Low temperature induced flowering ability in tulip bulbs (Tulipa gesneriana)
Walch, Karin

IMPORTANT NOTE: You are advised to consult the publisher's version (publisher's PDF) if you wish to cite from it. Please check the document version below.

Document Version
Publisher's PDF, also known as Version of record

Publication date:
1997

Link to publication in University of Groningen/UMCG research database

Citation for published version (APA):
The introduction of the tulip by Carolus Clusius was the start of a colourful career of the tulip. The tulip became a highly popular flower, which resulted in the so called tulipomania which raged across Holland between 1634 and 1637. In this period speculants tried to make enormous prices out of tulip bulbs, and many of them were bankrupt after the crash. The tulip remained however popular in Holland and is so still today. The enormous popularity of the tulip resulted in a strong effort of the bulb growers to produce flowering tulips all year round. The main principle of the year-round production is to manipulate the bulbs for the production of either early flowering or retarded flowering tulip bulbs. Manipulation of the flowering date can mostly be achieved by special temperature treatments. The temperature treatment is given after the harvest of the bulbs when the flowerbud has been completely developed. The most commonly used method to produce early flowering tulips is dry storage of the bulbs at 5°C for several weeks. The optimal cold period for *Tulipa gesneriana* cv. red Apeldoorn is 12 weeks at 5°C. The length of the cold period determines the flowering behaviour: when the cold period is too short the tulip shows a stunted stem length and a high percentage of flower abortion, when the cold period is too long the tulip shows a high growth rate with long stems and small flowers. For the production of late flowering tulips the bulbs are stored at -2°C for several months in humid soil in polyethylene bags. Tulips grown from these bulbs are also called “icetulips” because of this treatment. This method however, is expensive and only used when tulips grown by other methods are not available.

There is an extensive empirical knowledge of temperature treatments which give good flowering tulip bulbs. However, the physiological processes occurring during the cold induced development of well flowering tulip bulbs are only partially understood. An important component in plant functioning are plant membranes which normally consist for about 40% of lipids and for about 60% of proteins. Membranes are involved in communication and transport between cells and are an important site for physiological processes. The changes in composition of the different types of lipid molecules often indicate a changed physiological state of the plant.

In this study the role of the lipid composition related to the flowering ability of tulip bulbs was investigated in order to determine the physiological significance of membranes in the flowering process. The results were also used to determine which changes in membrane lipid composition could be used for a test for the flowering ability of tulip bulbs.
The effect on the hardening ability of several bulb organs after exposing tulip bulbs to low temperature was investigated with the experiments described in Chapter 2. Besides hardening, low temperature induced a better flowering ability in the tulip bulbs. Therefore, the low temperature induced changes in the membrane lipids which lead to better flowering ability were studied. The optimal cold period for good flowering ability for bulbs from *Tulipa gesneriana* cv. red Apeldoorn is 12 weeks at 5°C. During storage at 5°C and at 17°C (control) a possible relationship between the development of good flowering ability and (i) total lipid content (Chapter 3), (ii) fatty acid composition (Chapter 3) and (iii) phospholipid composition (Chapter 5) of the shoot, basal plate and scales was studied. Bulbs stored at -2°C also give good flowering tulips therefore, the fatty acid composition (Chapter 3) and the phospholipid composition (Chapter 8) of the shoot was determined in order to detect if similar changes were responsible for the development of good flowering ability. During dry storage the shoot continues to grow, although slowly. As a consequence, the developmental stage of the shoot could determine the changes in the lipid content and composition. Therefore, (i) the total lipid content (Chapter 4), (ii) the fatty acid composition (Chapter 4) and (iii) the phospholipid composition (Chapter 6) of the shoot were determined. Continued cold storage at 5°C induced a higher growth rate of the shoot after planting. In order to find a relation between the membrane lipid changes and the flowering ability of the bulb, (i) the total lipid content (Chapter 7 and 8) and (ii) the phospholipid composition (Chapter 7 and 8) of the shoot were studied during the first nine days of growth after different storage periods. The effect of gibberellin on the phospholipid content of the shoot is described in Chapter 9.

The results from Chapter 2 showed that dry stored tulip bulbs possessed a level of cold hardiness as low as -11°C, although flowering quality could be affected when bulbs were stored below -2°C. Furthermore, it was shown that the hardiness of bulbs was hardly affected by low temperature since the difference in survival of cells from cooled and uncooled bulbs at low temperatures was minimal. A high fluidity of membranes is often seen as an indication for hardening. The fluidity of the shoot membranes, as measured with Fourier Transform infrared spectroscopy (FTIR), showed that membranes of cooled bulbs were still fluid at a lower temperature than membranes from uncooled bulbs indicating hardening in cold stored tulip bulbs. A relation of the membrane fluidity with hardening can however be argued since hardening was not found when the survival of the tissue was determined. Changes in membrane fluidity might therefore not necessarily be involved in hardening since storage at low temperature also induces processes which lead to good flowering ability of the bulb. Development of the shoot and basal plate during storage indicated metabolic activity of the bulb. Shoot growth in uncooled bulbs was more rapid compared to the shoot growth of cooled bulbs. The total lipid content of the basal plate and the scales showed almost no reaction to low temperature whereas the total lipid content in the shoot increased during cold storage. A direct relation between the total lipid content of the shoot and the duration of cold storage was not found.
The relation between the fatty acid composition and development of good flowering was studied. The fatty acid composition of the basal plate and the scales hardly differed between the fatty acid composition of cooled and uncooled bulbs and was therefore not directly related with better flowering ability or adaptation to low temperature. The shoot of the dry stored bulbs however, showed a clear change in fatty acid composition as a reaction to low temperature storage: linoleic acid (18:2) increased and palmitic acid (16:0), oleic acid (18:1) and linolenic acid (18:3) decreased. The positive correlation between the ratio 18:2/18:3 and better flowering ability could only be observed when the bulbs were transferred to 5°C before half October. After this date the ratio 18:2/18:3 remained more or less at the level of uncooled bulbs. Since bulbs stored at -2°C showed a ratio of 18:2/18:3 similar to uncooled bulbs, this ratio was probably not directly related to good flowering ability. The decrease of 18:1, which was observed in the shoot during storage at 5°C, was independent from the developmental stage of the shoot and also occurred during storage at -2°C. This suggests that the decrease in 18:1 could be involved in the development of good flowering ability.

The total phospholipid content in both the basal plate and the scales reached a maximum value after 10-12 weeks of storage at 5°C, implying a relationship between the total phospholipid content in the basal plate and scales and the flowering ability of bulbs. A fast increase in the total phospholipid content shortly after a low temperature treatment is given is often attributed to low temperature adaptation. The phospholipid content in the shoot increased mainly in the first week during storage at 5°C, which suggested adaptation to low temperature rather than a relation with flowering ability. The suggestion of low temperature adaptation was sustained as the level of phospholipids also increased independent from the developmental stage at the start of the experiment and in bulbs stored at -2°C.

After storage, during the first nine days of growth, the phospholipid content showed a negative correlation with shoot growth; the faster the shoot grew after storage at 5°C or -2°C, the faster the phospholipid content decreased. Since the rate of shoot growth was correlated with a better flowering ability, the rate of decrease in phospholipids could be involved in the process for good flowering ability. A net breakdown of the phospholipids in the shoot seemed unlikely since the absolute phospholipid content per shoot increased during shoot growth. The decrease in phospholipid content (per mg lipid) was probably caused by an increased level of other lipid groups, e.g. galactolipids.

A possible involvement of the total phospholipid content on the flowering ability was not observed in the basal plate and scales since the phospholipid composition of the basal plate and the scales hardly changed under influence of low temperature. Contrary to the results from the basal plate and the scales, the phospholipid composition of the shoot changed as a result of low temperature storage and was almost independent from the date on which the bulbs were transferred to 5°C. The percentage of phosphatidyl choline (PC) and phosphatidyl ethanolamine (PE) in the shoot showed a fast decrease respectively increase during the first week of storage at 5°C implying a relation with low temperature adaptation rather than with flowering ability. Similar changes in PC and PE were also found in the shoot of bulbs stored at -2°C, also indicating involvement in low temperature adaptation. After optimal storage for
Summary
good flowering at low temperature, PC increased in the first 3 days of growth followed by a decrease. Similar changes in PC were observed in bulbs previously cooled longer than 12 weeks at 5°C and in bulbs previously cooled at -2°C. The reaction of PC in the shoot during growth was comparable with changes of PC during gibberellic acid (GA) induced growth in rye. A GA-induced change in PC in the shoot of tulip bulbs was however not likely, since no specific reaction of GA on the phospholipid composition was found during growth. Except after 12 weeks of storage at 5°C, when compared to bulbs treated without GA, a slight decrease in PC was determined. The low response of PC to GA suggested that the sensitivity to GA is more important than the amount of GA. Involvement of PC in the flowering process can therefore not be excluded in tulip bulbs. The gradual decrease in the percentage of PE in the shoot during the first nine days of growth in bulbs stored for more than 6 weeks at 5°C or at -2°C could not be related to flowering ability.

During and after storage at 5°C and at -2°C both PC and PE changed in a similar way. The changes in phosphatidyl inositol (PI) and phosphatidyl glycerol (PG) however, depended on the storage temperature. Storage at 5°C resulted in a gradual decrease respectively increase of PI and PG during the first five weeks of storage independent of the developmental stage of the shoot at the start of the experiment. A relation with low temperature adaptation seemed unlikely since hardly any change of PI and PG was observed in bulbs stored at -2°C. The turnover of PI and PG was not negatively affected by freezing temperatures in other plants and therefore it was unlikely that the turnover of PI and PG in the tulip bulbs was negatively affected during storage at -2°C. The difference in behaviour of PI and PG at 5°C and at -2°C might be explained by the changes in phospholipid composition during growth. The percentage of PI and PG hardly changed during growth when the bulbs were previously stored at 5°C. However, when the bulbs were previously stored at -2°C the percentage of PI and PG rapidly decreased respectively increased during the first three days after growth to the level detected in bulbs previously stored at 5°C. This indicated that the percentage of PI and PG in the shoot during growth could be essential for good flowering ability of the bulb. The importance of PI and PG in the development of good flowering ability at low temperatures was sustained by the observation that the percentage of PI and PG on the sixth day after planting correlated either negative (PI) or positive (PG) with the flowering ability in bulbs.

In conclusion, the lipid content and composition of the membranes in the shoot, basal plate and scales in tulip bulbs changes under influence of low storage temperatures. A relation between some of the investigated lipids and the flowering ability possibly occurred in the bulb organs. A direct relation with the changes in these lipids and the flowering ability is difficult to establish since the membrane lipid composition can also change as an adaptation to low temperatures. Therefore, further research is necessary to analyse the direct involvement between the changes in the lipid composition, as determined in this study, and the development of good flowering ability in tulip bulbs.