in 4 days. YARBROUGH (107) also shows that the

hypocotyledonary axis grows to a length of 146 mm in 5½ days, whereas the entire epicotyl does not exceed 20 mm, which is only slightly longer than the cotyledons themselves.

Whereas root growth is more energetic during the early stages of development, shoot growth surpasses it during the later stages. This result is clearly demonstrated in table 23, where the top – root ratios obtained 19 and 27 days after planting are compared.

TABLE 23 Top-root ratios of plants grown at three constant temperatures

Days after planting	24°C	28°C	33°C
19	2.22	2.38	2.87
27	3.72	3.78	4.11

Moreover, the tap roots of the plants at 24° and 28°C were longer than at 33°C, although actual comparisons were impossible due to the fact that the plants were grown in pots. It was observed, however, that some of the roots at 24° and 28°C even grew through the drainage opening of the Mitscherlich pots. This was not observed at 33°C.

By studying the effect of temperature on the loss of dry matter and carbohydrates with bean, milkweed and tomato plants, HEWITT and CURTIS (34) found that respirational losses of dry matter and carbohydrates increase with temperature.

Our results demonstrate that a greater proportion of photosynthates was used in the growth of roots at the low than at the high temperature. We may, therefore, assume that if the speed of translocation is higher at the high temperatures, less reserve material was available for transportation at the high temperature, due to a depletion of carbohydrates. When the energy level of the plant is low, and the growth temperature relatively high, it stands to reason that growth furthest removed from the photosynthesing cells will be affected most. Before sufficient energy can reach the roots, most of it is already used up in respiration and top growth, which may explain a higher top – root ratio at the high than at the low temperature.

It may be considered as rather risky to take solely the top – root ratio as an indication of sugar translocation; therefore a further experiment was conducted, where leaves at different temperatures were analysed for sugar.

5.3. SUGAR CONTENT OF LEAVES

Experimental: Plants of the Schwarz 21 cultivar were grown in a greenhouse for 30 days before they were transferred to 5 different thermostat cabinets. Shortly before

the plants were transferred at 2 p.m., some young fully developed leaves were clipped off to serve as a control. Plants were then divided into 5 groups, each group consisting of 12 plants (4 Mitscherlich pots with 3 plants each). Four groups were given a different nyctotemperature treatment of 12 and 24 hours, while the fifth group was given uninterrupted photoperiods of 12 and 24 hours in addition to the 8 hours sunlight received in the greenhouse.

The different treatments after transference from the greenhouse may be summarised as follows:

(a) 20°C. With dark periods of 12 and 24 hours.
(b) 24°C. With dark periods of 12 and 24 hours.
(c) 28°C. With dark periods of 12 and 24 hours.
(d) 33°C. With dark periods of 12 and 24 hours.
(e) A phototemperature of 28°C with photoperiods of (8 + 12) and (8 + 24) hours.

At the end of each treatment some of the young fully developed leaves were clipped off in order to determine their sugar content.

Before transference from the greenhouse had taken place, the plants were watered and drained off with water having the same temperature as in the temperature treatments, until constant soil temperatures were reached.

All leaf samples were oven-dried at 70°C and afterwards sent to the 'Bedrijfslaboratorium voor Grond- en Gewasonderzoek' at Oosterbeek where the sugar analysis was carried out. Sugar percentages were determined after inversion.

Results and discussions: The average sugar content of the leaves, when sampled in the greenhouse before transference to the various temperature treatments, was 2.9%. After a dark period of 12 hours at 20°C, the sugar content of the leaves dropped to 1.6%, whereas at nyctotemperatures of 24° and 28°C it dropped to 1.5 and 1.4% respectively, which demonstrates a gradual decrease in sugar content as the nyctotemperature was raised from 20° to 28°C. When the temperature was raised from 28° to 33°C, however, a sharp fall in leaf sugar content was observed from 1.4% at 28°C to 0.9% at 33°C.

When plants were left at the different nyctotemperatures for 24 hours, it appeared that the sugar percentages were in all cases almost the same. From sugar percentages of 1.6, 1.5, 1.4 and 0.9 after a nyctoperiod of 12 hours, there was a drop after a nyctoperiod of 24 hours to 1.1, 1.1, 1.0 and 0.8 at temperatures of 20°, 24°, 28° and 33°C respectively. It is thus clear that the bulk of the sugar was translocated during the first 12-hour dark period. It should be noted, however, that the sugar content in leaves kept at 33°C after a 12 hour nyctoperiod was even less than in the leaves held at the lower nyctotemperatures for 24 hours. Our findings are therefore the opposite of those of WENT (97); he, for instance, found that after warm nights in spite of greater respiration, the sugar content of tomato leaves was considerably higher than after cooler nights.

When the temperature was raised from 20° to 28°C during the 12 hour nyctoperiod,

TABLE 24 Sugar analysis of leaves from plants grown in a greenhouse and afterwards transferred to different nycto- and phototemperatures

Temp.°C	Photoperiod (hours)	Nyctoperiod (hours)	Sugar % after inversion
20	8	12	1.6
24	8	12	1.5
28	8	12	1.4
33	8	12	0.9
Greenhouse control	8	—	2.9
20	8	24	1.1
24	8	24	1.1
28	8	24	1.0
33	8	24	0.8
28	20	—	2.0
28	32	—	0.4

the difference in sugar content was 0.2%. This difference may, however, only have reflected a higher respiration rate at the higher temperature, and consequently does not justify the assumption of a higher translocation rate at 28° than at 20°C. However, when comparing the sugar content of the leaves of plants kept in the dark for 12 hours at 28° and 33°C a difference of 0.5% was observed ,which suggests that although translocation of sugars was more or less the same in the temperature range of 20° to 28°C, the translocation was speeded up when the temperature was raised from 28° to 33°C.

Apart from the nyctotemperature treatments, 6 plants were kept at 28°C at photoperiods of 20 hours (8 hours sunlight + 12 hours artificial illumination) and 32 hours (8 hours sunlight + 24 hours artificial illumination). The results shown in table 24 reveal that the sugar content of the leaves drops as the photoperiod is extended. From 2.9% as analysed in the control, the sugar percentages dropped to 2.0 and even to a value as low as 0.4, when the photoperiod was extended to 20 and 32 hours respectively. Considering the complexity of the problem and the fact that this study may be seen as preliminary, the result mentioned above needs further confirmation.

With respect to the other results, the following deductions may be made:

(1) The greatest percentage of sugars will be translocated during the first 12 hour nyctoperiod.

(2) If the sugar content of the leaves at the end of the night mainly reflects the amount translocated during the night (12 hour nyctoperiod), as pointed out by WENT (103), translocation increases as the temperature is raised from 28° to 33°C.

(3) The energy status of the plant, expressed as the sugar percentage of the leaves, will be higher at the low night temperatures than at a night temperature as high as 33°C.

5.4. Relation between growth and carbohydrate transport

Where the top - root ratios were determined, the results indicate that the top – root ratios increased as the temperature was raised. This suggests that a smaller amount of carbohydrates was available for root growth at the high than at the low temperature. Now one might assume that the translocation or transport is less at the high than at the low temperature. The analysis of the leaves reveals, however, that their sugar content was higher at the low than at the high temperature, which implies that translocation as well as respiration must also have been higher at the high than at the low temperature. Translocation may therefore not be considered as being critical, although less carbohydrates were transported from the leaves to the roots at the high temperature. So the smaller transport at the high temperature was most probably due to a quicker depletion of food materials, with the result that less carbohydrates were available for transport, and not to a slower translocation rate. Because of this quick depletion of food materials at the high temperature, the utilizing regions nearest to the photosynthesing cells will be favoured at the expense of those plant systems farthest removed from the energy producing regions. When the groundnut plant is grown at 33°C, the rapid vegetative growth results in a great distance between the energy supplying (the young fully developed leaves) and energy consuming regions *i.e.* fruits and roots. The gynophores are mostly concentrated at the lower nodes and at the beginning of flowering plants grown at 33°C already reached a main stem length of about 25 cm against 11 and 9 cm for plants grown at 24°C and at the control respectively. In some previous experiments it was shown that plants grown at 33°C slackened their vegetative growth and produced gynophores as soon as they were transferred to a temperature of 24°C. When, on the other hand, plants were grown at 24°C, fruits were produced but after being placed at 33°C, vigorous vegetative growth took place while no further or very few gynophores developed. These observations suggest that the assimilates are unevenly distributed throughout the plant at temperatures as low as 24°C or, on the other hand, as high as 33°C. It seems safe to say that in the case of the groundnut plant the balance between vegetative and generative growth shifts to the vegetative side at the high, and to the generative side at the low temperatures.

5.5. Summary

A short review of literature on the translocation of assimilates was given in the introduction.

To obtain more information on the distribution of carbohydrates in the groundnut plant, top – root ratios were determined and leaves of plants grown at different temperatures were analysed for sugar.

Top – root ratios were found to increase as the temperature was raised from 24° to 33°C. A greater proportion of assimilates was therefore used in root growth at the low than at the high temperature.

The results regarding sugar translocation do not justify the assumption of a higher translocation rate at 28° than at 20°C. Translocation increased, however, as the temperature was raised from 28° to 33°C.

When keeping plants at nyctoperiods of 12 and 24 hours, the results indicated that the greatest percentage of sugars was translocated during the first 12 hour nyctoperiod.

The experimental results further suggest that assimilates are unevenly distributed at both low (24°C) and high (33°C) temperatures. A more even distribution may thus be expected between these two extreme temperatures.

VI GENERAL SUMMARY AND CONCLUSIONS

The influence of temperature on growth and development of *Arachis hypogaea* L., cultivars Schwarz 21, Mallorca and Ukraine, was investigated. The majority of the experiments was carried out under artificial conditions and consequently the results and conclusions are only applicable to the conditions under which the plants were cultivated.

Except when stated otherwise, all conclusions are based on the results obtained with the Schwarz 21 cultivar.

Actual germination of the groundnut seed is not seriously influenced by temperature, provided it does not exceed 33°C or fall below 24°C. Nevertheless the rate of germination and further development of the seedlings are favoured at the high temperature, in this case 33°C.

The Mallorca cultivar, which needs a definitely lower temperature for normal growth and development, was more detrimentally influenced by a temperature of 33°C than the other two cultivars. With some exceptions occurring in the Mallorca cultivar, a constant temperature of 33°C led to an increase in vegetative growth, *i.e.* longer stems, more leaves, greater leaf area and consequently a higher dry weight of aerial parts of the plant. Together with the increase in vegetative mass at 33°C, a decrease in pod production was observed, some plants producing no pods, whereas vegetative growth and flowering continued uninterruptedly. At 24°C, however, vegetative growth was retarded, although larger leaves and more pods were produced than at 33°C. Thus, a temperature of 28°C may be considered as an optimum, whereas 24° and 33°C appear to be respectively too low and too high for optimum growth and development.

By altering the temperature from 24° to 33°C and *vice versa* at various stages during the development of the plant, it was shown that vegetative growth and flowering are complements in development; in combination, however, they constitute an opposing factor to fruit development.

The temperature during the vegetative phase of development has very little or no influence on the later generative development, but the rate of flowering and some flower characteristics, *e.g.* length of hypanthium, could be changed by merely altering the temperature during the flowering stage.

Although the majority of the groundnut plants flowered abundantly at 33°C, they produced hardly any pods. By studying various plant characteristics an explanation was sought for this unfruitfulness at the high temperature.

The viability of pollen from plants grown under greenhouse conditions as well as at constant temperatures of 24°, 28° and 33°C was studied in *vitro* at germination

temperatures of 21°, 24°, 27°, 30°, 33° and 36°C. The results obtained suggest:

(a) The viability of pollen is influenced by the date of sampling.
(b) The viability of pollen is influenced by the time of sampling during the day.
(c) Plant growth temperature has a marked effect on the character of pollen; pollen of low viability is produced at a temperature as high as 33°C.
(d) The character of pollen is not seriously influenced by the temperature during the day of flowering, but is influenced by the temperature 36 to 96 hours preceding the opening of the flowers.
(e) While an addition of boron to the germination medium stimulates the growth and germination of pollen, the same effect is not obtainable by spraying the plants.
(f) A substance such as boron or with the same effect as boron is primarily necessary for the production of active pollen. A hypothesis is presented stating that if the 'substance' is absent or present in too low a concentration in the pollen, this pollen cannot even be activated by adding boron to the germination medium.
(g) Smaller quantities of pollen are produced or set free at 33°C than at both 24° and 28°C.

The analysis of the leaves as well as the top – root ratio determinations suggest an uneven distribution of assimilates at 33°C, with the result that the balance between vegetative and generative growth shifts to the vegetative side.

In comparison to temperatures of 24° and 28°C it was found at 33°C that:

(1) The hypanthia are longer and the distance the pollen tube has to travel in order to effect fertilization is therefore also longer, in spite of the fact that pollen viability is lower.
(2) The lifespan of the flowers is shorter.
(3) The balance between vegetative and generative growth shifts to the vegetative side, which also explains why certain ovaries can remain dormant in their inflorescence.

We are of the opinion, therefore, that a combination of the above factors constitute the major cause for the unfruitfulness of the groundnut at the high temperature.

ALGEMENE SAMENVATTING EN CONCLUSIES

Een onderzoek werd ingesteld naar de invloed van de temperatuur op de groei en ontwikkeling van *Arachis hypogaea* L., cultivars Schwarz 21, Mallorca en Ukraine.

De proeven werden hoofdzakelijk gedaan onder kunstmatige voorwaarden en de resultaten en gevolgtrekkingen gelden dus slechts voor de omstandigheden, waaronder de planten gekweekt werden.

Tenzij anders vermeld, zijn alle conclusies gebaseerd op de uitkomsten bij de cultivar Schwarz 21.

Op zichzelf wordt de kieming niet sterk beïnvloed door de temperatuur, mits deze niet hoger dan 33°C en niet lager dan 24°C ligt. De snelheid van de kieming en de verdere ontwikkeling worden bevorderd bij de hoge temperatuur, in dit geval 33°C.

De Mallorca cultivar, die een duidelijk lagere temperatuur nodig heeft voor normale groei en ontwikkeling, had meer nadeel van een temperatuur van 33°C dan de twee overige cultivars. Met uitzondering van enkele gevallen bij de Mallorca cultivar, blijkt een constante temperatuur van 33°C een toename van vegetatieve groei als gevolg te hebben, dus langere stengels, meer bladeren, een groter bladoppervlak en dientengevolge ook een hoger drooggewicht van de bovengrondse delen van de plant. Tegelijk met deze toename van het vegetatieve gedeelte werd een afname geconstateerd in het aantal geproduceerde peulen, terwijl sommige planten zelfs bij onafgebroken vegetatieve groei en bloei in het geheel geen peulen opbrachten. Bij 24°C, daarentegen, werd de vegetatieve groei geremd, ofschoon grotere bladeren en meer peulen ontstonden dan bij 33°C. Hieruit blijkt dat een temperatuur van 28°C mag worden beschouwd als een optimum, terwijl 24°C en 33°C respectievelijk te laag of te hoog zijn voor normale groei en ontwikkeling.

Door wisselende temperaturen van 24° naar 33° en omgekeerd toe te passen tijdens verschillende perioden van de ontwikkeling van de planten, kon overtuigend worden aangetoond dat de vegetatieve groei en de bloei mogen worden beschouwd als elkaar aanvullend in de ontwikkeling, terwijl deze beide fasen tezamen een tegengestelde factor betekenen ten aanzien van de vruchtzetting. De temperatuur gedurende de vegetatieve fase heeft geen of zeer weinig invloed op de daaropvolgende generatieve ontwikkeling, maar het tempo van bloei en enkele eigenschappen van de bloem, zoals de lengte van het hypanthium bleken alleen al van temperatuurswisseling tijdens de bloeiperiode afhankelijk te zijn.

Ofschoon het merendeel van de aardnotenplanten overvloedig bloeide bij 33°C, had er vrijwel geen vruchtzetting plaats. Door bestudering van verschillende eigenschappen van de plant werd getracht een verklaring te vinden voor deze onvruchtbaarheid bij de hoge temperatuur.

De levensvatbaarheid van het stuifmeel van planten gekweekt onder kas-omstandigheden en daarnaast bij constante temperaturen van 24, 28 en 33°C, werd bestudeerd door kiemproeven in *vitro* bij 21, 24, 27, 30, 33 en 36°C.

Uit de resultaten hiervan mogen wij het volgende concluderen:

a) De levensvatbaarheid van stuifmeel wordt beïnvloed door de dag van verzamelen.
b) De levensvatbaarheid van stuifmeel wordt beïnvloed door het tijdstip van verzamelen gedurende de dag.
c) De temperatuur heersend gedurende de groei van de plant is van duidelijke invloed op de eigenschappen van het stuifmeel: stuifmeel van lage kiemkracht wordt geproduceerd bij de hoge temperatuur van 33°C.
d) De eigenschappen van het pollen worden niet aanmerkelijk beïnvloed door de temperatuur ten tijde van de bloei, maar wèl door de heersende temperatuur gedurende 36–96 uur vóórdat de bloemen opengaan.
e) Terwijl een toevoeging van borium aan het kiemsubstraat groei en kieming van stuifmeel stimuleert, wordt dit effect niet bereikt door middel van het besproeien der planten met borium.
f) Een stof als borium of met een gelijke werking als borium is absoluut noodzakelijk voor ontwikkeling van levensvatbaar stuifmeel. Als hypothese wordt gesteld dat, wanneer deze 'substantie' afwezig is, hetzij aanwezig in een te lage concentratie in het stuifmeel, toevoeging van borium aan het kiemsubstraat geen stimulans tot kieming bij dit stuifmeel kan teweeg brengen.
g) Bij een temperatuur van 33°C wordt een kleinere hoeveelheid stuifmeel gevormd, respectievelijk los gelaten, dan bij één der temperaturen 24°C of 28°C.

Zowel bladanalyses als bepalingen van de spruit/wortel verhouding leiden tot de veronderstelling dat bij 33°C een ongelijke verdeling van assimilaten plaats heeft, met het gevolg dat het evenwicht tussen vegetatieve en generatieve ontwikkeling een verschuiving ondervindt naar de vegetatieve zijde.

De resultaten verkregen bij 33°C in vergelijking met die bij 24°C en 28°C, kunnen als volgt worden geformuleerd:

1) De hypanthia zijn langer en de afstand, welke de stuifmeelbuis heeft af te leggen teneinde bevruchting te kunnen bewerkstelligen is dus ook langer en de levensvatbaarheid van het stuifmeel is geringer.
2) De levensduur van de bloemen is korter.
3) Het evenwicht tussen de vegetatieve en generatieve ontwikkeling verschuift naar de vegetatieve zijde: hiermede kan eveneens verklaard worden, waarom een aantal zaadknoppen in de bloeiwijze in blijvend ruststadium wordt aangetroffen.

De combinatie van bovengenoemde factoren is waarschijnlijk de voornaamste oorzaak van de slechte vruchtzetting van de aardnoot bij de hoge temperatuur.

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