Influence of dietary linseed on fatty acid composition of pig muscle and adipose tissue

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Introduction

- Meat consumption
  - industrialised countries ~ 90 kg per capita annually
  - fast growth in developing countries
  - pork – most frequently consumed meat worldwide

- Meat - significant source of fat in the diet
  - > total fat intake
  - > intake of saturated fatty acids (SFA)
  - > cholesterol intake
Introduction

- High fat, SFA and cholesterol intake
  - aetiology of today’s most frequent diseases, such as cardiovascular disease

- PUFA and MUFA in diet
  - Preventive role of polyunsaturated (PUFA) and monounsaturated (MUFA) fatty acids
Introduction

- **Nutritional recommendations** *(WHO/FAO 2003)*
  - fat intake: max 15 - 30% of total diet energy
  - max 10% of energy intake from SFA
  - 6 -10% from polyunsaturated (PUFA)
  - 10 - 15% of monounsaturated (MUFA)
  - cholesterol < 300mg/day

- **Optimal P/S ratio ≥ 0.4** *(Dept. of Health, 1994)*
Introduction

• Opposing effects of n-6 and n-3 PUFA:
  ➢ linoleic acid (LA, 18:2n-6; cereals, vegetable oils...):

  ![Linoleic acid structure]

  ➢ alpha-linolenic acid (ALA, 18:3n-3; green leaves, oils from linseed and rapeseed):

  ![Alpha-linolenic acid structure]

  ➢ essential for mammals
Introduction

- In organism longer PUFA are metabolized:
  - from LA: arachidonic (AA,20:4n-6; eggs, meat)
  - from ALA: eicosapentaenoic (EPA,20:5n-3; fish oil),
    docosahexaenoic (DHA,22:6n-3; fish oil)
  - in the membrane phospholipids

- C20 PUFA - precursors for eicosanoides:
  - mediators of physiological processes in tissues
    (blood clotting, inflammation…)
Introduction

- Different effects of n-6 and n-3 derived eicosanoids:
  - from n-3: more anti-inflammatory and inhibitory
  - from n-6: more pro-inflammatory and more pro-active in other disease-promoting effects

- Nutritional recommendations:
  - optimal n-6/n-3 ratio in the diet 4 - 5 and less

( WHO/FAO 2003; Dept. of Health, 1994 )
• Composition of fats in pig meat

- SFA and MUFA - *de novo* synthesis in the body
- PUFA primarily from diet, absorbed unchanged
- Industrial pork: low in n-3, high in n-6 due to high proportion of LA in cereal-based feeds
- **P/S ratio** ≤ 0.4 nutritionally positive
- n-6/n-3 ratio 7-10 and higher! nutritionally unfavorable!
• Modification of pork fats by feeding

- primary goal - increase in n-3, reduction of n-6/n-3
- long-chain n-3 rich feeds e.g. fish oil (EPA, DHA)
- plant sources rich in ALA e.g. rapeseed, canola
- linseed (ALA ~ 50%, n-6/n-3 = 0.2 -0.3)
- > PUFA - possible negative side effects (reduced oxidative stability of fats and off-flavors)
- increased dietary levels of antioxidants (vitamin E)
Aim of the work

• To investigate the influence of dietary linseed on fatty acid composition of pig muscle and adipose tissue

• To achieve better pork composition related to recommended n-6/n-3 PUFA ratio for human diet
Material and methods

* Pigs and diet:*
  - commercial crossbreds (IHAN farm, Slovenia)
  - usual fattening from ~ 25 to 105 kg
  - 36 experimental pigs fed 3 % linseed diet + alpha-tocopherol 97.5 mg/kg
  - 6 conventionally fed farm pigs as a control
  - linseed diet: ALA = 19%, n-6/n-3 = 2.4
  - control diet: ALA = 3.7%, n-6/n-3 = 15.4
**Material and methods**

- **Sampling and analyses:**
  - meat quality (*m.longissimus dorsi* pH, colour $L^*$)
  - longissimus muscle and back fat samples (last rib)
  - chemical analyses (EMONA RCP, Slovenia)
  - fatty acid composition (GLC method)
    
    *(in situ) trans-esterification, Park and Goins, 1994)*
  - Rancimat test for back fat oxidative stability
    
    *(Läubli et al., 1998)*
  - Student t-test and correlations *(SAS, 1999)*
Meat quality of longissimus muscle of pigs fed linseed or control diet

<table>
<thead>
<tr>
<th></th>
<th>Linseed</th>
<th>Control</th>
<th>Sign.</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH(_1)</td>
<td>6.26 ± 0.28</td>
<td>6.58 ± 0.18</td>
<td>*</td>
</tr>
<tr>
<td>pH(_2)</td>
<td>5.66 ± 0.14</td>
<td>5.56 ± 0.26</td>
<td>ns</td>
</tr>
<tr>
<td>Lightness ((L^*))</td>
<td>55.13 ± 3.84</td>
<td>55.82 ± 4.95</td>
<td>ns</td>
</tr>
</tbody>
</table>

mean ± standard deviation

*P ≤ 0.05; **P ≤ 0.01; ns–not significant (P > 0.05)
Fatty acid composition (g/100g of total FA) of longissimus muscle of pigs fed linseed or control diet

<table>
<thead>
<tr>
<th>Fatty acid</th>
<th>Linseed</th>
<th>Control</th>
<th>Sign.</th>
</tr>
</thead>
<tbody>
<tr>
<td>14:0 myristic</td>
<td>1.30 ± 0.19</td>
<td>1.38 ± 0.71</td>
<td>ns</td>
</tr>
<tr>
<td>16:0 palmitic</td>
<td>22.65 ± 0.10</td>
<td>24.57 ± 0.82</td>
<td>***</td>
</tr>
<tr>
<td>16:1 palmitoleic</td>
<td>3.15 ± 0.61</td>
<td>4.43 ± 0.63</td>
<td>***</td>
</tr>
<tr>
<td>18:0 stearic</td>
<td>12.01 ± 0.54</td>
<td>9.45 ± 1.46</td>
<td>**</td>
</tr>
<tr>
<td>18:1 oleic</td>
<td>36.95 ± 3.72</td>
<td>38.84 ± 2.54</td>
<td>ns</td>
</tr>
<tr>
<td>18:2 n-6 LA</td>
<td>12.47 ± 2.64</td>
<td>13.03 ± 2.83</td>
<td>ns</td>
</tr>
<tr>
<td>18:3 n-3 ALA</td>
<td>2.48 ± 0.34</td>
<td>0.56 ± 0.14</td>
<td>***</td>
</tr>
<tr>
<td>20:4 n-6 AA</td>
<td>2.79 ± 0.88</td>
<td>3.90 ± 1.21</td>
<td>**</td>
</tr>
<tr>
<td>20:5 n-3 EPA</td>
<td>0.94 ± 0.29</td>
<td>0.28 ± 0.10</td>
<td>***</td>
</tr>
<tr>
<td>22:6 n-3 DHA</td>
<td>0.25 ± 0.15</td>
<td>0.33 ± 0.16</td>
<td>ns</td>
</tr>
</tbody>
</table>

Mean ± standard deviation

*P ≤ 0.05; **P ≤ 0.01; ***P ≤ 0.001; ns – not significant (P > 0.05)
Results – muscle tissue

Proportions of total saturated, monounsaturated and poly-unsaturated fatty acids in logissimus muscle of pigs fed control or linseed diet

Control diet
- SFA: 36.0%
- MUFA: 43.7%
- PUFA: 19.9%

Linseed diet
- SFA: 36.7%
- MUFA: 40.6%
- PUFA: 21.8%

P/S ~ 0.6
Results – muscle tissue

Proportion of total n-6 i n-3 PUFA in longissimus muscle of pigs fed control or linseed diet

Control diet
- 18.1 (Total n-6 PUFA)
- 1.8 (Total n-3 PUFA)

Linseed diet
- 16.4 (Total n-6 PUFA)
- 5.4 (Total n-3 PUFA)

***P ≤ 0.001

Total n-3 increased 3 x
n-6/n-3 reduced from ~10 on 3
### Results – adipose tissue

Fatty acid composition (g/100g of total FA) of back fat of pigs fed linseed or control diet

<table>
<thead>
<tr>
<th>Fatty acid</th>
<th>Linseed mean ± standard deviation</th>
<th>Control mean ± standard deviation</th>
<th>Sign.</th>
</tr>
</thead>
<tbody>
<tr>
<td>14:0 myristic</td>
<td>1.35 ± 0.16</td>
<td>1.42 ± 0.12</td>
<td>ns</td>
</tr>
<tr>
<td>16:0 palmitic</td>
<td>21.61 ± 2.10</td>
<td>22.58 ± 0.86</td>
<td>***</td>
</tr>
<tr>
<td>16:1 palmitoleic</td>
<td>2.30 ± 0.36</td>
<td>2.26 ± 0.43</td>
<td>ns</td>
</tr>
<tr>
<td>18:0 stearic</td>
<td>10.87 ± 2.13</td>
<td>14.15 ± 1.00</td>
<td>***</td>
</tr>
<tr>
<td>18:1 oleic</td>
<td>39.17 ± 2.97</td>
<td>38.78 ± 1.09</td>
<td>ns</td>
</tr>
<tr>
<td>18:2 n-6 LA</td>
<td>14.35 ± 2.42</td>
<td>13.56 ± 1.64</td>
<td>ns</td>
</tr>
<tr>
<td>18:3 n-3 ALA</td>
<td>6.61 ± 0.97</td>
<td>1.01 ± 0.15</td>
<td>***</td>
</tr>
<tr>
<td>20:4 n-6 AA</td>
<td>0.18 ± 0.04</td>
<td>0.25 ± 0.04</td>
<td>***</td>
</tr>
<tr>
<td>20:5 n-3 EPA</td>
<td>0.10 ± 0.02</td>
<td>0.03 ± 0.01</td>
<td>***</td>
</tr>
<tr>
<td>22:6 n-3 DHA</td>
<td>0.05 ± 0.03</td>
<td>0.09 ± 0.01</td>
<td>**</td>
</tr>
</tbody>
</table>

*P≤0.05; **P≤0.01; ***P≤0.001; ns–not significant (P>0.05)
Results – adipose tissue

Proportions of total saturated, monounsaturated and poly-unsaturated fatty acids in back fat of pigs fed control or linseed diet

- **Control**:
  - SFA: 41.8%
  - MUFA: 41.8%
  - PUFA: 16.0%

- **Linseed**:
  - SFA: 34.7%
  - MUFA: 42.2%
  - PUFA: 23.0%

***P ≤ 0.001

P/S raised from ~ 0.4 to 0.7
Results – adipose tissue

Proportion of total n-6 and n-3 PUFA in back fat of pigs fed control or linseed diet

Control

- Total n-6: 14.6%
- Total n-3: 1.4%

Linseed

- Total n-6: 15.2%
- Total n-3: 7.8%

Total n-3 increased ~ 5.5 x

n-6/n-3 reduced from ~ 10.3 to 1.9

***P≤0.001
## Results – back fat oxidative stability

Oxidative stability of fats in back fat of pigs fed control or experimental diet

<table>
<thead>
<tr>
<th>Rancimat test</th>
<th>Control</th>
<th>Linseed</th>
<th>Sign.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Induction time (h)</td>
<td>4.38 ± 0.51</td>
<td>2.38 ± 0.58</td>
<td>***</td>
</tr>
</tbody>
</table>

mean ± standard deviation; *** P ≤ 0.001

Reduced ~ 46 %!
Results – back fat oxidative stability

Relation between oxidative stability of backfat and proportion of total PUFA, ALA and total n-3 PUFA in pigs fed linseed diet

ALA \( (r = -0.40) \), Total n-3 \( (r = -0.39) \), Total PUFA \( (r = -0.35) \)

\( P \leq 0.05 \)
Conclusions

- Linseed - effective feed for the increase of n-3 PUFA content of pork
- Feeding 3% linseed diet to fatlings lowered n-6/n-3 ratio in muscle and adipose tissue and improved nutritional quality of pork fats
- To improve oxidative stability of fats higher dietary levels of vitamin E are needed
- Including linseed in commercial pig feeds - a practical way for the rising population intake of n-3 PUFA through consumption of nutritionally improved pork
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THANK YOU FOR THE ATTENTION!

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