Effects of salinity on fatty acid composition of canola (Brassica napus L.)

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Abstract

The growth and fatty acid composition of canola (Brassica napus L.) at different salinities (0, 50, 100, 150 and 200 mM NaCl) were analyzed. The results showed that salt treatment reduced significantly the plant growth and the total fatty acids (TFA) content by 25% at 200 mM NaCl. Oleic, linoleic, palmitic and α-linolenic acid were the major fatty acids. Moreover, the polyunsaturated fatty acids decreased, while the monounsaturated ones increased with respect to increasing salinity.

Key words: Canola, Brassica napus L., salinity, fatty acid composition.

Introduction

Salinization of land has been received more attention because of salinity increasing progressively throughout the world 1-2. It is estimated that about a third part of world’s irrigation lands and half of the lands in semiarid and coastal regions are affected by salinization and 10 Mha irrigated lands are abandoned annually because of excessive salinity. Of the 1.5 Bha that is cultivated, about 5% is affected by salt 4. Hence, it should be found an effective way to use saline lands by the cultivation of tolerant cultivars or other agrotechiques. All salts can affect plant growth, but not all inhibit growth. Plants subjected to high salinity levels undergo various physiological and biochemical changes leading to numerous changes in the structure and function of cell membranes. Fatty acids are among the most prominent constituents of cell membrane lipids which play a fundamental role in cell permeability 5. Increasing soil salinity levels strongly influence the essential oil biosynthesis 6. Nevertheless, investigations dealing with the effect of this stress on essential oil production are scarce. With increasing salt stress, both increase and decrease in chlorophyll content and changes in mineral composition of different plant organs have been reported 8-10. However, effect on lipid metabolism of oilseed crops has not been adequately studied 11-14. Canola (Brassica napus L.) is an important oilseed crop of Northern Iran, where decreased oil content with salinity has been reported 15, but information regarding the effect of salinity on quality aspects of canola oil is very scarce. So the present investigation was carried out to examine the effect of different levels of salinity on lipid components of canola seeds.

Material and Methods

Seeds of canola cultivar SLM046 was planted into the pots filled with perlite and vermiculite (1:1). The pots were kept in a glasshouse with natural sunlight and temperature ranging between 30±3°C and 20±3°C in the day and at night, respectively. The experiment consisted of 5 treatments with 5 salinity concentrations as NaCl (0, 50, 100, 150 and 200 mM), each treatment being replicated four times. Modified Hoagland’s solution 16 was used to prepare the nutrient solution. At the end of the experiment two plants were harvested from each pot. After recording their fresh biomass, they were oven-dried at 70°C for two days and dry biomass was recorded. Plant growth was determined by measuring the dry weight.

Lipid extraction: The cell biomass was determined by centrifuging the cell suspension, washing twice with distilled water and freeze-drying. Total lipid was extracted with chloroform: methanol (2:1) for 1 h. The extracted lipid was centrifuged to obtain a clear supernatant, and anhydrous sodium sulphate was added to remove any residual moisture. The solvent was removed by flushing with nitrogen, and the total lipid content was estimated by gravimetric method 17.

Fatty acid analyses: The dried cells were suspended in 0.4 M methanolic KOH at 60°C for 1 h, and fatty acids were esterified at 60°C for 1 h in BF3/methanol (14% w/w) reagent. The esterified fatty acids were extracted with n-hexane and analyzed by Varian-CP-3800 EC equipped with a FID and a DB-23 capillary column (50 mm×0.25 mm). Nitrogen was used as carrier gas. Initial column temperature was set at 175°C for 1 min, raised to 250°C at 15°C/min and maintained 23 min. The injector was kept at 250°C with an injection volume of 1 µl under splittes mode. The FID detector temperature was set at 260°C. Fatty acid methyl esters (FAMEs) were identified by comparison with the retention time of authentic standards (Sigma Co., USA). The quantities of individual FAMEs were estimated from the peak areas on the chromatogram using nonadecanoic acid (19:0) (Sigma Co., USA) as an internal standard.

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**Statistical analyses:** All extractions and determinations were conducted in triplicate. Data are expressed as mean ± SD. The means were compared by using the one-way and multivariate analysis of variance (ANOVA) followed by Duncan’s multiple range tests. The differences between individual means were deemed to be significant at P < 0.05. All analyses were performed by using the SAS software.

**Results and Discussion**

As shown in (Fig.1), a significant (P<0.05) decrease of plant dry biomass was observed following the application of the different NaCl levels. The application of 50 mM caused a light drop in dry biomass production, while concentration of 100 and 150 mM NaCl reduced plant biomass production by 25 and 35%, respectively, as compared to control plants. A consistent decrease by 55% in dry biomass was observed at 200 mM NaCl. Salt stress, like many other abiotic stresses, inhibits plant growth and is one cause of growth rate reduction. Under salt stress inadequate photosynthesis is owing due to stomatal closure and consequently reduced plant biomass production by 25 and 35%, respectively (Table 1). Varying levels of NaCl (50 to 150 mM) in the growth medium decreased the SFA and the PUFA and increased the MUFA fraction due to gadoleic acid increase. It is noteworthy to mention a possible stimulation effect of these NaCl concentrations on the eicosanoyl desaturase activity 20. Besides, it is worth to highlight the relatively opposite evolution in the percentages of oleic and linoleic acids at all NaCl levels in comparison to the control. Furthermore, the DBI (double-bound indices), assessed in order to evaluate the unsaturation degree of the fatty acids pool, decreased in comparison to the control (Table 1). In summary, low (50 mM), moderate (100 mM) and high (200 mM) NaCl levels decreased the degree of fatty acid unsaturation. This fact could be explained by a possible reduction of the desaturase activity which appeared as an adaptive feature to salinity 21, since some plants could be protected against the oxidative effects of salt ions through restructuring membranes with less polyunsaturated fatty acids 22. Moreover, this low unsaturation degree limited the membrane fluidity 23-25 and restricted permeability to Na and Cl ions 26, 27.

**Figure 1.** Dry biomass of canola influenced by different NaCl levels.

**Table 1.** Fatty acids percentage, DBI and TFA content of canola seeds under different NaCl concentrations.

<table>
<thead>
<tr>
<th>Fatty acid</th>
<th>0</th>
<th>50</th>
<th>100</th>
<th>150</th>
<th>200</th>
</tr>
</thead>
<tbody>
<tr>
<td>C 16:0 (palmitic)</td>
<td>5.21 ± 0.06</td>
<td>6.34 ± 0.06</td>
<td>5.18 ± 0.04</td>
<td>4.89 ± 0.07</td>
<td>3.65 ± 0.04</td>
</tr>
<tr>
<td>C 16:1 (palmitoleic)</td>
<td>0.58 ± 0.02</td>
<td>0.68 ± 0.02</td>
<td>0.55 ± 0.01</td>
<td>0.52 ± 0.02</td>
<td>0.48 ± 0.01</td>
</tr>
<tr>
<td>C 18 (stearic)</td>
<td>2.34 ± 0.07</td>
<td>2.88 ± 0.03</td>
<td>2.18 ± 0.03</td>
<td>2.13 ± 0.03</td>
<td>1.98 ± 0.03</td>
</tr>
<tr>
<td>C 18:1 n-9(oleic)</td>
<td>66.79 ± 0.09</td>
<td>62.48 ± 0.3</td>
<td>63.82 ± 0.3</td>
<td>62.94 ± 0.3</td>
<td>63.18 ± 0.4</td>
</tr>
<tr>
<td>C 18:2 n-6 (linoleic)</td>
<td>16.58 ± 12</td>
<td>15.49 ± 1</td>
<td>18.26 ± 0.1</td>
<td>19.34 ± 0.1</td>
<td>20.24 ± 0.1</td>
</tr>
<tr>
<td>C 18:3 n-3 (α–linolenic)</td>
<td>6.48 ± 0.02</td>
<td>7.36 ± 0.04</td>
<td>7.88 ± 0.08</td>
<td>7.96 ± 0.09</td>
<td>8.11 ± 0.08</td>
</tr>
<tr>
<td>C 20:0 (arachidic)</td>
<td>0.76 ± 0.04</td>
<td>0.88 ± 0.02</td>
<td>0.96 ± 0.01</td>
<td>0.99 ± 0.04</td>
<td>1.10 ± 0.02</td>
</tr>
<tr>
<td>C 20:1 n-9 (gadoleic)</td>
<td>1.26 ± 0.06</td>
<td>2.89 ± 0.01</td>
<td>1.18 ± 0.02</td>
<td>1.27 ± 0.05</td>
<td>1.26 ± 0.03</td>
</tr>
<tr>
<td>SFA</td>
<td>19.7 ± 0.08</td>
<td>19.8 ± 0.8</td>
<td>19.6 ± 0.07</td>
<td>19.2 ± 0.09</td>
<td>19.36 ± 0.09</td>
</tr>
<tr>
<td>MUFA</td>
<td>21.6 ± 0.09</td>
<td>24.36 ± 0.2</td>
<td>25.26 ± 0.06</td>
<td>24.66 ± 0.2</td>
<td>25.18 ± 0.11</td>
</tr>
<tr>
<td>PUFA</td>
<td>59.4 ± 0.32</td>
<td>56.36 ± 0.04</td>
<td>57.32 ± 0.2</td>
<td>57.88 ± 0.3</td>
<td>58.3 ± 0.22</td>
</tr>
<tr>
<td>DBI</td>
<td>2.66 ± 0.05</td>
<td>2.26 ± 0.03</td>
<td>2.32 ± 0.04</td>
<td>2.22 ± 0.04</td>
<td>2.46 ± 0.06</td>
</tr>
<tr>
<td>TFA</td>
<td>18.9 ± 0.03</td>
<td>18.34 ± 0.08</td>
<td>18.26 ± 0.09</td>
<td>16.98 ± 0.3</td>
<td>14.21 ± 0.08</td>
</tr>
</tbody>
</table>

Values (Means of three replicates ± SD) (SFA saturated fatty acids; MUFA monounsaturated fatty acids; PUFA polyunsaturated fatty acids; DBI double bound index). Values of standard error of mean are given after.
Conclusions

The application of NaCl concentrations reduced the plant growth and caused a subsequent gradual decrease in TFA content. Moreover, a reduction in PUFA in favour of MUFA was noted. This fact could be considered as one of the aspects of canola (cv. SLM046) adaptation in saline conditions. In summary, the reduction in plant growth and total fatty acid content by the salt constraint may be a result of a new pattern of resource partitioning providing more carbon skeletons for terpene biosynthesis and accumulation.

References