

also help understand how the pollutants act on the organisms. The present findings clearly show that the starfish sperm bioassay based on the effect of chemicals on O<sub>2</sub> consumption is more sensitive than the sea urchin fertilization and cleavage assays Kobayashi described. Using sea urchin sperm, Kobayashi<sup>5,10</sup> found that fertilization was reduced by 50% at about 1.8 ppm and 0.35 ppm for KCN and HgCl<sub>2</sub> respectively, whereas in the present starfish sperm assay 50% reduction in O<sub>2</sub> consumption occurred at about 0.07 ppm and 0.3 ppm respectively. Therefore, sperm assay based on O<sub>2</sub> consumption was much more sensitive than that based on fertilization for KCN (25 times) and equally sensitive for HgCl<sub>2</sub>. Because most animals could be artificially induced to spawn (e.g. sea urchins) or ejaculate (e.g. bull) use of in vitro sperm toxicity assay will not only be quick and cheap but also alleviate the unnecessary suffering of the test animals.

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### Distribution of 4-methoxy-3-indolylmethyl-glucosinolate (4-methoxy-glucobrassicin) in Brassicaceae

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**Summary.** 4-methoxy-3-indolylmethyl-glucosinolate was detectable by HPLC-methods in a representative profile of members of the brassicaceae. In many species it proved to be the main glucosinolate present in young seedlings.

**Key words.** Brassicaceae; indole-glucosinolate.

	A	B	C
<b>Alysseae</b>			
<i>Alyssoides utriculata</i> (L.) Med.	6	40	n.d.
<i>Ptilotrichum purpureum</i> (Lag. Rodr.) Boiss.	10	52	n.d.
<b>Arabideae</b>			
<i>Arabis alpina</i> L.	n.d.	11	n.d.
<i>Cardamine chenopodiifolia</i> Pers.	44	40	n.d.
<b>Brassicaceae</b>			
<i>Brassica oleracea</i> L.	76	38	71
<i>Sinapis alba</i> L.	47	64	n.d.
<b>Drabeae</b>			
<i>Draba aurea</i> Vahl	n.d.	11	n.d.
<i>Draba fladnizensis</i> Wulf.	n.d.	n.d.	10
<i>Draba incana</i> L.	n.d.	3	3
<b>Hesperideae</b>			
<i>Erysimum comatum</i>	8	18	6
<i>Erysimum perovskianum</i> Fisch. et Mey.	31	62	n.d.
<b>Lepidieae</b>			
<i>Lepidium graminifolium</i> L.	675	25	84
<i>Lepidium sativum</i> L.	84	190	51
<i>Biscutella laevigata</i> L.	67	31	18
<i>Aethionema coridifolium</i>	n.d.	43	22
<i>Aethionema grandifolium</i> Boiss. et Hohenack.	10	11	4
<i>Thlaspi arvense</i> L.	10	51	50
<i>Teesdalia nudicaulis</i> (L.) Br.	49	69	5
<i>Cochlearia arctica</i> Schlechtend.	8	50	8
<i>Cochlearia officinalis</i> L.	44	22	172
<b>Lunarieae</b>			
<i>Lunaria annua</i> L.	47	n.d.	n.d.
<i>Peltaria turcmena</i> Lipsky	22	50	50
<b>Matthioleae</b>			
<i>Matthiola incana</i> (L.) Br.	14	15	3
<b>Sisymbrieae</b>			
<i>Camelina alyssum</i> (Mill.) Thell.	107	21	28
<i>Camelina sativa</i> (L.) Crantz	3	8	7

Contents of desulfo-indole-glucosinolates in seedlings of different species of brassicaceae. A.: ds-glucobrassicin, B.: ds-4-methoxy-glucobrassicin, C.: ds-neoglucobrassicin. ( $\mu\text{g}$  ds-glucobrassicin-equivalents/g fresh wt)

With newly developed methods for the HPLC-analysis of indole-glucosinolates, 4-methoxy-3-indolyl-methyl-glucosinolate has been identified as the main mustard oil glucoside not only in seeds of cabbage and rape<sup>1-3</sup>, but also in seedlings of *Brassica oleracea* and *Isatis tinctoria*. In all TLC-systems used so far for the separation of indole-glucosinolates, 4-methoxy-3-indolyl-methyl-glucosinolate co-chromatographs with glucobrassicin. This could have been the reason for the failure to detect it in other glucosinolate-synthesizing plant species. Therefore it seemed worthwhile to re-examine different taxonomic groups of the family Brassicaceae for the existence of this new secondary indole compound using HPLC-separation of the desulfoglucosinolates. 10-15-day-old seedlings (0.5-1 g fresh wt), grown in continuous white light (10 Wm<sup>-2</sup>; 21  $\pm$  0.5 °C), were extracted with boiling methanol and analyzed for indole-glucosinolates according to the methods of Götz and Schraudolf<sup>4</sup>. Since crystallized 4-methoxy-3-indolylmethyl-glucosinolate was not available, the composition of the indole-glucosinolates in the species analyzed are presented in the table as glucobrassicin-equivalents. These data furnish the proof that 4-methoxy-glucobrassicin is not only a common secondary plant product in most members of the Brassicaceae but, moreover, is very often the predominating glucosinolate in young seedlings of plants of this family. The data also demonstrate that a broad spectrum of indole-ring substituting enzymes is active in Brassicales, as well as the low specificity of the indole-glucosinolate synthesizing enzyme complex with regard to the substitution pattern of the indole ring, as previously postulated by Götz and Schraudolf<sup>4</sup>.

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