INFLUENCE OF TEMPERATURE ON SEED GROWTH AND SEED RIPENING IN SUNFLOWER*

By

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Abstract

Rumanian sunflower hybrids HS 90 were grown in the glasshouse until the beginning of flowering and then transferred to climatic rooms with three different day/night temperature regimes: 30/22°C (I), 25/17°C (II), 20/12°C (III). Light intensity was about 80 W m^{-2}, daylength 14 h, and relative humidity 65%. Starting two weeks after fertilization, seed samples were drawn from one plant per treatment at a rate of once to twice a week. The fresh weight, dry weight, total content of lipids and triglycerides, and fatty acid composition were determined in hulls and kernels separately. Rate of desiccation of both hulls and kernels was positively correlated with temperature. At any time the hull contained more moisture than the kernel. Desiccation of the kernels almost stopped between the 42nd and 64th day after flowering (DAF) in regimes I (28%) and II (32%) and between the 52nd and 70th DAF in regime III (37%). The kernels reached their maximum content of triglycerides, viz. 61% (I), 61% (II) and 58% (III), at 44, 44 and 54 DAF, respectively. Afterwards there was a small decrease. The triglyceride content of the hulls was less than 1% until the 60th DAF at all treatments, but then it suddenly rose to 15% (I), 11% (II) and 4% (III), more or less concommitant with the final desiccation period. The oleic acid content was positively and the linoleic acid content negatively correlated with the temperature. In the kernels these contents were complementary during the entire experimental period. Their sum was about 87% of the total triglyceride content. In the hulls the complementarity was less manifest. From our results it follows that when average temperatures during maturation period of sunflowers are between 16 and 26°C, harvesting should not be later than two months after flowering.

Introduction

The period of seed growth and seed maturation is distinguished by a number of changes in the composition of the seed. These concern both the absolute and the relative amounts of the various components. Most of these compounds keep on increasing until a constant level has been reached, but the rate of the different increases is neither equal nor constant in the course of time, resulting in a widely changing procentual composition. In many cases the absolute amounts of certain components decrease after a maximum has been reached. On the other hand also the loss of moisture, either from the seed taken as a whole or from the kernel need not show a continuous course. The outcome of

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all this is that the moment at which moisture content may be deemed sufficiently low to permit harvesting and subsequent storage with a minimal risk of damage does not necessarily coincide with the moment of maximum seed or oil yield. A compromise will thus have to be found.

In this report we present a detailed description of the time-course of changes in moisture content, dry matter, fat and fatty acids that occur in devleoping seeds of sunflower at different day/night temperature regimes. In the discussion we have tried to lay a link between the results of our determinations, seed quality and best period for harvesting.

Somewhat akin to the question when yield or dry weight of the grains is at its maximum is the matter of relative growth of hull and kernel. Because in hull and kernel dry matter is not accumulated at the same rate and even may be transferred from one to the other especially towards the end of the maturation period, it appeared of interest to follow developments in both parts separately.

Materials and Methods

One hundred sunflower plants (Helianthus annuus, HS 90, a Rumanian hybrid) were grown in 271 baskets in light soil in the greenhouse until the onset of flowering. Out of this number 60 plants were selected for equal height and development and from these 20 were transferred to each of three climatic rooms. Day/night temperatures in these rooms were $30/22^{o}$, $25/17^{o}$ and $20/12^{o}C$. The photoperiod was 14 h, but the high temperature period was only 12 h, starting one hour later and ending one hour earlier than the light period. Light intensity was about 80 Wm^{-2}, relative humidity was 65-70%.

In the climatic rooms the plants were hand pollinated in the course of the first three days following transfer. On the 12th day after the beginning of flowering (DAF) two plants in each room were harvested and afterwards one plant per room was taken every 3 to 7 days depending on temperature treatment and stage of development.

The sampling of the seeds was done in two ways. Two samples (A), consisting of 30 full seeds were taken from one region in the outer third part of the head. In these seeds fresh and dry weight, content of total lipids and triglycerides and fatty acid composition were determined in hulls and kernels separately.

Samples B, also taken in duplicate, consisted of a natural mixture of full and empty seeds collected radially from brim to center in two opposite sectors of the inflorescence. In these samples only fresh and dry weight of the whole seeds were determined.

Total lipids were determined in two different ways, both suitable for small samples (about 250 mg):

1. A residue method according to Franzke (1963) and Marquard (1972);
2. A GLC-method with methyl heptadecanoate as an internal standard.

With the residue method 250 mg of sample is extracted in a small stainless steel vessel with petroleum ether and a stainless steel ball. Extract and residue are quantitatively transferred into a 15 ml centrifuge tube and centrifuged. The solvent is drawn off and the residue is washed 3 times with petroleum ether. After drying the residue is weighed. Loss in weight corresponds with oil weight.

With the GLC-method the same extraction procedure is used in which, however, the internal standard is dosed with a 5 ml petroleum ether solution into the vessel. After transfer of the extract and centrifugation a small part of the extract is saponified and esterified giving methyl esters of all triglyceride fatty acids. The GLC peak areas are used to calculate the oil content and the fatty acid composition.

The GLC-method determines only the triglyceride fraction and can be compared with an apolar Soxhlett extraction when no other apolar compounds (waxes, etc.) are present.

The residue method normally gives higher values (2-3%) than the GLC-method because of incomplete transfer of the extract and small losses of solids during drawing off.

Results

Moisture

The rate of desiccation of the seeds is not constant (Figure 1a). During the first two weeks it is slow and hardly influenced by temperature. Then moisture content begins to decrease more rapidly at a rate that slightly increased with temperature. Desiccation now continues until a level is reached that lies between 30% at 30/22^{o} and 25/17^{o} and between 52 and 70 DAF at 20/12^{o}C. After this stage there is a rapid final desiccation until 10% moisture at the moment the experiment ended. From Figure 1b it appears that this pattern is almost entirely due to events in the kernel. Desiccation rate of the hulls (Figure 1c) is more constant. At all temperatures desiccation of the hulls is slower than that in the kernels. At any moment moisture content of the kernels is smaller than that of the hulls.

Fresh and Dry Weight

Seed growth, particularly the accumulation of reserve material in the kernels is strongly dependent on temperature. This appears in the first place from Figure 2 in which proportion of kernel is plotted against time. There is a maximum of 80-85% kernel that is about equal for all treatments, but this value is attained at 42 DAF at 30/22^{o}, at 48 DAF at 25/17^{o} and at 54 DAF at 10/12^{o}C. During the latter part of the ripening period there is a small decrease in kernel percentage at all temperatures, which will be discussed later.

The rate of increase in dry weight of kernels and seeds and the maximum level attained were almost equal at the two highest temperature regimes (Figures 3 and 4). At 20/12^{o}C both the accumulation rate and the maximum dry weight were lower. During the second part of the ripening period dry weight of the kernels decreased at a rate that seems to be negatively correlated with temperature. This may mean that the temperature coefficient of the sum total

FIGURE 1. Moisture content (% of fresh weight) in sunflower seeds as a function of days after flowering at various day/night temperature regimes.

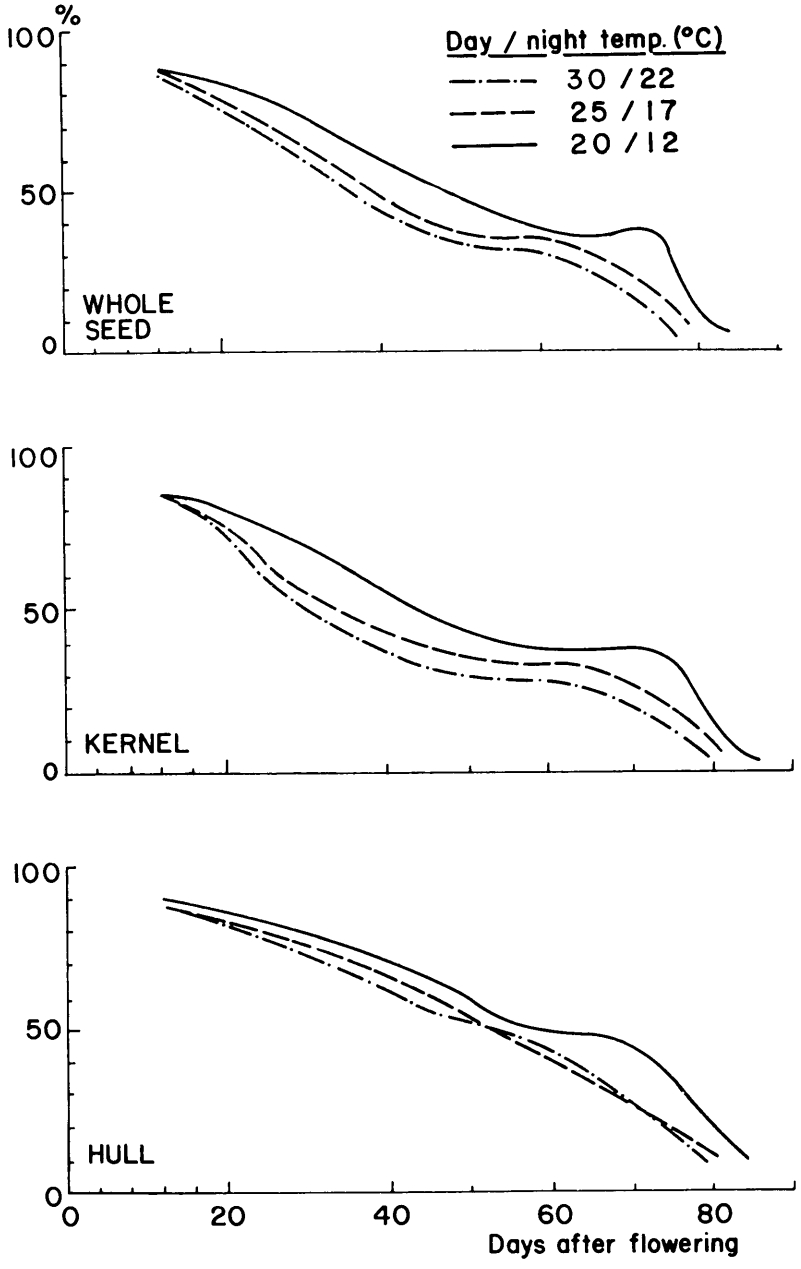


FIGURE 2. Kernel fresh weight (% of seed) as a function of days after flowering at various day/night temperature regimes.

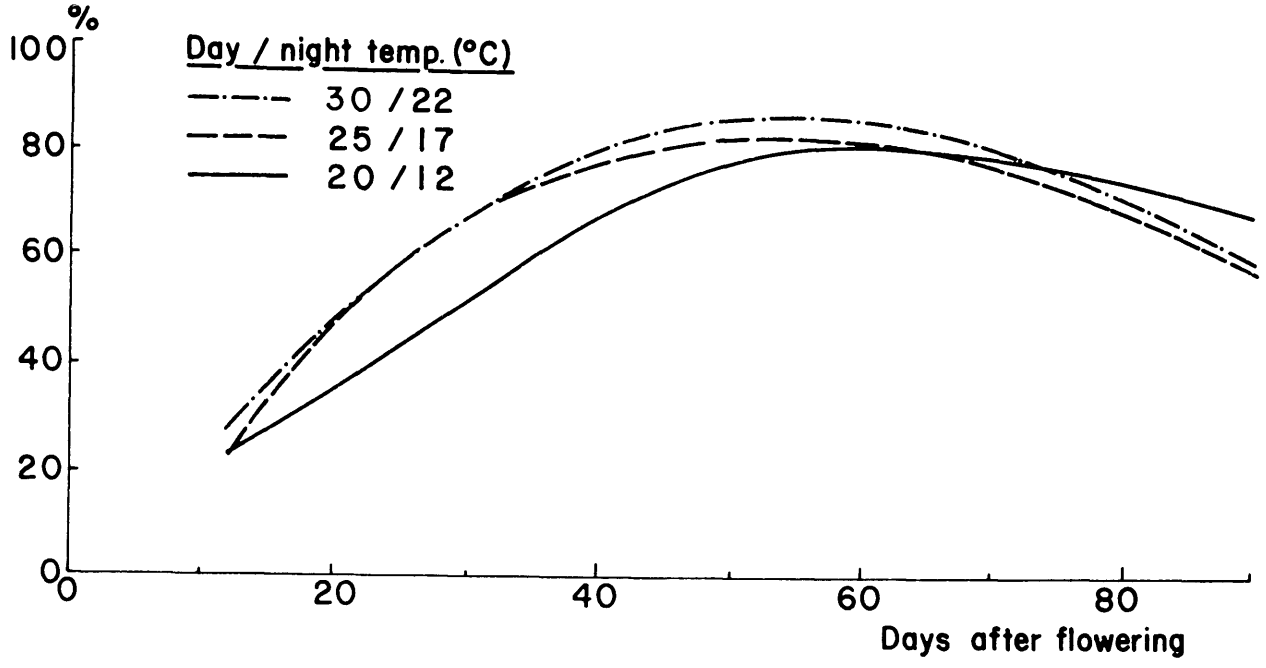


FIGURE 3. Accumulation of dry matter in kernels (mg) as a function of days after flowering at various day/night temperature regimes.

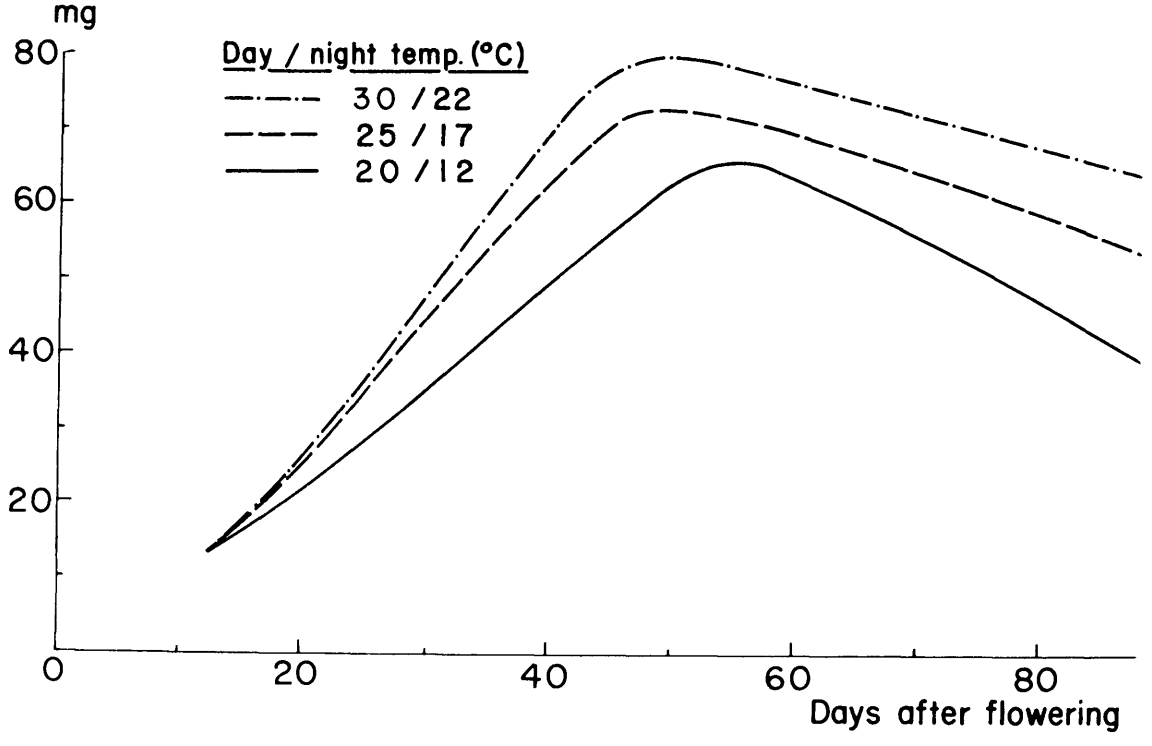


FIGURE 4. Accumulation of dry matter in - full - seeds (g) as a function of days after flowering at various day/night temperature regimes.

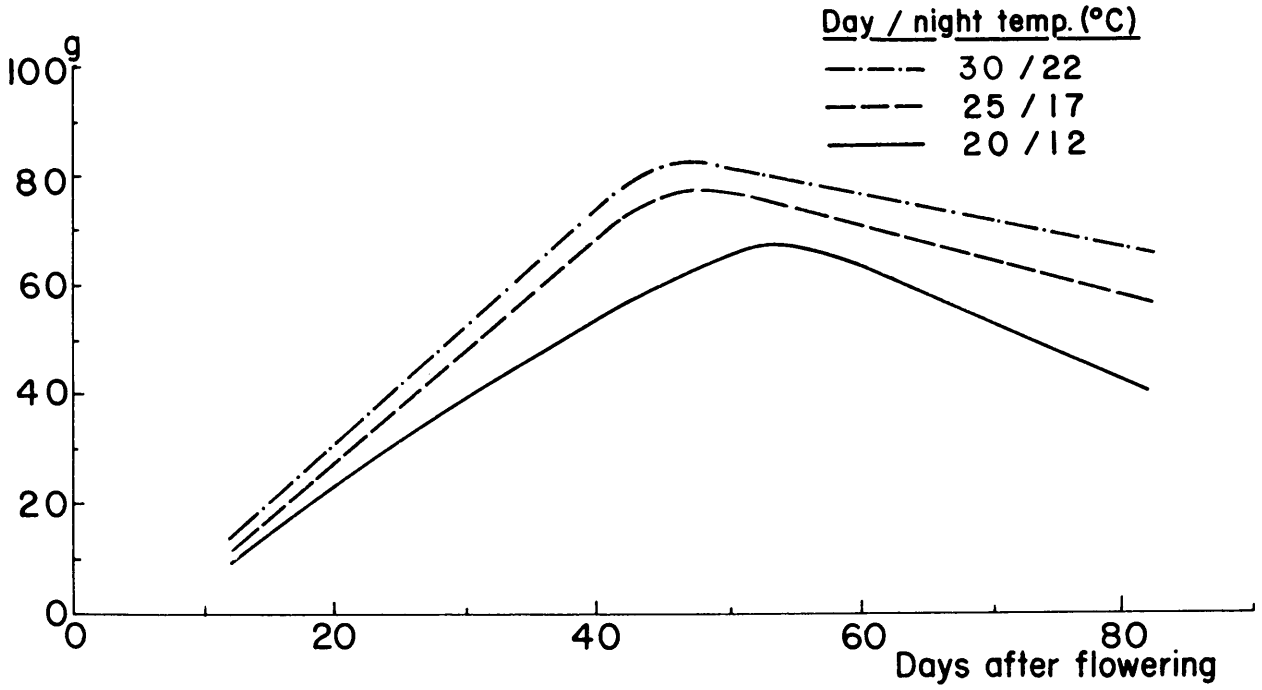
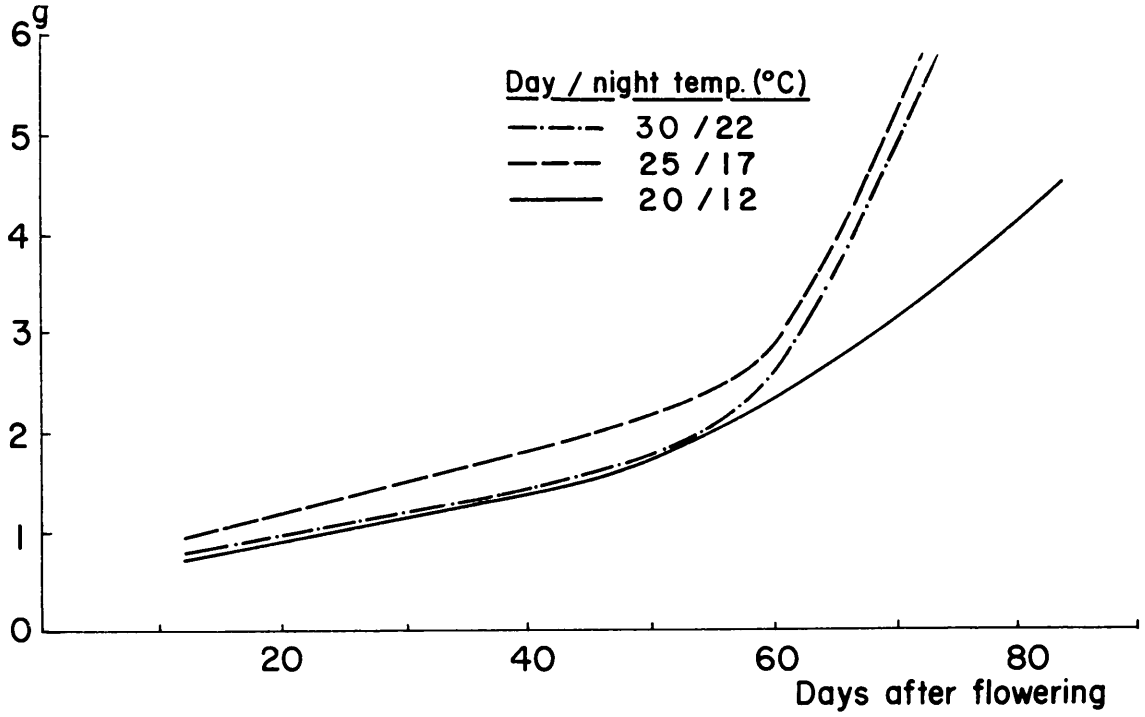


FIGURE 5. Accumulation of dry matter in radially sampled good and faulty seeds (g hundred-seed weight) as a function of days after flowering at various day/night temperature regimes.



of the anabolic processes in the plant that control seed filling is higher than that of the dissimilation processes in the seed, but other causes may also play a role.

The course of the dry weight as an average of full and empty seeds that have been sampled according to method B presents a different picture (Figure 5). There is a slow but steady increase during the first 50 days and then a rise that at the two higher temperature treatments is sudden and steep. In a later stage we shall discuss that oil content in the hulls shows a similar pattern.

Lipids and Triglycerides

In Figures 6a and b and 7a and b the time-courses of lipids and tryglycerides contents in kernels and hulls are presented. In some instances those at the two higher temperatures coincide. At the 20/12°C treatment both accumulation rate and maximum values are lower. In the kernels the proportion of non-triglycerides in the total of lipids must be very small since the respective curves for the three treatments in Figures 6a and 7a are rather similar. But in the hulls (Figures 6b and 7b) things are different.

During the first five weeks at the higher temperatures, resp. eight weeks at 20/12°C, the content of lipidic compounds is very low, and of these tryglycerides amount to no more than 20%. Then there is a rather rapid increase in fatty substances that is for the greater part due to an increase in triglycerides. Whereas during the former period there was hardly any difference between the temperature treatments, this rise is temperature dependent.

During the first few weeks the rate of increase of oil content in the kernel exceeds the growth rate of the seed in terms of dry matter. After that oil content continues to rise at a much lower rate than dry matter as a whole and during the last part of the maturation period there is a slight decrease. Peak values are almost equal at all temperatures. End values show differences. Eventually higher temperatures appear to cause higher oil content.

Fatty Acid Composition

Kernels

The time courses of fatty acids in the kernels are presented in Figures 8a, b, and c. The contents of oleic and linoleic acids are complementary over the whole ripening period. If one curve rises the other goes down. The sum, however, is practically independent of temperature. Commencing at the end of the fifth week sums of oleic and linoleic acid contents are 88, 87 and 86% at 30/22°, 25/17° and 20/12°C, respectively. The final contents of both fatty acids are strongly influenced by temperature (Figure 9).

Hulls

The course of the fatty acid pattern in the hulls is less straightforward and clear than that in the kernels. It is conceivable that this is due to the fact that the changes in content of the various acids are not only the direct result of synthetic processes occurring on the spot but are also influenced by events in the kernel as this is the center of the whole organ. Yet there are some interesting general points to make, in particular concerning the relation between oleic and linoleic acids. Therefore, I have lifted these two out and

FIGURE 6. Accumulation of Triglycerides (% of dry matter) in kernels and hulls as a function of days after flowering at various day/night temperature regimes.

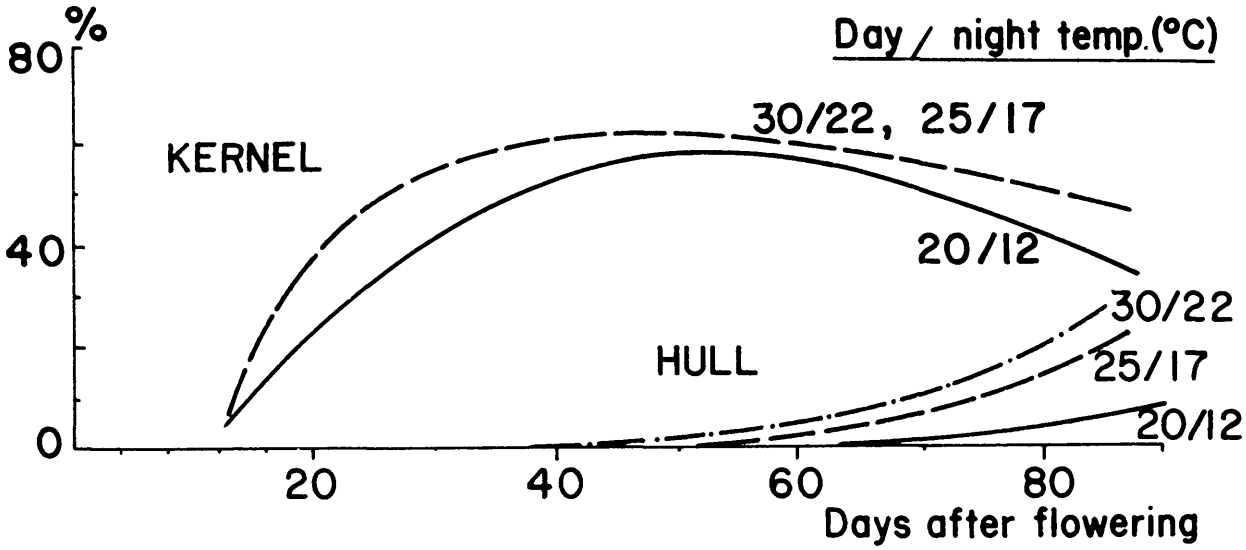


FIGURE 7. Accumulation of lipids (% of dry matter) in kernels and hulls as a function of days after flowering at various day/night temperature regimes.

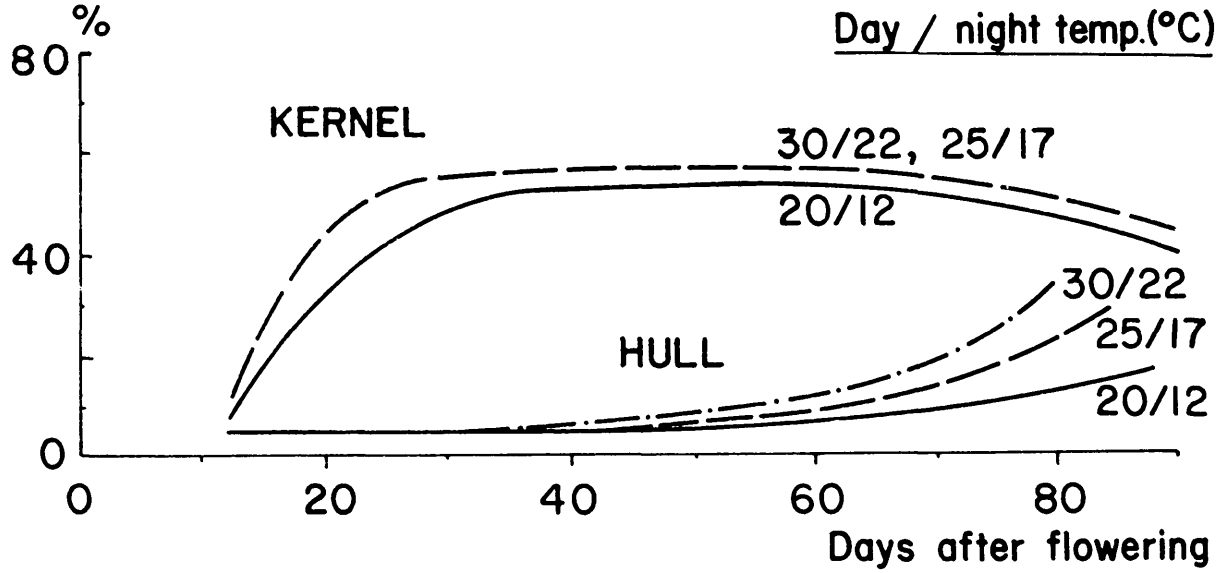


FIGURE 8. Fatty acid composition (% of total fatty acid) of oil from the kernels as a function of days after flowering at various day/night temperature regimes.

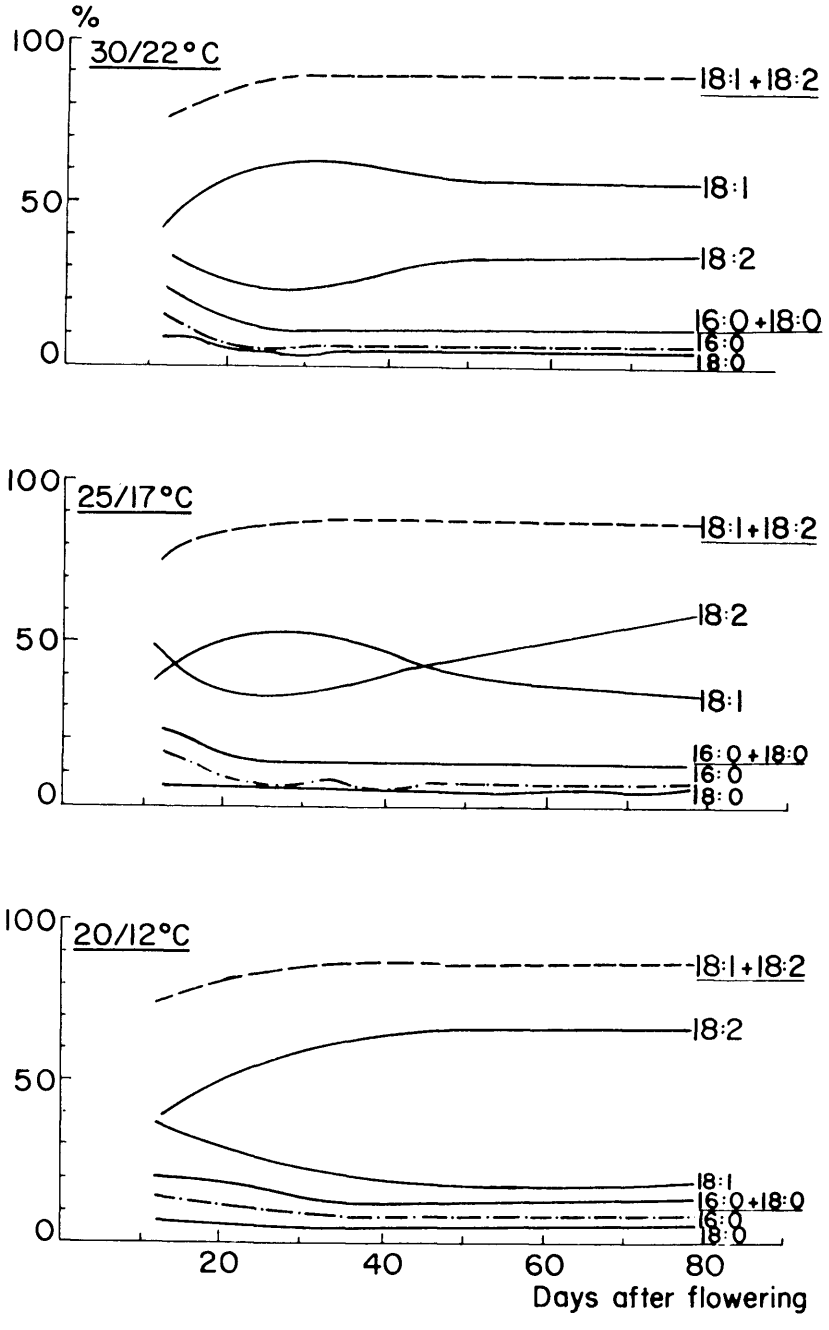


FIGURE 9. Effect of temperature on oleic and linoleic acid content (% of total fatty acid) of the oil from the kernels.

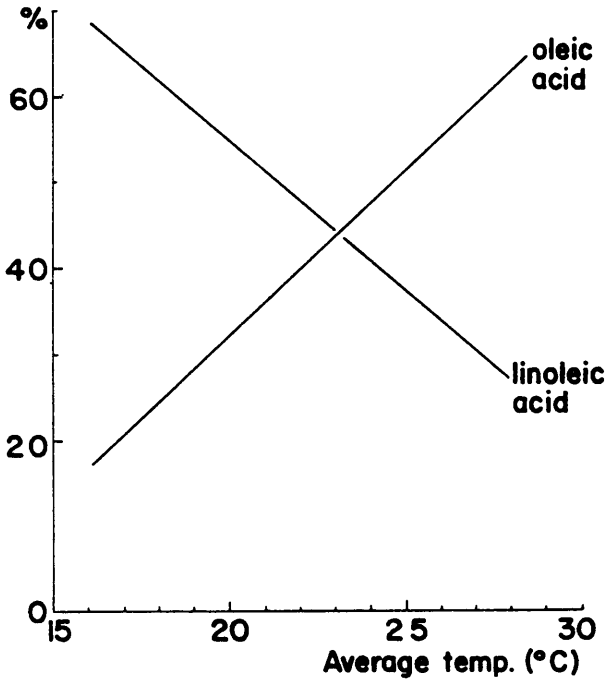
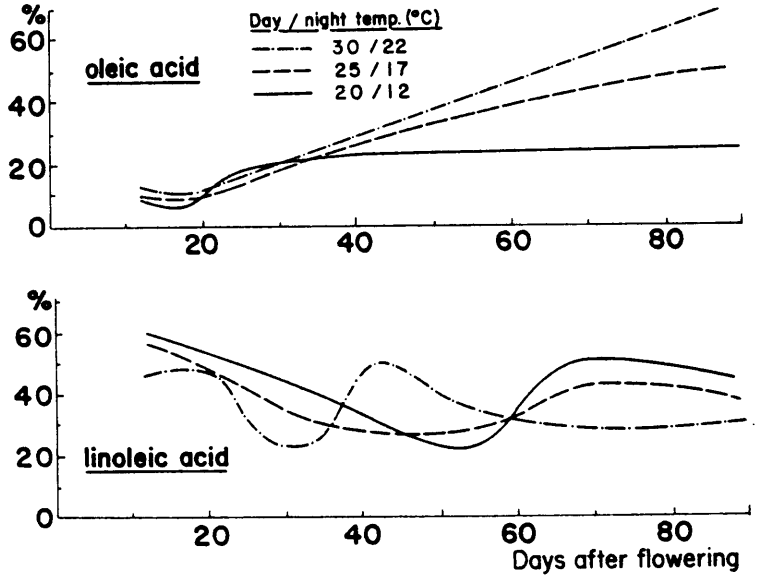


FIGURE 10. Unsaturated fatty acid content (% of total fatty acid) of the oil from the hull as a function of days after flowering at various day/night temperatures.



presented them in the Figures 10a and b. The contents of the two fatty acids are grossly complementary but not so strictly as in the kernels. Both are influenced by temperature in the same sense as in the kernels but to a smaller extent, especially linoleic acid. In oleic acid the temperature effect increases in the course of the maturation period. It follows that the sum of the two contents is not constant over a longer period. It is also more affected by temperature and it is especially low at the end of the lowest temperature treatment (Table 1). At the lowest temperature treatment the contents of 16:0 and 18:0 are higher than those in the kernels.

TABLE 1. Final Contents of Oleic and Linoleic Acids in Hulls and Kernels of Sunflower Seeds that have Matured at Three Different Temperature Regimes (in % of Total Fatty Acids).

Day/Night Temperatures (°C)	Hull			Kernel		
	18:1	18:2	18:1+18:2	18:1	18:2	18:1+18:2
30/22	58	29	87	55	34	89
25/17	46	36	82	37	50	87
20/12	24	40	64	17	69	86

Oil Quality

Instead of by the contents of the single most important fatty acids, quality of the seed might also in a kind of shorthand way be assessed by either the ratio of linoleic (18:2/18:1) or by that of the main unsaturated to the main saturated acids (18:1 + 18:2/16:0 + 18:0). This is done in Table 2, and some interesting conclusions may be drawn from this way of presentation.

TABLE 2. Influence of Temperature on Oil Quality.

Day/Night Temperatures (°C)	Ratio $\frac{18:2}{18:1}$		Ratio $\frac{18:2 + 18:1}{16:0 + 18:0}$	
	Kernel	Hull	Kernel	Hull
30/22	0.621	0.502	8.568	7.937
25/17	1.340	0.767	7.029	5,405
20/12	4.065	1.423	6.553	2,187

The quality of the oil from the kernels is at all temperatures and by both standards better than that of the oil from the hulls. But the higher the temperature the smaller the difference.

The ratio linoleic/oleic in the kernel is more temperature dependent than that in the hull. With the ratio unsaturated/saturated, however, the reverse is true.

According to the linoleic/oleic standard oil quality declines with maturation temperature whereas it improves according to the second standard. Yet any of the standards may be found applicable at some time or other.

Conclusion

After the maximum values had been reached grain weight decreased. This decrease was also found by Anderson (1975), Izzo et al (1976) and Dompert and Beringer (1976). In the two former reports and in our Figures 6a and 7a there seems to be also a small decrease in oil content in percentage dry matter, but this might not be significant. From a comparison of Figures 3 and 4 it follows that the decrease in achene dry weight is almost entirely on the account of the kernel, due to the fact that with increasing senescence of the leaves the dissimilatory processes in the seed get the upper hand of the accumulation of reserve substances. Moreover, we noted a marked increase in triglycerides in the hulls after the end of the 7th week (Figures 6b, 7b). Parallel to this we found a rapid increase in dry matter in samples that had an increased proportion of hull material (Figure 5). So, the decrease in kernel weight may be caused at the same time by respiration and by a transfer of lipidic substances to the hull. The walls of the cells of the hulls are much thicker and harder than those in the kernel, which impedes the extrusion of the oil.

Migration of oil from kernel to hull inevitably will thus cause a certain loss at processing. Morever, at low temperatures the hull oil is of lower quality than the kernel oil by the 18:2/18:1 standard, as was shown in Table 2. This means that under these circumstances not only good oil may be lost but that also a small part may be replaced by oil of a lesser quality. Harvesting should thus take place before this migration is likely to occur, that is about the 8th week after flowering, depending on temperature, and at a moisture content not lower than 20%.

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