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effect of UVB exposure

Jäpelt, Rie Bak; Silvestro, Daniele; Smedsgaard, Jørn; Jensen, Poul-Erik; Jakobsen, Jette

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Vitamin D₃ in plants – effect of UVB exposure

Rie Bak Jæpelt, Daniele Silvestro, Jørn Smedsgaard, Poul-Erik Jensen, Jette Jakobsen

aNational Food Institute, Technical University of Denmark, e-mail: riba@food.dtu.dk
bDepartment of Plant Biology and Biotechnology, University of Copenhagen

Introduction
As a surprise for many not only vitamin D₂, but also vitamin D₃ can be found in plants. Vitamin D₃ is formed in the skin of vertebrates by exposure to UVB light (Fig. 1). The synthesis of vitamin D₃ in plants is on the other hand unresolved and contradicting results regarding the dependence of UVB-light has been presented (1,2,3). The aim of this study was, therefore, to investigate vitamin D₃ synthesis and metabolism in plants and how it changes upon UVB-exposure. Most work on vitamin D₃ in plants has been done with non-selective methods such as bioassays, but this study utilizes LC-MS/MS with derivatization to improve sensitivity and selectivity.

Fig. 1. Biosynthesis of vitamin D₃ from 7-dehydrocholesterol. UVB exposure of 7-dehydrocholesterol breaks the B-ring to form previtamin D₃, which undergoes thermally induced rearrangement to vitamin D₃

Material
Plants were grown in growth chambers with or without UVB light. Three Solanaceous species were used:

- Solanum glaucophyllum Desf. (waxy leaf nightshade)
- Solanum lycopersicum L. (tomato)
- Capsicum annuum L. (pepper)

Method
The leaves were harvested, freeze-dried and saponified over-night. The vitamin D₃ metabolites were extracted from the non-saponified matter followed by solid phase clean-up. Further clean-up was performed with semi-preparative HPLC. Fractions of vitamin D₃, 25-hydroxy vitamin D₃ and 1,25-dihydroxy vitamin D₃ were collected separately and derivatized with 4-Phenyl-1,2,4-triazoline-3,5-dione (PTAD) to increase sensitivity in ESI-MS/MS. The derivatized extracts were subsequently analyzed by LC-ESI-MS/MS. The vitamin D₃ metabolites were quantified using their deuterated form as internal standard.

Results
Vitamin D₃ was identified in S. glaucophyllum, S. lycopersicum and C. annuum (Table 1). The vitamin D₃ content in the UVB-exposed plants was 18-64 times higher than for the not UVB-exposed plants. 25-hydroxy vitamin D₃ was only identified in the UVB-exposed plants, whereas 1,25-dihydroxy vitamin D₃ only was found in UVB-exposed S. glaucophyllum (Table 1).

Table 1. Content of vitamin D₃, 25-hydroxy vitamin D₃ and 1,25-dihydroxy vitamin D₃ in plants grown with (+UVB) or without (-UVB) UVB light

<table>
<thead>
<tr>
<th>Plant</th>
<th>D₃ (ng per gram dry weight)</th>
<th>25OHD₃ (ng per gram dry weight)</th>
<th>1,25(OH)₂D₃ (ng per gram dry weight)</th>
</tr>
</thead>
<tbody>
<tr>
<td>S. glaucophyllum (+UVB)</td>
<td>200</td>
<td>31</td>
<td>32</td>
</tr>
<tr>
<td>S. glaucophyllum (-UVB)</td>
<td>3.2</td>
<td>0.8</td>
<td>&lt;0.1</td>
</tr>
<tr>
<td>S. lycopersicum (+UVB)</td>
<td>100</td>
<td>4.3</td>
<td>&lt;0.1</td>
</tr>
<tr>
<td>S. lycopersicum (-UVB)</td>
<td>1.7</td>
<td>&lt;0.02</td>
<td>&lt;0.1</td>
</tr>
<tr>
<td>C. annuum (+UVB)</td>
<td>2.9</td>
<td>0.5</td>
<td>&lt;0.1</td>
</tr>
<tr>
<td>C. annuum (-UVB)</td>
<td>&lt;0.02</td>
<td>&lt;0.02</td>
<td>&lt;0.1</td>
</tr>
</tbody>
</table>

Conclusion
It is remarkable that the leaves of the Solanaceous family contain high amounts of vitamin D₃ bearing in mind that the fruits from, e.g. tomato is an important food for humans. Thus, the potential of plants as a vitamin D₃ source exists. This study demonstrates that both UVB-dependent and independent pathways for biosynthesis of vitamin D₃ exist in plants.

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We acknowledge The Danish Ministry of Food, Agriculture and Fisheries, Directorate for Food, Fisheries and Agri Business (3304-FVFP-07-774-02) and Technical University of Denmark for financial support. We would also like to thank Astrid Kvindebjerg for technical assistance.

References