Effects of temperature on low-linolenic rapeseed oil fatty-acid composition

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Abstract

Rapeseed oil, rich in alpha-linolenic acid (C18:3), is easily oxidized when heated and therefore not suitable for deep frying. According to Everard (2004), the typical unpleasant « room odor » appears above a threshold content of 2.5 % C18:3, while its content varies around 8 to 10% in usually grown double-low varieties. Low-linolenic varieties with C18:3 content lower than 3.5% have been selected, but a large variability is still observed among locations and years. In order to meet the target content of 2.5 %, it is important to identify factors affecting the fatty acid profile of oilseed rape.

Fatty acid composition is affected by environmental conditions, temperature being the main factor. Merrien (2005) showed for conventional rapeseed varieties that low minimal temperatures during the 60 days following the onset of flowering are related to high linolenic acid content with R^2=0.37.

Monitoring the fatty acid profile of low-linolenic varieties from the beginning of grain filling to maturity showed that linolenic acid synthesis occurs mainly between 550 and 850 degree-days after the onset of flowering, that is during the 20 first days of grain filling in Swiss conditions, i.e. 40 to 60 days after the onset of flowering. During this period, the correlation between final linolenic acid content and minimal temperature is even better, with R^2=0.87.

An easier way to show this relationship is based on the assumption that fatty acid desaturases regulated by temperature are active at low temperatures only. It consists in counting how many times during this period daily minimal temperature reaches a lower level than a threshold temperature of 13°C. The correlation between final linolenic acid content and number of days with minimal temperature below 13°C is as good as the one presented before, i.e.R^2=0.85. It could also be used as a very simple predictive model and showed a good accuracy in 2005 in Changins.

Key words: alpha-linolenic acid, temperature, grain filling, low-linolenic cultivars

Introduction

Conventional rapeseed oil contains about 7 to 10% alpha-linolenic acid, polyunsaturated fatty acid of the omega-3 family. It is therefore recognized as a very healthy edible oil. However, high content of omega-3 fatty acids can cause difficulties when heated: Everard (2004) showed that above a threshold content of 2.5% C18:3, heating the oil produces a typical unpleasant odour called "room odor". Oils rich in polyunsaturated fatty acids like rapeseed oils need an additional industrial process called "hydrogenation" to lower the unsaturation and therefore improve their technological properties. New varieties with less than 3.5% linolenic acid have been bred to provide industries with a new raw material for the production of frying media and avoid hydrogenation. However, a large variability of linolenic acid content is often observed and the target of 2.5% linolenic acid is not always met. Many studies already showed an impact of climatic conditions on oil content and composition. When grain filling occurs at lower temperatures, more polyunsaturated fatty acids are produced. This was observed in conventional varieties (Trémolière & al 1997, Pritchard et al. 2000) and for a low-linolenic one (cv. Stellar) (Deng and Scarth, 1998). Drought stress could also increase linolenic acid storage (Triboi-Blondel and Renard 1999). Izquierdo et *al.* (2002) in sunflower and Merrien (2005) in conventional double-low rapeseed varieties were the first who quantified this trend with a linear negative relationship between minimal temperatures (monitored from the onset of flowering) and final linoleic (for sunflower) or linolenic (for rapeseed) acid content.

The goal of the present study was to define if temperature affects linolenic acid content in recently bred low-linolenic cultivars and which growth stage is temperature sensitive. A second goal was to quantify the impact of temperature on final linolenic acid content in seeds of low-linolenic rapeseed varieties.

Materials and methods

Grain filling

Sequential harvests (every 7-10 days) were carried out from the beginning of grain filling until maturity with cultivar Splendor (Monsanto). Farmer's fields were selected in Calève in 2004 and Monnaz in 2005 (VD, Switzerland). They were large enough, free of volunteer and well isolated from other rapeseed fields to avoid cross-pollination. The 2006 values came from a well-isolated field at the Changins experimental station (VD, Switzerland). Three replicates were taken each time. Pods were harvested by hand and grain dried at 60°C before analysis of fatty acid composition by gas chromatography. Oil content

was analysed by nuclear magnetic resonance (NMR). Sub samples were used to determine grain dry matter content (100 °C, 24 hours) and thousand seed weight.

Temperature impact

Low-linolenic varieties MSP01, Splendor and MSP11 (Monsanto) were tested in the swiss varietal testing network since harvest 2002. In each location, individual plot size was 25 to 29m^2 with three replicates in a lattice design. Low-linolenic plots were not isolated and received pollen from any neighbour plots. We assume that cross-pollination was equivalent for each plot and therefore can be compared with any other low-linolenic rapeseed plot of the network. Sowing density ranged from 80 to 100 plants/m2 and usual crop management practices for oilseed rape were used, however without fungicide or growth regulator application. Mean and minimal daily temperatures were recorded by the closest meteorological station. The onset of flowering date was recorded for each variety when the stage 61 (beginning of anthesis) is reached on the basis of the international BBCH (Biologische Bundesanstalt, Bundessortenamt, Chemische Industrie) scale. Plots were machine harvested at full maturity. Fatty acid composition was analyzed by gas chromatography and oil content by near infrared spectrometry.

Results and discussion

Grain filling

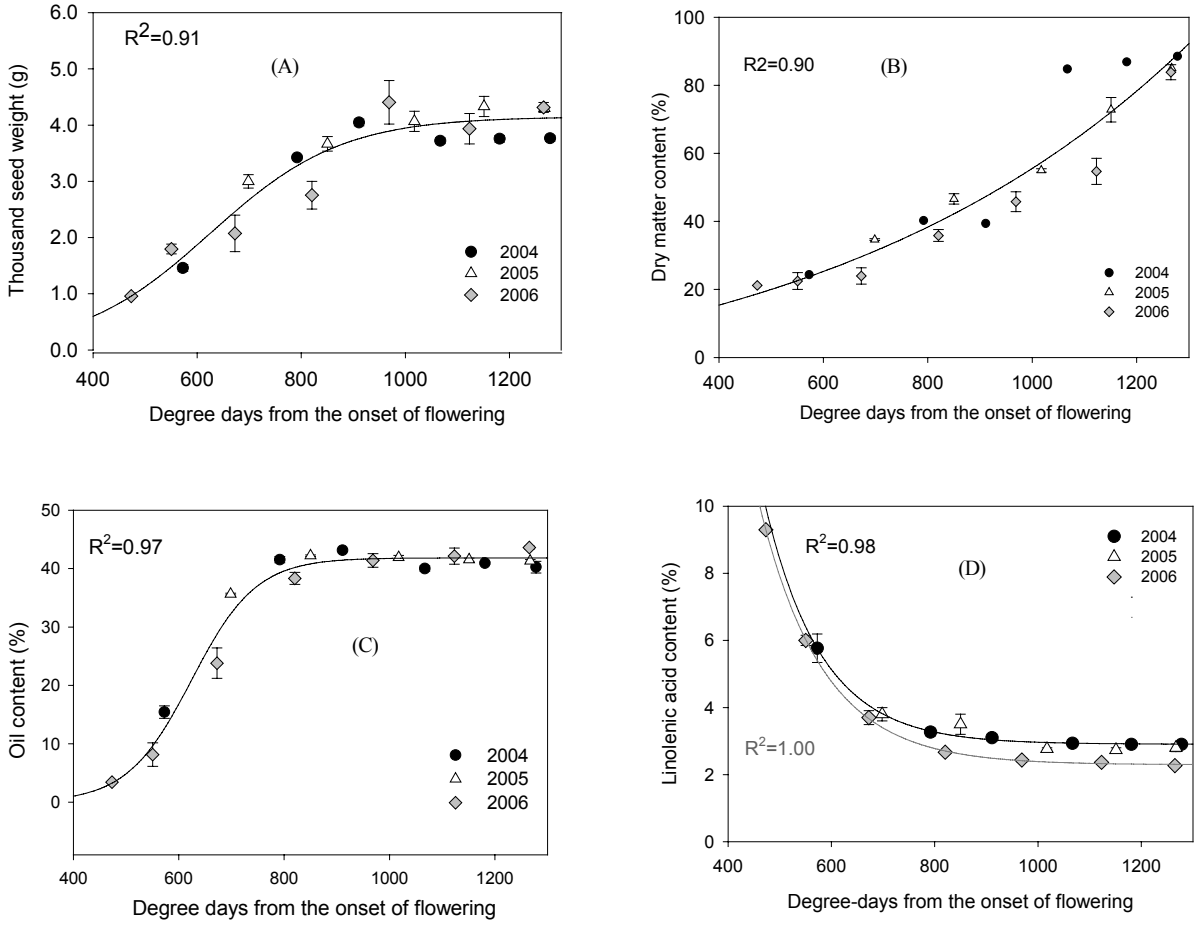

Fig 1. Changes in rapeseed seed components from the beginning of grain filling to maturity. A. Thousand seed weight (g). B. Dry matter content (%). C. Oil content (%). D. Linolenic acid content (%). Error bars = standard error of the mean, n=3.

Time is expressed in days or in degree-days (base 0°C) after the onset of flowering in order to compare experimental data collected during three years.

Thousand seed weight was about 1.0g at the first harvest, 470 degree-days after the onset of flowering. It increased steadily until 800 degree days and then slowly to reach its final value about 1000 degree days after flowering (Fig 1A). Dry matter content increased exponentially until full maturity (Fig 1B). Oil content increased during the first part of grain filling to reach a maximum content around 900 degree-days after the onset of flowering (Fig 1C). Linolenic acid content was higher at the beginning of grain filling and decreased until maturity (Fig 1D). The major changes in grain weight, oil and linolenic acid contents occurred between 500 and 850 degree-days after the beginning of flowering. No difference in oil content was noticed among the three locations and years. However, the evolution of linolenic acid content between 550 and 850 degree-days was more rapid in 2006 than in 2004 and 2005 and it reached a lower final value at full maturity (2.3% in 2006 as compared to

2.9% and 2.8% in 2004 and 2005, respectively). These results showed that oil production occurs mainly during the period between 550 and 850 degree-days (i.e. 40 to 60 days) after the onset of flowering.

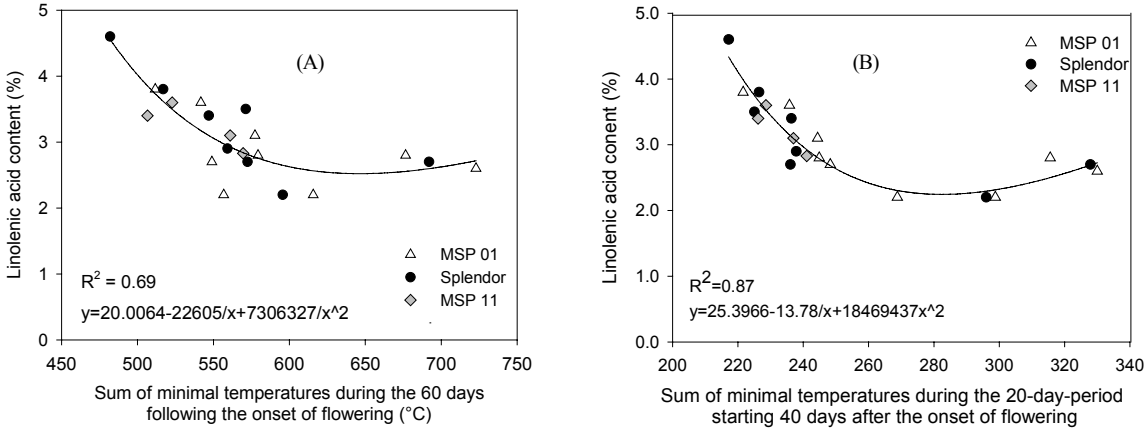

Fig 2. Relationship between the sum of daily minimal temperatures registered during grain filling and final linolenic acid content. A – temperatures registered during a 60-day period starting at the onset of flowering. n=21. B – temperatures registered during a 20-day period starting 40 days after the onset of flowering. n=21.

Temperature impact

The sum of daily minimal temperature measured was negatively correlated with final linolenic acid content (Fig. 2A and 2B). However, the data collected in Switzerland showed that the negative correlation based on a 60-day measurement period was stronger than the one observed by Merrien (2005) for conventional rapeseed. Furthermore, it was possible to improve this relationship using a shorter period of 20 days, starting at the onset of flowering+40 days with an R^2=0.87 (Fig 2B). This showed that the impact of temperature on linolenic acid content was even stronger, during the 20-day period of strong oil synthesis (Fig 1 A and C).

The influence of minimal temperature can be expressed in a different way, considering the number of days with minimal temperature below a "threshold temperature". The best relationship was obtained with a threshold temperature of 13°C (Fig 3).

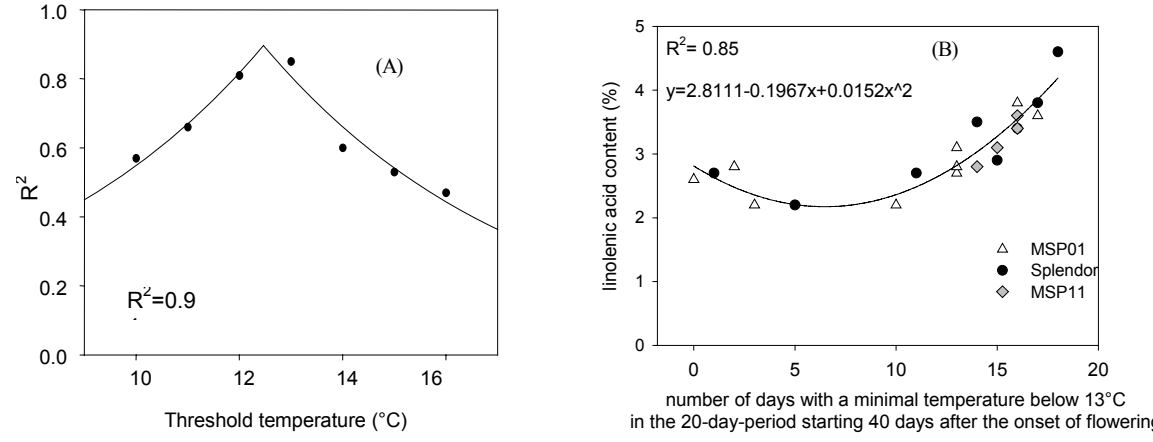

Fig 3. A – determination of the threshold temperature. The highest R2 comes from the relationship between the number of days below a threshold temperature and the final linolenic acid content. B - Relationship between the number of days with a minimal temperature below 13 and the final linolenic acid content in rapeseed seeds. n=21.

The lowest linolenic acid contents were obtained with 3 to 11 days with minimal temperatures below 13°C during the temperature sensitive 20-day period. No difference could be evidenced between the three low-linolenic varieties.

Conclusions

The changes in seed composition during grain filling suggested that oil production and thus fatty acid synthesis mainly occurs between 550 and 850 degree-days after the onset of flowering. This period corresponds in our conditions to a 20-day period starting 40 days after the onset of flowering. The relationship between the sum of temperature recorded during this period and final linolenic acid content showed that in low-linolenic varieties, linolenic acid synthesis is highly influenced by

minimal temperature during grain filling and oil synthesis. As Matsuda & al (2005) noticed for Arabidopsis thaliana, we can make the hypothesis that the activity of at least one of the omega-3 desaturases left in low-linolenic varieties is regulated by temperature.

Taking the impact of temperature on final linolenic acid content into account will make it possible to forecast alpha-linolenic acid content before harvest, or to choose the best production area in order to produce low linolenic oilseed rape.

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References

Deng X. & Scarth R. (1998). Temperature effects on fatty acid composition during development of low-linolenic oilseed rape (Brassica napus L.) JAOCS, 75, 7.

Everard J. (2004). Acides linolénique et oléique: les clés pour diversifier les usages de l'huile de colza, Oléoscope 75: 23-25.

Izquierdo N. & al. (2002). Night temperature affects fatty acid composition in sunflower oil depending on the hybrid and the phonological stage. Field Crop Research, 77: 115-126

Matsuda O. & al. (2005). A temperature-sensitive mechanism that regulates post-translational stability of a plastidial omega-3 fatty acid desaturase (FAD8) in Arabidopsis leaf tissues. Journal of biological chemistry, 280(5) : 3597-3604

Merrien A. (2005). Contribution à l'explication de la variabilité de la teneur en acide linolénique chez le colza d'hiver culture en France. Rencontres annuelles du Cetiom, décembre 2005. 75-90.

Pritchard F. & al. 2000: Environmental effects on seed composition of Victorian canola, Australian Journal of Experimental agriculture, 40: 679-685

Trémolières H. & al. (1977). Effects of light and temperature on fatty acid desaturation during the maturation of rapeseed. Phytochemistry, 17: 685-687

Triboi-Blondel A-M. & Renard M. (1999). Effects of temperature and water stress on fatty acid composition of rapeseed oil. CGIRC. 10th Intetrnational Rapeseed Congress, Canberra, Australia.