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HEAT AVOIDANCE A NEW APPROACH FOR BREEDING FOR HEAT RESISTANCE

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EXECUTIVE SUMMARY

Breeding for resistance to an environmental stress in general and to heat stress in particular has been one of the most difficult problems faced by plant breeders While progress in heat resistance for yield and quality has been made in a few crops, overall progress has been slow and limited The primary cause has been the inability to identify efficient, reliable and rapid selection criteria for heat resistance Until such criteria can be identified, breeding for heat resistance will be extremely difficult and effectively a serendipitous exercise

In this program we are examining a new approach, heat avoidance, for improved heat resistance As a model we are using two crops important to Kazakhstan and Israel, tomatoes and wheat The traits examined include those which allow a plant to avoid heat stress by (a) reducing the amount of energy absorbed by solar radiation, and/or (b) by increasing cooling efficiency The traits include stomatal conductance, density of stomata and leaf hairs, leaf area and architecture, plant architecture and reflection of solar radiation

In Kazakhstan the research in the second year focused on examining the genetics of leaf rolling in selected wheat varieties They identified leaf rolling as a wheat leaf trait that has been associated with their more heat and drought resistant cultivars They concluded that leaf rolling may potentially be a useful selection criterion for these stresses They produced a series of monosomic lines for leaf rolling and examined them for genetic relationships and for testing for heat and drought resistance

The Israelis in the second year focused their program on tomatoes From their original program they selected two cultivars, 'd' and 'g', the former having few if any heat avoidance traits and the latter many These include (a) high stomatal conductance, (b) a plant architecture that increases shading of underleaves, and (c) changes in leaf architecture that reduces the amount of solar radiation absorbed by the leaves and/or increases the water efficiency for leaf cooling either by increasing humidity or by allowing the same drop of water to be evaporated several times in the funnel that is produced by leaf curling They examined the two parental lines plus reciprocal F1 and F2 generations Their results further confirm the potential of this approach

Collaboration in the second year included visits to the other country, exchange of scientists for research, exchange of germplasm and joint analysis of the results

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SECTION I

A RESEARCH OBJECTIVES

1 to determine the potential of an array of physiological and morphological heat avoidance characters as rapid and reliable selection criteria for future breeding programs for heat resistance,

2 to begin to determine the genetic control of the more promising heat avoidance characters

B RESEARCH ACCOMPLISHMENTS

1 GENERAL INTRODUCTION

The continuous improvement of yield and product quality in most crops to meet the increasing demand of the world's population is an important, if not central, goal for agricultural researchers An important limiting factor in maximizing yield and quality is the relatively poor resistance of present cultivars to heat stress Plants grown under high stress inducing temperatures usually have significantly reduced yields and/or quality then when grown under more optimum temperatures Thus while breeding programs have lead to dramatic increases in yield/quality in almost all crops when grown under environmentally non-stressed conditions, they have usually failed to improve heat resistance This is primarily due to two reasons

1 the absence of rapid and reliable selection criteria - the large number of studies that have been conducted to improve heat resistance, whose two components are heat tolerance (the ability of metabolism and structures to function normally or nearly so under high temperatures) and heat avoidance (the ability of a plant to prevent the buildup of high plant temperatures when grown at high temperatures), have mostly concentrated on heat tolerance characters and very little on heat avoidance characters (Blum, 1988, Shapelier and Blum, 1986, Zeng and Khan, 1984) While these studies have added to our knowledge of the effects of high temperatures on plant physiology and biochemistry, they have not provided breeders with any reliable selection criterion,

2 little information on the genetic control of heat resistance characters - while some information is known for a few characters, it is still not sufficient for a breeder

to use *a priori* for a specific character (Zeng and Khan, 1984, Lu and Zeiger, 1994, Schaff, 1985, Wessel-Beaver and Scott, 1992, Abdul-Baki and Stommal, 1995)

Heat avoidance as an alternative approach to heat tolerance for improved heat resistance has recently been proposed (Lu and Zeiger, 1994, Radin et al, 1994) Two characters, increased stomatal conductance and reduced leaf size were suggested The Israeli team examined these characters in leaf and plant architecture in a historic collection of five elite cultivars of tomatoes, one from each decade from 1940-1980, which were bred for improved quality, yield and disease resistance under the high temperatures of central California Their results closely followed those expected from this hypothesis The Kazakh team did a similar study in wheat selected under high temperatures and drought and concluded that plant and leaf morphology can play an important role in stress resistance

This report is the result of a joint effort by both teams to examine heat avoidance traits in two crops which are important to both countries, tomatoes and wheat Due to logistical and financial limitations, it was decided that in the second year the Israeli team would concentrate on tomatoes and the Kazakh team on wheat

2 *KAZAKHSTAN*

Abstract

Our investigations revealed that the rolling leaves adaptive trait is controlled by the RL1 and RL2 dominant genes which are localized in chromosomes 6A and 4D, respectively We have started the introduction of RL1 and RL2 genes into cultivars of different ecotypes cultivated in Kazakhstan to create both new cultivars with additional adaptive traits as well as a series of isosomic lines Isosomic lines are valuable for breeding, genetics, physiology, biochemistry and biotechnology research

Results and Discussion

Water regime and photosynthetic activity of isosomic lines and respective parental cultivars

Expression of wheat genotypic peculiarities is achieved, first of all, by changes in the intensity of water turnover which to a high degree determines the total plant

metabolism We accomplished a comparative study of the water regime of isosomic lines carrying RL1 and RL2 genes and parental wheat cultivars We compared the water status parameters – total water content and its fraction composition, transpiration rate and leaf water-retaining capacity

Table 1 shows the ratio of water fractions in the leaves of winter wheat during the vegetative period The water content varied substantially during the whole vegetative season Reduction in water content of wheat tissues caused by high temperatures was followed by certain changes in the ratio of water fractions The Krasnovodopadskaya isosomic line did not differ from the control in total water content, but was superior to the control in bound water content at tillering, ear formation and ear formation-anthesis This suggests a higher ability of the isosomic line for adaptive response

Isosomic lines of the Omskaja 9 and Sibirjachka 8 cultivars had slightly lower content of both free and bound water The data points to the fact that if the amount of free water determines the intensity of physiological processes, the bound water quantity is responsible for plant resistance against unfavorable environmental conditions Transpiration rates of isosomic lines and controls were different (Table 2) Daily transpiration rate was more variable in the controls with an increase of water loss for transpiration at noon and a steep decrease at 18 00 Isosomic lines had lower transpiration rate regardless of the measurement time Water-retaining capacity is an important trait that allows the determination of the range of adaptive plasticity and estimate plant resistance Water-retaining capacity was determined by evaporation and the content of retaining water at dehiscence of leaves separated from plants The measurements were taken at intervals (15-180 min) and the water-retaining capacity was calculated as percentage of initial content Noteworthy, the differences in tolerance to dehydration were seen within 15 min, suggesting different initial leaf water-retaining capacity of the studied wheat lines (Table 3) Leaves of isosomic lines showed high water-retaining capacity at all measurement times at both ear formation and grain maturation The higher amount of bound water, lower variation in total water content, higher water retaining capacity and lower water transpiration rates can be considered as evidence of higher drought resistance of wheat isosomic lines as compared with parental cultivars

Photosynthetic activity and structural organization of the photosynthetic apparatus in isosomic lines and cultivars were determined and compared We determined the following parameters Hill reaction rate, activity of non-cycle electron transport, cycle and non-cycle phosphorylation, degree of energy coupling

in chloroplasts, Rubisco activity, intensity of real photosynthesis by gas exchange and the number of chloroplasts in suspension

Physiological characteristics were determined at main vegetative stages Chloroplasts were obtained from the flag leaf by the conventional procedure Chloroplast photochemical activity was measured using spectrophotometer to determine the rate of electron transport photophosphorylation Ammonium chloride (10 mkM per sample) was used as an uncoupler of electron transport and photophosphorylation For estimation of Rubisco activity we used a radiometric method from fresh leaf extracts The amount of chloroplasts was calculated using Goryaev counting chamber according to Ivanchenko In order to measure photosynthetic intensity we used an Infralit gas analyzer with a CO_2 concentration 0 03% (real photosynthesis), calculated on a unit of dry weight and unit of leaf area Analysis was done at the end of stem formation, transition to the reproductive period, anthesis, grain formation and maturation (Table 4)

The chloroplasts of the Omskaja isosomic line were characterized by a higher rate of electron transport during stem formation compared to the Omskaja control cultivar The addition of a phosphate-acceptor system to the reaction medium which imitates the electron transport in isolated chloroplasts significantly accelerated electron flow in isosomic lines The maximal rate of electron transport in isosomic lines was detected if the phosphate-acceptor system was supplied together with ammonium chloride

The Omskaja isosomic line did not significantly differ from the control cultivar for non-cycle phosphorylation activity at stem formation, but was superior in activity of cycle phosphorylation and total phosphorylation flow Besides, the proportion of cycle phosphorylation to non-cycle one in isosomic line increased by 50% as compared to parental cultivar Although most of the energy is provided in higher plants by non-cycle phosphorylation, cycle phosphorylation is important for ATP synthesis during CO_2 photosynthetic assimilation and suspends high ATP NADP ratio The Omskaja isosomic line possessed high potential activity of chloroplast electron-transport chain to electron transport and higher activity of cycle phosphorylation as compared with Omskaja 9 control

Photosynthetic chloroplast activity of Krasnovodopadskaya isosomic line and its control cultivar Krasnovodopadskaya 210 represented another pattern The Krasnovodopadskaya isoline was inferior to the control cultivar by the electron transport activity and phosphorylation It is plausible that the results were affected by the differences in developmental stages of the isoline and control at the time of measurements the end of stem formation in Krasnovodopadskaya 210 coincided

with ear formation in the isosomic line It is known that the enhancement of primary energetic processes of photosynthesis is closely related to the developmental stage and is expressed in wheat at stem formation before grain formation (transition to reproduction)

It is worth noting that the activity of Rubisco, a key enzyme in photosynthesis, was lower in the Omskaja isosomic line versus control but higher in the Krasnovodopadskaya isosomic line versus its control at stem and ear formation This difference correlated to photosynthetic intensity (integral photosynthetic activity) evaluated by gas exchange

There was a considerable increase in non-cycle phosphorylation activity at spike formation-anthesis and flowering for the Omskaja isoline and increase in cycle phosphorylation for both isolines A higher activity of the total flow of phosphorylation in the isolines was found as compared to controls The intensity of photosynthesis during this period of time was much higher in isosomic lines

At the end of the vegetative period, at the stages of grain formation and maturation, activity of primary energetic and enzymatic processes in chloroplasts of isolines and controls were the same as was the total decrease of the corresponding absolute values Decreased activity of particular photosynthetic reactions led to decreased integral gas exchange and intensity of photosynthesis Determination of photochemical activity of chloroplasts in suspension revealed an increased number of chloroplasts in the Omskaja 9 cultivar as compared with corresponding isolines We can conclude that the Omskaja isoline is characterized by considerable activity of the electron transport chain and photophosphorylation in a single chloroplast (Table 5)

Dry matter accumulation, formation of leaf assimilatory surface, yield parameters of isosomic lines and cultivars

We studied accumulation of biomass and its vegetative/reproductive ratio from anthesis to waxy maturation Potential grain yield and the way of its realization was compared and contrasted between Grecum 476 cultivars and their isosomic lines and a new cultivar carrying the RL1 and RL2 genes determining leaf rolling Grecum 476 had significantly greater leaf area than the other cultivars The same trend was found in isosomic lines (Table 6)

There are some traits which characterize the relationship between a spike and its assimilation apparatus, e g biomass distribution between plant organs before the onset of grain formation and afterwards The spike/tiller ratio during anthesis is an

indicator of genetically determined spike demand A large spike biomass at anthesis (initial mass) was typical for the studied isosomic lines (Table 7)

The highest total biomass at anthesis was Grecum 476, followed by Sibirjachka 8 and Omskaja 9 isosomic lines The last two lines also exceeded parental cultivars for biomass of leaves, shoots and spikes Spike/tiller biomass ratio was higher in Sibirjachka 8 than its parental cultivar

Changes in stem, spike and the whole tiller biomass during grain maturation determine the degree of assimilation stock to spike demand and indirectly characterizes reutilization The period between anthesis and waxy maturation was characterized by a decrease of tiller dry matter in all lines and cultivars studied The biomass of leaves and stems decreased and the spike biomass increased (with an exception of a conspicuous increase in the Sibirjacka 8 isoline)

Analysis of productivity compounds revealed that isosomic lines are superior to parental cultivars in the number of reproductive tillers, spike weight and length and mass of thousand seeds, but have shorter stem lengths (Table 8) which is important for intensive cultivation technology

Table 1 Water regime of winter wheat leaves

Line/ cultivar	Water fraction content, %														
	Tillering			Stem formation			Ear formation			Ear formation-anthesis			Grain maturation		
	total	free	bound	total	free	bound	total	free	bound	total	free	bound	total	free	bound
Krasnovodopadskaja 210	82 34	79 83	2 51	82 53	58 61	23 92	78 25	56 15	22 10	76 14	55 31	20 83	75 1	52 7	22 37
Krasnovodopadskaja isosomic lines	81 12	68 89	12 26	81 08	55 13	25 95	78 16	49 58	28 58	75 52	50 05	25 47	75 45	47 7	27 74
Omskaja 9	80 75	66 98	13 77	76 79	57 97	18 75	77 72	51 10	26 62	74 00	60 71	13 28	73 6	44 0	29 25
Omskaja isosomic lines	82 36	60 80	21 56	78 75	54 15	24 60	75 88	44 21	31 67	76 61	50 52	26 09	73 4	45 5	27 82
Sibirjachka 8	78 00	69 72	8 28	78 50	54 62	23 88	74 78	47 29	27 49	73 68	54 85	18 83	72 7	41 3	31 35
Sibirjachka isosomic lines	80 14	63 39	16 75	78 72	55 50	23 28	74 62	46 37	28 25	75 53	54 11	21 42	73 1	37 7	35 39

Table 2 Intensity of transpiration of winter wheat leaves mg/h/1 g of fresh biomass at ear formation

Line/ cultivar	Hours			
	9 00	13 00	18 00	Mean/day
Grekum 476	493	1088	694	758
Omskaja 9	1528	2021	709	1419
Omskaja isosomic lines	837	1398	701	879
Sibirjachka 8	943	1621	655	1076
Sibirjachka isosomic lines	630	1055	522	736

Table 3 Water-retaining capacity of winter wheat leaves, percent of total

Developmental stage, line	Water loss in % of initial weight							Total water content
minutes	15	30	45	60	90	120	180	
Ear formation								
Grekum 476	12 67	17 82	20 28	23 36	29 22	31 16	48 64	70 68
Omskaja 9	12 50	18 47	21 59	25 00	28 69	32 67	40 49	66 66
Omskaja isosomic lines	13 30	16 41	21 49	23 96	27 20	32 21	39 75	67 87
Sibirjachka 8	13 60	18 18	21 75	24 70	31 45	33 83	44 33	65 48
Sibirjachka isosomic lines	11 19	16 38	19 46	22 13	25 25	29 06	35 09	69 97
Grain filling								
Grekum 476	18 24	21 59	26 99	32 43	43 08	49 22	58 24	67 30
Omskaja 9	22 21	25 24	28 79	35 09	42 79	48 06	54 22	67 72
Omskaja isosomic lines	19 14	22 46	27 27	34 13	39 09	45 88	54 31	65 66
Sibirjachka 8	20 51	23 22	25 70	35 33	35 80	43 99	55 09	64 87
Sibirjachka isosomic lines	18 51	21 05	25 32	29 63	38 39	42 53	52 18	67 99

Table 4 Photosynthetic activity of isosomic lines of bread wheat

Trait	Stem formation		End of stem formation		Ear formation		Anthesis		Grain maturation			
	Omskaja 9		Krasn 210		Omskaja 9		Krasn 210		Omskaja 9		Krasn 210	
	con	iso	con	iso	con	iso	con	iso	con	iso	con	iso
Hill reaction, mkm red $K_3Fe(Cl)_6$/mg * l/h	272	317	310	337	168	390	351	300	317	315	232	257
Electron transport in presence of Phospate-acceptor system	559	611	763	771	889	790	876	833	648	621	589	591
Electron transport uncoupled with photophosphorylation	620	703	948	786	982	841	976	983	778	722	801	891
Rise of electron transport in presence of PAS	287	295	453	434	721	400	527	533	332	306	357	333
Rise of electron transport in presence of PAS & NH_4Cl	348	386	638	449	814	451	624	683	462	407	570	433
Non-cycle phosphorylation mkM inorg P/mg chl per hour	226	220	739	497	356	771	683	500	396	414	1012	595
Cycle phosphorylation	113	220	369	316	267	313	244	375	308	276	253	191
Total cycle + non-cycle phosphorylation, - "-	340	440	1108	814	622	1084	927	875	703	690	1264	786
Non-cycle (% of total)	67	50	67	61	57	71	74	57	56	60	80	76
Cycle (% of total)	33	50	33	39	43	29	26	43	44	40	20	24
Rubisco activity, nm CO_2/mg protein per min	436	345	358	437	-	-	-	-	174	175	167	224
Intensity of photosynthesis, mg CO_2/g dry matter per hour	18	11 6	10 1	11 6	10 8	13 6	10 7	12 4	5 4	6 2	6 4	5 0
Intensity of photosynthesis, mg CO_2/dm^2 dry matter per hour	7 8	4 4	4 0	4 6	6 0	7 2	6 4	7 3	5 2	3 8	3 8	3 3

Table 5 Number of chloroplasts in suspension for isosomic lines

Control/line	Date	Developmental stage	Number of chloroplasts
Omskaja 9	26 05	stem formation	18,840,000
Omskaja isosomic lines	26 05	stem formation	1,540,000
Krasnovodopadskaja 210	26 05	end of stem formation	1,016,000
Krasnovodopadskaja isosomic lines	26 05	start of ear formation	1,100,000
Omskaja 9	09 06	ear formation	19,600,000
Omskaja isosomic lines	09 06	ear formation	1,852,670
Krasnovodopadskaja 210	09 06	anthesis	863,330
Krasnovodopadskaja isosomic lines	09 06	anthesis	863,330
Omskaja 9	23 06	grain formation	1,628,670
Omskaja isosomic lines	23 06	grain formation	1,372,000
Krasnovodopadskaja 210	23 06	grain formation	1,866,670
Krasnovodopadskaja isosomic lines	23 06	grain formation	1,255,330

Table 6 Assimilative surface of leaves (cm^2), ear formation-anthesis

Control/line	Flag leaf	Penultimate leaf	Total
Grekum 476	32 7	33 3	66 0
Omskaja 9	20 2	21 4	41 6
Omskaja isosomic lines	33 7	32 3	66 0
Sibirjachka 8	20 8	20 4	41 2
Sibirjachka isosomic lines	27 3	27 0	54 3
Krasnovodopadskaja 210	22 4	22 1	44 5
Krasnovodopadskaja isosomic lines	25 6	29 3	55 0

Table 7A The structure of main tiller dry matter at the anthesis stage

Control/line	Dry matter, g				Organ fraction in the total tiller mass, %		
	whole tiller	leaves	spike	straw	leaves	straw	spike
Grekum 476	4 79	0 39	1 54	2 86	8 14	59 71	32 15
Omskaja 9	3 26	0 21	0 79	2 26	6 14	69 33	24 33
Omskaja isosomic lines	4 17	0 48	0 99	2 70	11 51	64 75	23 74
Sibirjachka 8	3 14	0 27	0 64	2 23	8 60	71 02	20 38
Sibirjachka isosomic lines	4 41	0 37	1 06	2 98	8 39	67 57	24 04

Table 7B Dry matter in different organs at three developing stages

Control/line	Dry matter, g								
	leaves			straw			spike		
	anthesis	grain formation	waxy maturation	anthesis	grain formation	waxy maturation	anthesis	grain formation	waxy maturation
Grekum 476	0 39	0 37	0 33	2 86	2 15	2 02	1 54	1 71	2 43
Omskaja 9	0 21	0 41	0 30	2 26	3 06	1 74	0 79	2 00	2 20
Omskaja isosomic lines	0 48	0 45	0 26	2 70	2 51	1 81	0 99	2 14	2 18
Sibirjachka 8	0 27	0 28	0 25	2 23	1 66	1 39	0 64	1 23	1 35
Sibirjachka isosomic lines	0 37	0 36	0 31	2 98	1 67	2 03	1 06	1 64	2 12

Table 8 Productivity traits of parental cultivars and their isosomic lines

Cultivar/line	Productive tillering	Length in cm		Number per spike		1000 seeds weight
		stem	spike	spikelets	seeds	
Grekum 476	10 47±0 27	109 41±1 21	12 94±0 26	16 29±0 24	48 76±1 11	54 40
Omskaja 9	5 76±0 25	117 08±1 68	7 76±0 17	15 36±0 30	29 92±0 84	42 30
Omskaja isosomic lines	9 48±0 99	111 04±0 78	10 56±0 15	16 96±0 25	41 04±1 22	46 80
Krasnovodopadskaja 210	5 15±0 25	99 20±0 63	9 75±0 18	19 80±0 20	49 75±1 33	42 34
Krasnovodopadskaja isosomic lines	6 47±0 31	89 42±0 75	12 47±0 27	20 68±0 36	62 42±1 88	48 80
Sibirjachka 8	4 44±0 24	128 76±0 72	8 08±0 15	15 72±0 23	25 16±0 72	46 10
Sibirjachka isosomic lines	10 32±0 40	117 16±0 81	11 60±0 17	15 52±0 17	29 92±0 84	48 34

3 *ISRAEL*

Introduction

From our preliminary study of the five historic cultivars, we found that stomatal conductance increased and leaf area decreased from the older cultivars to the newer "elite" cultivars, as we had hypothesized for improved heat avoidance In addition we found that the newer cultivars had undergone changes in their leaf and plant morphology which caused a reducing in available surface area for solar absorption In the first year of the program our main tomato experiment was to reexamine in an array of heat avoidance characters the three cultivars which gave the most extreme responses in respect to heat avoidance characters in our preliminary study 'c' (1940), 'd' (1950) and 'g' (1980) In addition, we examined the F1 and F2 generations between 'c' and 'g' (Mendlinger et al 1998)

In the second year of the program, due to major advantages of working with cultivar 'd' over 'c', we shifted over research to comparing 'd' with 'g' We planned two field experiments (1) a nethouse experiment comparing an array of heat avoidance traits in parental cultivars 'd' and 'g' and their reciprocal F1 and F2 crosses - the characters to be examined included stomatal conductance, plant and leaf architecture, and (2) an open field trial of selected F4 generation lines from dxg and gxd crosses Due to sever disease problems involving both viruses spread by whitefly and fusarium, we had to stop the second experiment half way through it

Materials and Methods

Three identical nethouses, each covered with a fine-mesh net which prevented insects from entering, were used in this experiment Each nethouse was sown at a different time of the summer nethouse-1 (NH-1) in late spring, nethouse-2 (NH-2) in early summer and nethouse-3 (NH-3) in late summer Seeds from each line, the two parents and the reciprocal F1s and F2s, were sown in petri dishes, incubated for three days at 25^{0}C and then placed in seedling trays for three weeks After three weeks when the plants had 3,4 true leaves, they were transplanted into 15-liter buckets, one plant per bucket, filled with a commercial potting mixture for horticultural crops Each nethouse was divided into two blocks with 3 plants per 'd', 'g' and reciprocal F1 and 6 plants per reciprocal F2 per block Two minimum/maximum thermometers were placed in each of the nethouses, one at 25cm and the other at 150cm above the ground and the temperatures recorded daily

Fertigation was given via drippers with 2 drippers per bucket The irrigations began with 1 per day and increased with increasing temperature and plant size to 5 per day, each irrigation was 2 liters/bucket The amount of irrigation needed was determined by measuring the outflow of water from 9 randomly chosen buckets Fertilizer was given in every irrigation Our target was a daily outflow of 25% which prevents salt buildup while maintaining water saturation of the potting mixture The plants were periodically examined for diseases and insect infestation and prophylactic fungicide treatment was performed during the growing season

Each plant was examined for an array of characters These included

1 plant architecture - leaf and stem fresh and dry biomass, length of the main stem, number of nodes and side branches (only in the first nethouse)

2 leaf architecture - the amount of leaf curling (the bending of the sides of the leaves upwards and inwards producing a cylinder shape leaf - sometimes called leaf rolling in cereal papers) was scored five times during the growing season using a scale from 1-5 with 1 being no curling and 5 a complete cylinder leaf, leaf rotation (the rotation of a leaf on its stem) with no rotation being scored as 0, 90^0 rotation scored as 1 and 180^0 rotation scored as 2, leaf area, number of leaflets/leaf, leaf and stem biomass,

3 stomatal conductance - stomatal conductance and leaf temperatures were measured on the first fully developed leaf using a Licor 1600 steady state porometer for each plant at five life stages before flowering, onset of flowering, fruit set, fruit filling and fruit ripening These stages were approximately one week apart,

4 fruit yield and quality - for each plant the mean fruit weight, number of fruits per plant and total yield were determined, from 5 fruits/plant the total soluble solids (TSS) were determined

Results and Discussion

The results of the plant architecture traits are presented in Table 1 Cultivar 'd' had significantly more dry leaf biomass than did 'g' but no differences were found for dry stem biomass The F1 and F2 generations had dry leaf biomass closer to parent 'd' than 'g' No significant differences were found in number of nodes or sidebranches It appears that 'd' invested more of its energy into leaf production than did 'g' This difference in leaf biomass, which is a function of more leaves, leaflets/leaf and/or leaf size (see below) superficially may be considered as a heat avoidance mechanism in tomatoes as the more leaf biomass, the more the understory leaves may be shaded This would reduce solar absorption and thus

prevent leaf temperature increases (Comstock and Mahall, 1988, Neufeld et al , 1988, Save et al 1993) However as heat is a serious problem in the summer when plants produce more than sufficient amounts of photosynthates for their fruit needs the increased investment in leaves may not be worthwhile (Mendlinger and Fossen,1993) The larger the leaf biomass, the more water may be lost via stomatal transpiration and increase of water load needed for maintenance This type of shading maybe less efficient and require more water than if the leaf has a mechanism for selfshading, i e curling or rotation

The number of leaflets/leaf and leaf area at three census periods in the three nethouses are presented in Table 2 Over all three nethouses, 'd' had more leaflets/leaf and leaf area than 'g' The F1 and F2 generations did not significantly differ from the parental midpoint (t-test) and were overall equally close to both 'd' and 'g'

Cultivar 'g' had significantly greater curling and rotation than 'd' or the reciprocal F1 and F2 generations (Table 3) Therefore it showed a higher grade of selfshading and a reduction in solar absorption (Mendlinger et al 1998) The F1s and F2s were not significantly different from each other When the results from the F1s and F2s were pooled over the three nethouses, they had significantly less curling and rotation than the parental midpoints (Chi-Square, $p<0\ 01$) A significant positive correlation was found between curling and rotation (Fig 1) Both curling and rolling are important heat avoidance character as both reduce solar absorption (DiBenedetto and Cogliatti, 1990, Neufeld et al , 1988, Young,1985, Heckathorn and DeLucia, 1991) In addition, we found that leaf curling allows for improved and more efficient leaf cooling as a molecule of water cools the leaf when first transpirated from the stomata, is prevented from escaping by condensing in the curled leaf tunnel and then evaporated again, i e the same molecule is used several times for cooling

Cultivar 'g' had significantly higher stomatal conductance than did 'd', the F1 and F2 generations (Table 4) This is similar to the results obtained last year (Mendlinger et al , 1998) Stomatal conductance was found to decrease over time from before flowering to fruit set ($r=-0\ 89^*$, $df=3$) Stomatal conductance is highest at the earliest stages of growth, when the plants need all the photosynthates that they can produce for rapid vegetable growth, flower production and early fruit growth, and lowest at fruit set when there is little need for large amounts of photosynthates in deterministic cultivars

A significant correlation was found in both leaf curling and rotation with stomatal conductance (Fig 2) Significant regressions were found between stomatal

conductance and leaf temperatures when all life stages are pooled (Fig 3) or when all generations are pooled (Fig 4) The regressions were strongest in NH-2 and NH-3 when the plants were grown during the hottest part of the summer When the R2s for the regression of leaf temperature on stomatal conductance in the different generations were compared using a similar sample size (n), we found that both F2 generations were significant higher than the F1s (The R2s for the F2 generations were calculated using the mean of a series of R2s generated from both F2s, n=90)

Total yield did not differ between 'd' and 'g' (Table 5), however, it must be remembered that they were not grown under commercial agromanagement techniques The F1 and F2 generations all had higher yields than did their parents This may indicate a heterosis effect from the two parental homozygous cultivars The F1 and F2 generations also had higher mean fruit weight 'G' had the highest TSS No significant relationship was found between TSS and stomatal conductance or leaf temperature (Fig 5)

Broad sense heritability estimates (h^2) were not significant different between the two reciprocal crosses (Table 6) Most characters had moderate levels of heritability indicating the potential for breeding The relatively high h^2 for curling does not fit the results from Table 3 (see above) We cannot yet explain this discrepancy

Table 1 Vegetative growth characters of the first nethouse, 1998

cultivar	FW (g)	DW leaf (g)	DW stem (g)	length of MS (cm)	No of nodes	No of SB
d	1941	199 b	117	71 8	19 5	7 2
g	1194	107 a	116	66 7	18 8	9 8
dxg	1730	188 b	121	72 5	22 3	11 4
gxd	1629	153 ab	99	68 7	16 3	9 8
dxg F2	1690	176 ab	103	72 9	15 9	9 0
gxd F2	1778	186 b	113	81 5	17 3	9 4
parental midpoint	1567	153	116	69 3	19 2	8 5

Std Dev cultivar	FW	DW leaf	DW stem	length of MS	No of nodes	No of SB
d (n=6)	264	26	18	30 3	6 2	1 5
g (n=6)	107	5	12	7 4	5 5	2 2
dxg (n=6)	233	36	19	13 3	6 0	0 5
gxd (n=6)	506	29	16	14 9	6 3	2 8
dxg F2 (n=24)	580	65	25	17 7	3 3	1 8
gxd F2 (n=24)	478	50	32	17 0	4 7	2 3

Table 2 Leaflets per leaf and leaf area at three physiological plant stages in three nethouses, 1998

Nethouse 1

cultivar	before flowering leaflet/ leaf	before flowering leaf area cm^2	flowering leaflet/ leaf	flowering leaf area cm^2	fruit set leaflet/ leaf	fruit set leaf area cm^2
d	9 7 a	194 a	12 3	124	9 2	116
g	8 7 a	165 a	10 8	126	7 8	79
dxg	10 7 ab	222 ab	11 5	108	9 2	79
gxd	11 8 b	228 ab	13 8	198	8 5	80
dxg F2	11 2 b	237 b	11 2	123	9 7	92
gxd F2	10 1 a	205 a	11 8	145	9 1	96
parental midpoint	9 2	179	11 6	125	8 5	98

Nethouse 2

cultivar	before flowering leaflet/ leaf	before flowering leaf area cm^2	flowering leaflet/ leaf	flowering leaf area cm^2	fruit set leaflet/ leaf	fruit set leaf area cm^2
d	13 0 b	272 c	11 5	155 abc	10 0 bc	144 b
g	10 2 a	182 a	10 5	119 a	8 7 a	101 a
dxg	11 2 ab	238 bc	10 3	128 ab	10 2 c	115 ab
gxd	12 8 b	275 c	9 8	126 ab	9 7 abc	130 ab
dxg F2	12 6 b	242 bc	10 4	176 bc	9 2 ab	141 b
gxd F2	11 4 ab	218 ab	10 5	190 c	9 3 abc	122 ab
parental midpoint	11 6	226	11 0	137	9 4	122

Nethouse 3

cultivar	before flowering leaflet/ leaf	before flowering leaf area cm^2	flowering leaflet/ leaf	flowering leaf area cm^2	fruit set leaflet/ leaf	fruit set leaf area cm^2
d	12 3 bc	184 b	10 0	107 ab	10 7	168
g	10 0 a	131 a	9 5	120 ab	9 3	129
dxg	11 0 ab	193 b	9 2	91 a	9 7	132
gxd	13 2 c	243 c	9 2	126 ab	9 2	120
dxg F2	11 4 ab	202 b	10 2	137 b	9 3	131
gxd F2	12 5 bc	212 bc	9 3	96 a	9 2	118
parental midpoint	11 2	158	9 8	113	10 0	148

Table 3 Curling and rotation of tomato leaves in three summer seasons, 1998

	Curling			Rotation		
cultivar	NH 1	NH 2	NH 3	NH 1	NH 2	NH 3
d	1 08 a	1 28 a	1 17 a	0 00 a	0 00	0 00 a
g	4 35 d	3 64 c	3 83 d	2 08 c	1 31	1 83 b
dxg	2 10 b	1 11 a	1 50 ab	0 35 ab	0 00	0 00 a
gxd	2 36 bc	1 69 b	2 00 bc	0 30 ab	0 00	0 00 a
dxg F2	3 00 c	1 74 b	2 29 c	0 80 b	0 04	0 08 a
gxd F2	2 48 bc	1 67 b	1 91 b	0 38 ab	0 01	0 09 a
parental midpoint	2 71	2 46	2 51	1 04	0 66	0 92

Std Dev	Curling			Rotation		
cultivar	NH 1	NH 2	NH 3	NH 1	NH 2	NH 3
d (n=6)	0 204	0 429	0 408	0 000	0 000	0 000
g (n=6)	0 505	0 326	0 408	0 508	0 162	0 408
dxg (n=6)	0 316	0 274	0 548	0 399	0 000	0 000
gxd (n=6)	0 344	0 221	0 000	0 274	0 000	0 000
dxg F2 (n=24)	0 770	0 484	0 550	0 607	0 141	0 408
gxd F2 (n=24)	0 748	0 376	0 526	0 587	0 035	0 417

Table 4 Stomatal conductance and leaf temperature at 5 growing stages of tomato plants in summer 1998

Nethouse 1

cultivar	stom con bef flo	leaf tem bef flo	stom con beg flo	leaf tem beg flo	stom con flower	leaf tem flower	stom con beg fr set	leaf tem beg fr set	stom con fruit set	leaf tem fruit set
d	0 53	34 5	0 58 a	33 9	0 54	34 0	0 57 ab	32 3	0 55 ab	32 8 ab
g	0 55	35 1	0 76 b	32 7	0 67	33 8	0 72 b	32 4	0 60 b	33 2 ab
dxg	0 57	34 3	0 61 ab	32 9	0 58	34 2	0 48 a	31 8	0 36 a	33 9 b
gxd	0 55	34 5	0 57 a	34 0	0 57	34 2	0 52 a	32 5	0 48 ab	33 3 ab
dxg F2	0 59	34 7	0 62 ab	33 9	0 63	33 3	0 55 a	32 3	0 48 ab	33 5 b
gxd F2	0 55	34 8	0 56 a	33 8	0 59	33 4	0 52 a	32 0	0 49 ab	32 7 a

Nethouse 2

cultivar	stom con bef flo	leaf tem bef flo	stom con beg flo	leaf tem beg flo	stom con flower	leaf tem flower	stom con beg fr set	leaf tem beg fr set	stom con fruit set	leaf tem fruit set
d	0 56 ab	34 2	0 57 ab	31 3	0 50 a	33 1	0 47	33 4	0 51	33 5 b
g	0 65 b	33 4	0 65 b	32 2	0 65 b	32 9	0 49	33 7	0 56	34 1 b
dxg	0 56 ab	33 1	0 56 ab	31 7	0 49 a	34 0	0 51	33 0	0 67	31 1 a
gxd	0 54 ab	35 1	0 61 ab	31 2	0 51 ab	34 4	0 48	33 7	0 55	33 0 b
dxg F2	0 53 a	34 5	0 55 a	31 5	0 54 ab	32 6	0 47	33 7	0 59	32 6 b
gxd F2	0 53 a	34 9	0 56 ab	31 5	0 55 ab	33 0	0 47	34 0	0 56	33 0 b

Nethouse 3

cultivar	stom con bef flo	leaf tem bef flo	stom con beg flo	leaf tem beg flo	stom con flower	leaf tem flower	stom con beg fr set	leaf tem beg fr set	stom con fruit set	leaf tem fruit set
d	0 50 ab	31 5	0 41	33 5 b	0 36	35 7 b	0 54	30 8	0 39	32 8
g	0 62 b	31 2	0 54	33 4 b	0 47	33 5 a	0 57	30 7	0 34	33 8
dxg	0 49 a	32 1	0 46	33 2 b	0 48	32 6 a	0 53	30 1	0 40	31 9
gxd	0 56 ab	30 6	0 48	33 0 ab	0 48	32 6 a	0 50	30 8	0 34	32 5
dxg F2	0 53 ab	32 0	0 51	32 8 a	0 45	33 4 a	0 57	30 4	0 35	33 4
gxd F2	0 53 ab	31 8	0 49	33 1 b	0 45	33 0 a	0 56	30 7	0 36	32 9

Table 4 cont Standard Deviations

Nethouse 1

Std Dev cultivar	stom con bef flo	leaf tem bef flo	stom con beg flo	leaf tem beg flo	stom con flower	leaf tem flower	stom con beg fr set	leaf tem beg fr set	stom con fruit set	leaf tem fruit set
d	0 030	0 549	0 046	0 582	0 055	1 100	0 053	0 436	0 123	0 564
g	0 006	0 153	0 130	1 678	0 080	1 198	0 106	0 882	0 143	0 523
dxg	0 070	0 216	0 068	0 659	0 042	1 231	0 019	0 896	0 134	1 037
gxd	0 010	0 469	0 046	0 479	0 040	0 568	0 084	0 335	0 154	0 434
dxg F2	0 121	0 923	0 089	1 196	0 102	0 805	0 103	1 270	0 133	0 913
gxd F2	0 052	0 619	0 069	0 986	0 091	0 940	0 081	1 471	0 085	0 948

Nethouse 2

cultivar	stom con bef flo	leaf tem bef flo	stom con beg flo	leaf tem beg flo	stom con flower	leaf tem flower	stom con beg fr set	leaf tem beg fr set	stom con fruit set	leaf tem fruit set
d	0 077	0 787	0 048	0 595	0 067	1 737	0 040	1 406	0 052	0 880
g	0 092	1 504	0 087	1 282	0 084	1 847	0 053	0 753	0 093	1 437
dxg	0 047	0 758	0 033	0 294	0 038	0 436	0 044	0 532	0 039	0 709
gxd	0 042	0 962	0 058	0 585	0 027	0 063	0 026	0 516	0 094	1 658
dxg F2	0 077	1 560	0 054	0 669	0 061	1 938	0 037	0 854	0 089	1 620
gxd F2	0 058	1 188	0 065	0 941	0 108	1 948	0 051	0 708	0 112	2 105

Nethouse 3

cultivar	stom con bef flo	leaf tem bef flo	stom con beg flo	leaf tem beg flo	stom con flower	leaf tem flower	stom con beg fr set	leaf tem beg fr set	stom con fruit set	leaf tem fruit set
d	0 037	0 335	0 021	0 121	0 037	0 554	0 027	0 860	0 039	1 791
g	0 068	1 038	0 067	0 760	0 094	3 048	0 059	0 650	0 055	0 637
dxg	0 066	1 521	0 068	0 200	0 210	3 179	0 075	1 355	0 69	0 836
gxd	0 055	0 589	0 065	0 133	0 157	3 405	0 057	0 509	0 019	0 449
dxg F2	0 069	1 151	0 088	0 348	0 104	3 096	0 077	1 210	0 058	2 030
gxd F2	0 072	0 868	0 053	0 682	0 125	2 887	0 062	1 070	0 079	1 442

Table 5 Fruit characters at the end of the season in nethouse 1, 1998

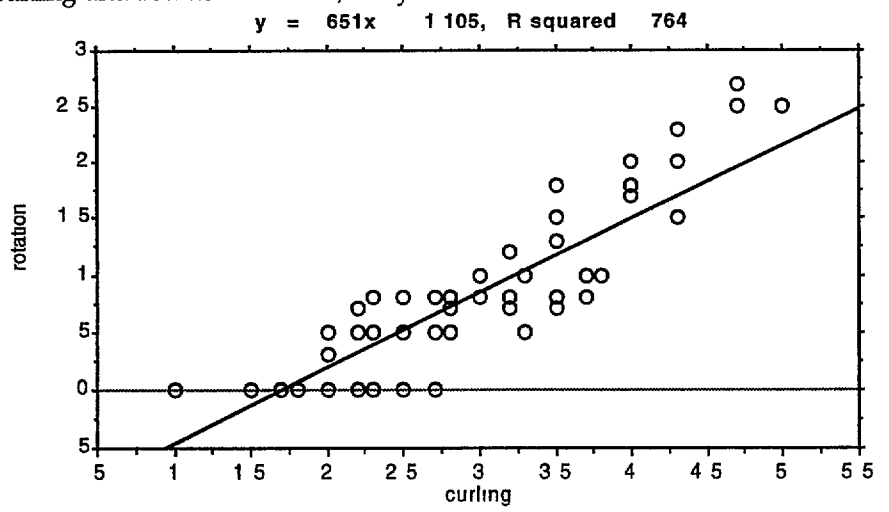
cultivar	yield (g)	mean fruit weight (g)	TSS (%)	EC (dS/m)	pH	acidity
d	1738±491	34 9 a	4 76 abc	3 42	4 05 a	3 87 ab
g	1705±187	46 2 ab	5 14 c	3 33	4 18 b	4 17 b
dxg	2607±462	52 3 b	4 45 a	3 18	4 14 b	3 49 a
gxd	2118±672	56 6 b	4 58 ab	3 21	4 10 ab	3 84 ab
dxg F2	2169±705	50 3 b	4 96 bc	3 29	4 14 b	3 73 ab
gxd F2	1829±775	43 1 ab	4 89 bc	3 29	4 16 b	3 84 ab
parental midpoint	1722	40 6	4 95	3 38	4 12	4 02

Table 6 Broad based heritability estimates (h^2) of the parameter examined in the tomato experiment 1998

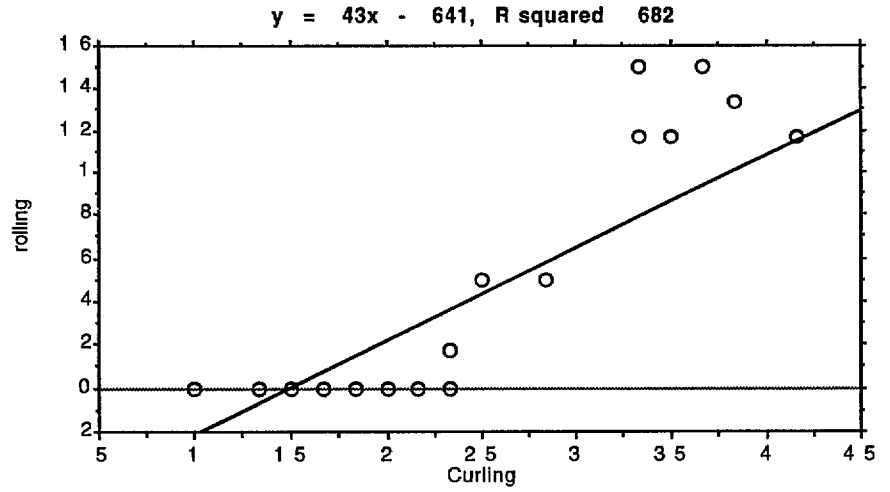
Parameter	cross	
	dxg	gxd
Dry weight - leaf	84	80
Curling - Aug	22	25
Curling - Aug -Sep	75	78
Curling - Sep	70	40
Rotation - Aug	38	32
Rotation - Aug -Sep	46	86
Rotation - Sep	33	32
Stomatal Conductance	86	90
Leaf Temperature	85	63

Fig 1 Regression between curling and rotation over three time periods in summer 1998

Curling and rotation in NH 1, early summer 1998



Curling and rotation in NH 2, summer 1998



Curling and rotation in NH 3, late summer1998

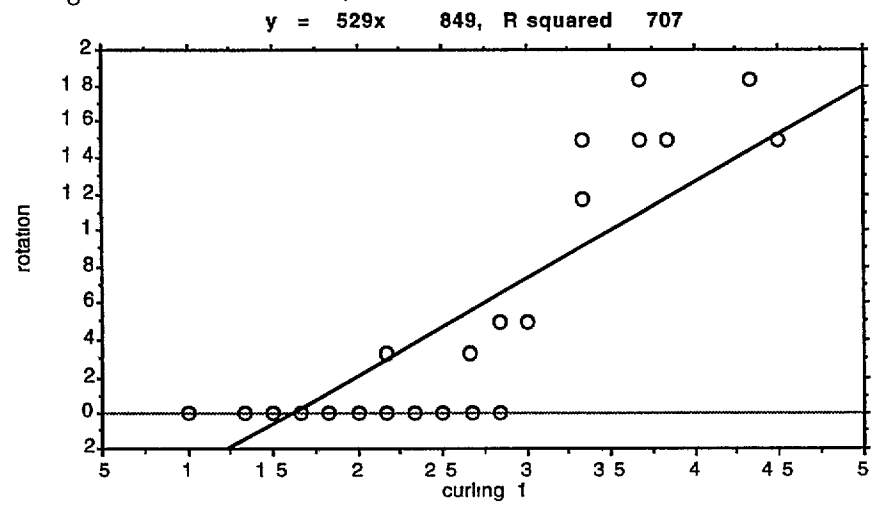


Fig 2 Regression between curling, rotation and stomatal conductance, leaf temperature in NH 1 n=72

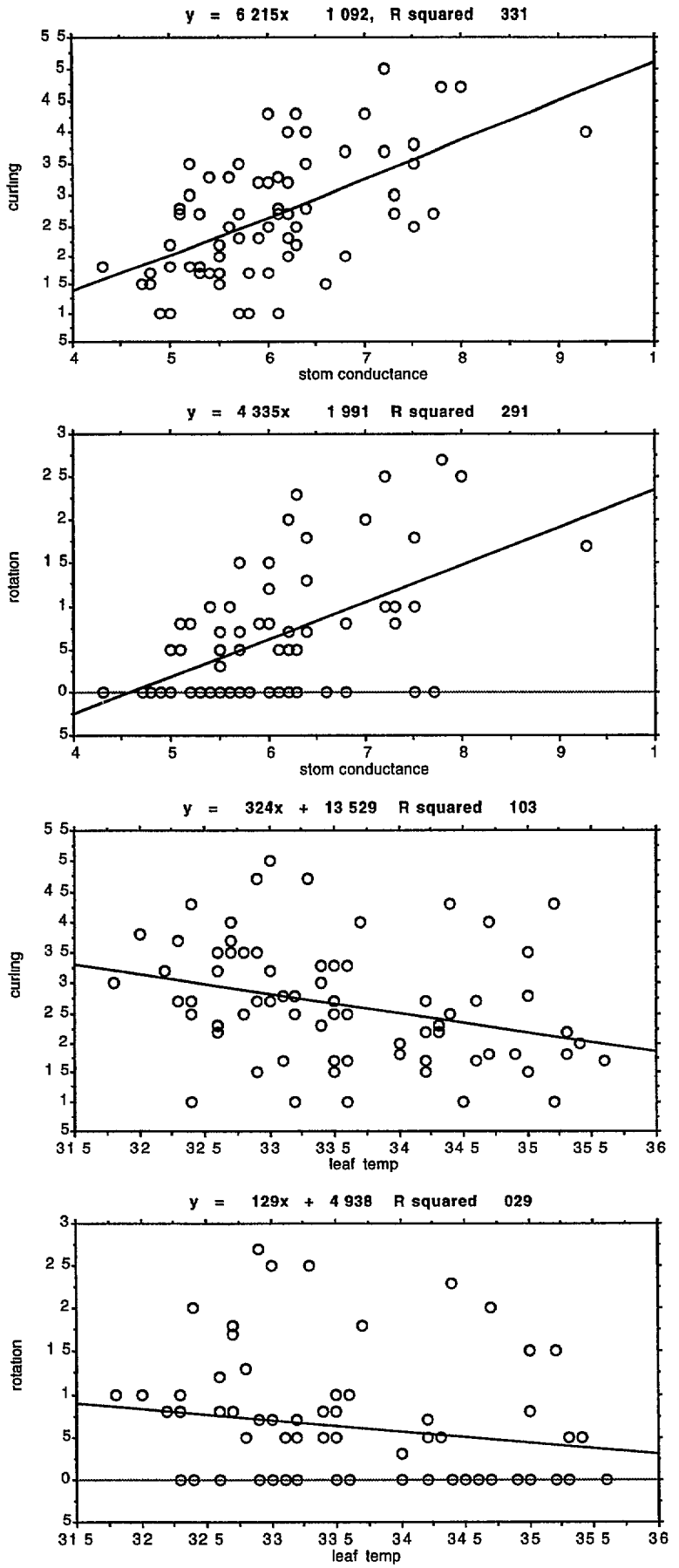
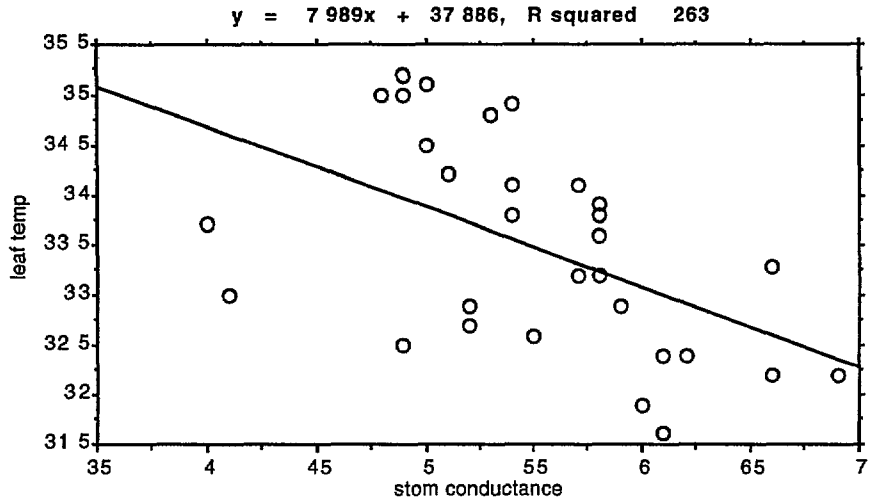
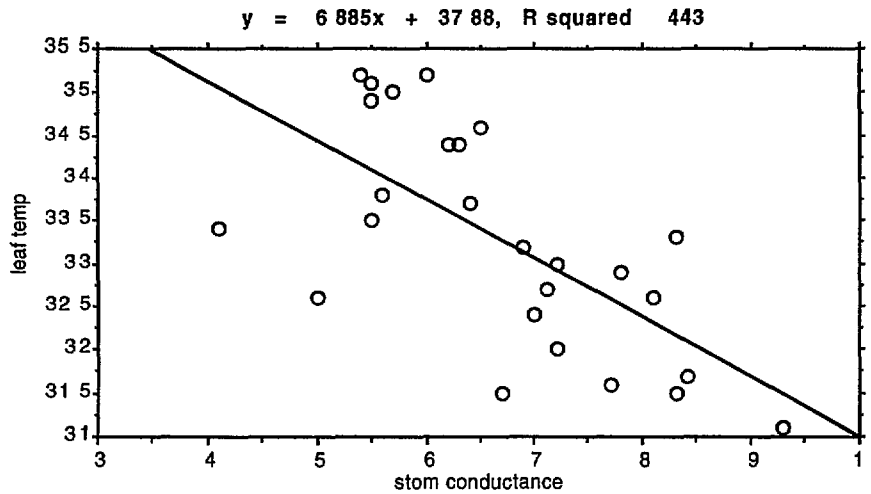


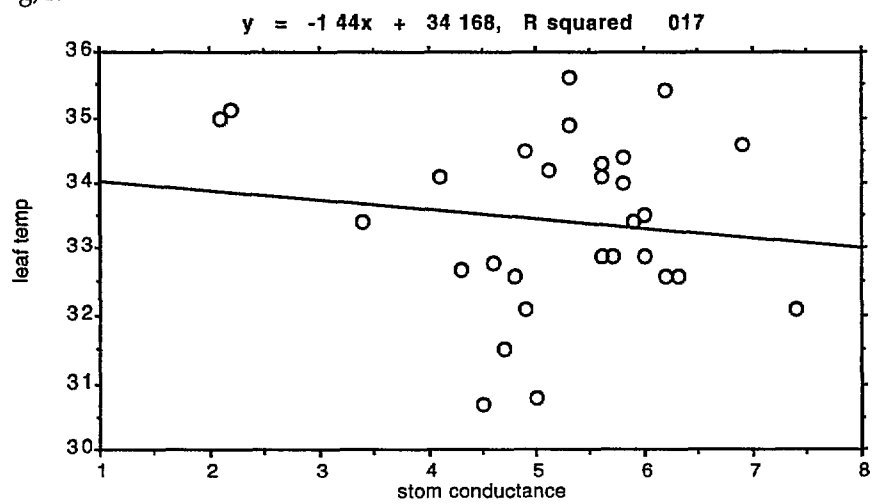
Fig 3 Regression of stomatal conductance and leaf temperature at the 6 genotypes over all nethouses
d, n=72



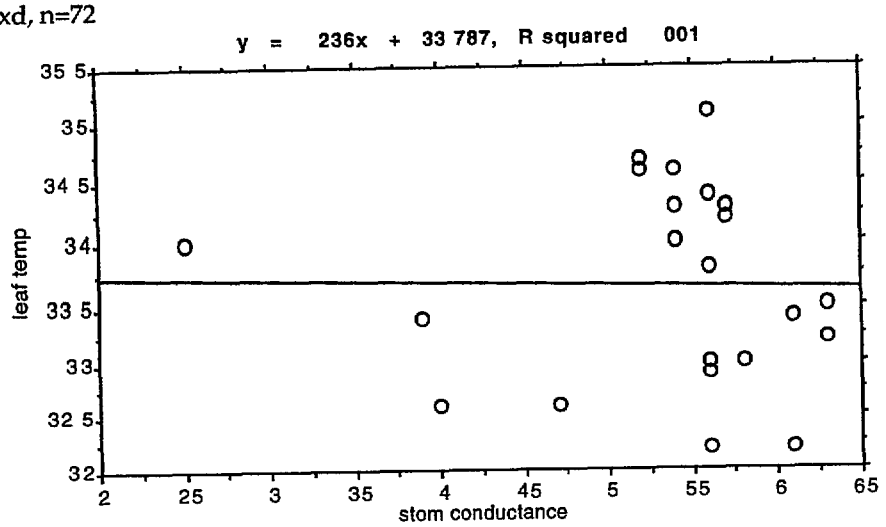
g, n=72



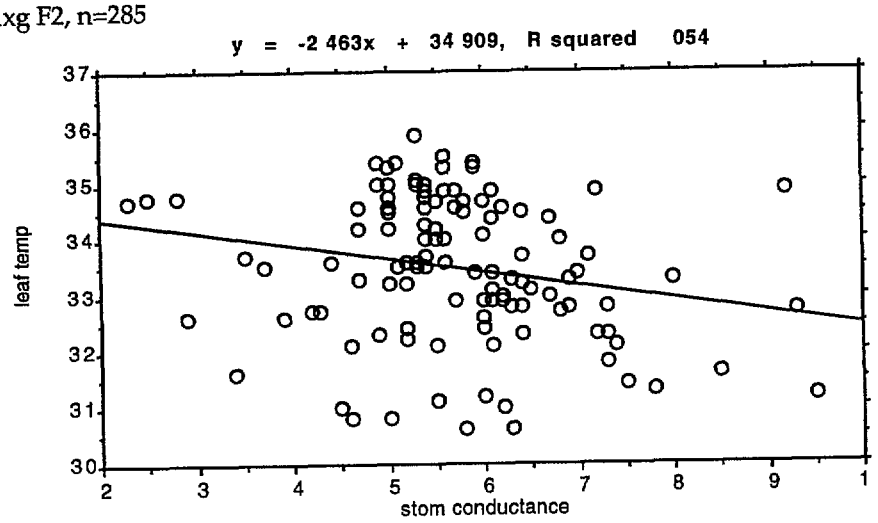
dxg, n=72



gxd, n=72



dxg F2, n=285



gxd F2, n=285

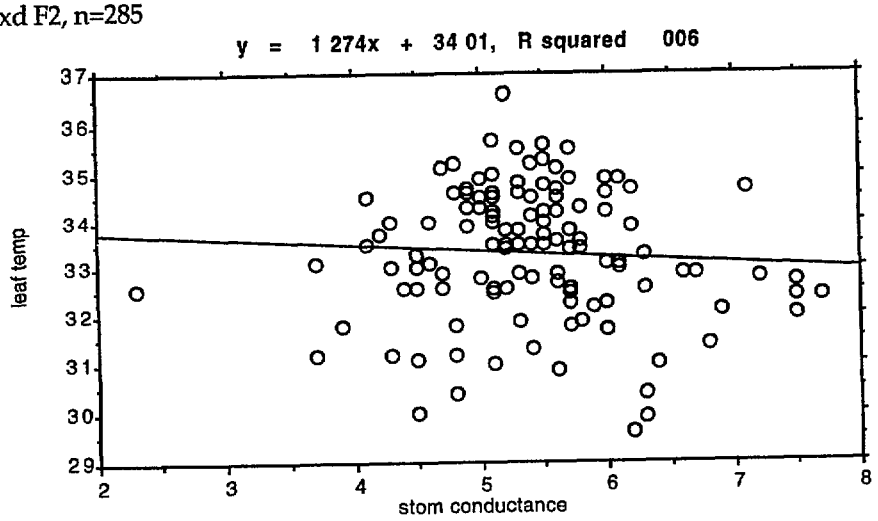
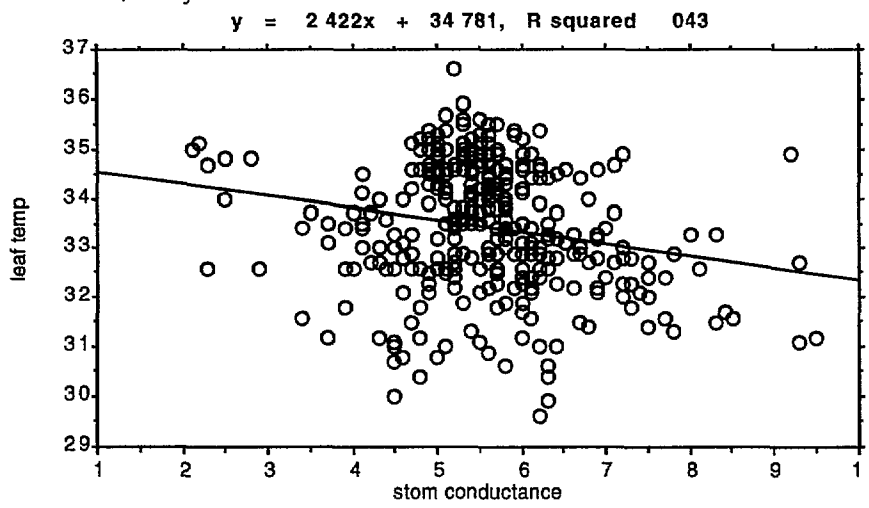
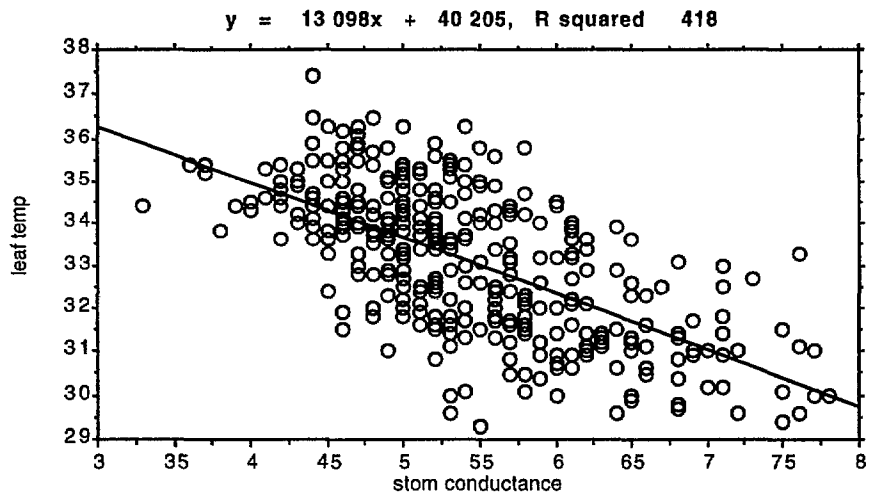


Fig 4 Regression between leaf temperature and stomatal conductance over three time periods, all genptypes pooled

Nethouse 1, early summer 1998



Nethouse 2, summer 1998



Nethouse 3, late summer 1998

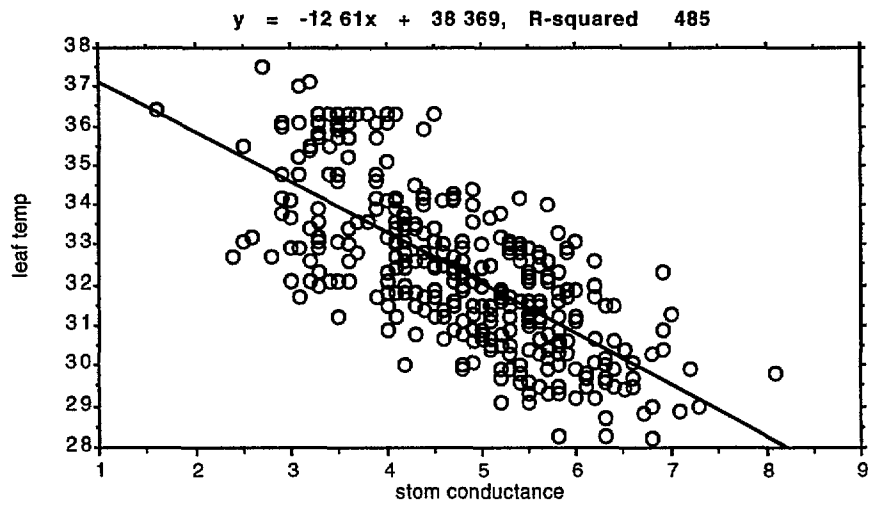
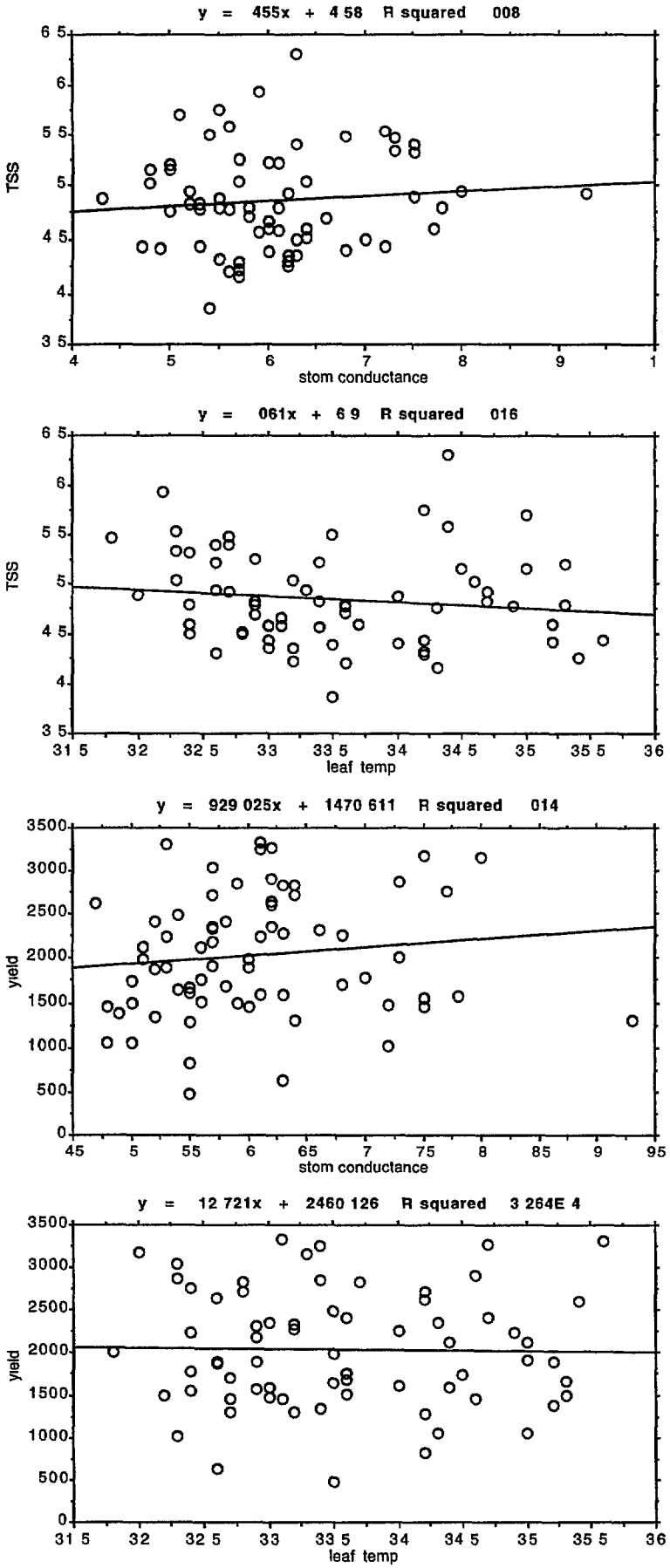


Fig 5 Regression between TSS, yield and stomatal conductance leaf temperature



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C SCIENTIFIC IMPACT OF COLLABORATION

The P I s fully organized the research program as an integrated unit Drs Mendlinger, Polimbetova and Tal have spent about 33% of their time on the project and Ventura 100% Dr Mendlinger sent his Ph D student Sergei Volis to Kazakhstan in September 1998 to further the coordination

D DESCRIPTION OF SCIENTIFIC IMPACT

It is still too early for any major claims but the results of the first and second years' study strongly support our hypothesis of the use of heat avoidance traits for improving heat resistance

E STRENGTHENING OF DEVELOPING COUNTRY INSTITUTIONS

In the second year the project helped strengthen the Institute for Plant Physiology, Genetics and Biotechnology in two ways

(a) training - several Kazakh scientists spent time in Israel obtaining training and working on the project

(b) purchase of equipment - several needed equipment was purchased and additional equipment will be purchased in the second year

F FUTURE WORK

The project is on schedule and both sides will be completing the third year of the project

SECTION II

A MANAGERIAL ISSUES

No major managerial issues have arisen The transfer of funds to Kazakhstan has gone smoothly

B BUDGET

No major changes have occurred in the budget

C SPECIAL CONCERNS

We have no major special concerns in this project

D. COLLABORATION, TRAVEL, TRAINING AND PUBLICATION -

The degree of cooperation in planning and conducting the research program has been described above In respect to travel, Dr Mendlinger visited Kazakhstan in August 1987 and his Ph D student, Serjei Volis in September 1998 to examine the experimental plots and discuss the project with Dr Polimbetova Due to this project three Kazakh scientists have worked in Israel

1 Dr Intyke Shayakmetova - spent 6 months working on the tomato project,

2 Dr Elizabetha Bogdanova - spent 2 weeks visiting the wheat fields,

3 Ms Anel Tormanova - spent 5 months in Israel working on the project

In addition, junior scientists in both countries have been working on the project