

Molecules of interest

Distribution of isoflavonoids in non-leguminous taxa – An update

Zuzana Mackova, Radka Koblovská, Oldrich Lapcik *

Department of Chemistry of Natural Compounds, Faculty of Food and Biochemical Technology, Institute of Chemical Technology in Prague, Technická 5, 166 28 Praha 6, Czech Republic

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Abstract

Common emphasis of the fact that isoflavonoids are characteristic metabolites of leguminous plants sometimes leads to overlooking that the presence of isoflavonoids has been reported in several dozen other families. The spectrum of isoflavonoid producing taxa includes the representatives of four classes of multicellular plants, namely the Bryopsida, the Pinopsida, the Magnoliopsida and the Liliopsida. A review, recently published by Reynaud et al. [Reynaud, J., Guilet D., Terreux R., Lussignol M., Walchshofer N., 2005. Isoflavonoids in non-leguminous families: an update. *Nat. Prod. Rep.* 22, 504–515], provided listing of 164 isoflavonoids altogether reported in 31 non-leguminous angiosperm families. In this contribution we complement the abovementioned inventory bringing the references on further 17 isoflavonoid producing families and on additional 49 isoflavonoids reported to occur in non-leguminous plants.

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Keywords: Isoflavone; Pterocarpane; Rotenoid; Non-leguminous**1. Introduction**

Isoflavonoids draw the attention of specialists from multiple branches of science – from chemotaxonomy (Aparecida et al., 1987) and plant physiology (Dakora and Phillips, 1996) to medicine and nutrition (Cornwell et al., 2004). Common emphasis of the fact that isoflavonoids are characteristic metabolites of leguminous plants sometimes leads to overlooking another reality – that the presence of isoflavonoids has been reported in several dozen other families. The spectrum of isoflavonoid producing taxa includes the representatives of four classes of multicellular plants, namely the Bryopsida, the Pinopsida, the Magnoliopsida and the Liliopsida (Reynaud et al., 2005). In the majority of cases, these reports represent sporadic identification of one or a few compounds in one or a few species falling into the respective family. Some of them have been published in journals not excerpted in generally used databases (e.g., WOS, PubMed and ScienceDirect) and thus

might have been missed by a considerable number of researchers. Certain useful data may also be hidden in the results of research originally not intended as phytochemical – e.g., in screening of food sources of phytoestrogens (Mazur and Adlercreutz, 1998; Mazur et al., 1998; Mazur, 2000). The concise and well-written review by Reynaud et al. (2005) provides an important contribution to this area, enumerating altogether 164 isoflavonoids reported in 31 non-leguminous angiosperm families, three gymnosperm families possessing 15 structures and 3 isoflavonoids found in one Bryophyte (Table 1). Coincidentally, a short time before this review has been published, we started working on our own contribution intended to cover identical field. Not intending to duplicate the work of those who have preceded us we still would like to complement it. In addition to the taxa listed by Reynaud et al. (2005), we bring references to other 4 monocot and 13 dicot families reported to produce isoflavonoids (Table 2), including 33 isoflavones, 7 pterocarpanes and 9 rotenoids, which were not mentioned in the review (Table 3 and Fig. 1). We would also like to add a few comments concerning the occurrence of isoflavonoids in beer (Rosenblum et al., 1992; Lapcik et al., 1998)

* Corresponding author. Tel.: +420 220 443 240; fax: +420 220 444 422.
E-mail address: oldrich.lapcik@vscht.cz (O. Lapcik).

Table 1
Occurrence of isoflavonoids in non-legumes: families listed in Reynaud et al. (2005)

<i>Bryopsida</i>
Bryaceae
<i>Gymnosperms</i>
Araucariaceae, Cupressaceae, Podocarpaceae
<i>Monocotyledons</i>
Eriocaulaceae, Iridaceae, Liliaceae, Poaceae, Stemonaceae, Zingiberaceae
<i>Dicotyledons</i>
Amaranthaceae, Apocynaceae, Asteraceae, Bombacaceae, Caryophyllaceae, Celastraceae, Chenopodiaceae, Clusiaceae, Cucurbitaceae, Euphorbiaceae, Malvaceae, Menispermaceae, Moraceae, Myricaceae, Myristicaceae, Nyctaginaceae, Ochnaceae, Papaveraceae, Polygalaceae, Polygonaceae, Rosaceae, Rutaceae, Scrophulariaceae, Solanaceae, Vitaceae

and bourbon (Gavaler et al., 1987), which was doubted a little in the above mentioned review.

2. Isoflavonoid producing families not included in previous reviews

2.1. Monocots

In addition to the six monocot families listed by Reynaud et al. (2005), isoflavones have been reported in two *Juncus* species (Juncaceae) (Abdel-Mogib, 2001) and furthermore in *Aloe vera* (Asphodelaceae) (Saxena and Sangeeta, 2000) and *Smilax glabra* (Smilacaceae) (Yi et al., 1998). Certain confusion exists in the literature concerning taxonomic classification of the last two species. The genus *Aloe* has been classified to the Alooidae subfamily of the Aspho-

delaceae (Watson and Dallwitz, 1992) or to the Liliaceae (Gutterman and Chauser-Volfson, 2000), alternatively the Aloeaceae has been treated as an independent family (<http://plants.usda.gov/>, 5th September 2005). The genus *Smilax* has been repeatedly assigned to the Liliaceae as well (Dua et al., 2005; Guo et al., 2004). Most recently, Maver et al. (2005) reported bioassay-guided isolation of six isoflavones with interesting fungicidal activities from the stem of *Eriophorum scheuzeri* (Cyperaceae).

2.2. Dicots

Besides the 25 non-leguminous dicot families mentioned by Reynaud et al. (2005), we have found the literature references on other 13 families reported to contain isoflavones (Table 2), which appear reliable to us, and several others, which should “be taken with a pinch of salt”. The latter will be mentioned under miscellaneous. Two references were only available as abstracts because full texts of those journals were inaccessible. We will state this fact for each particular case.

3. Isoflavonoids in beer and bourbon and in plants of which they are made

In 1992, Rosenblum reported the detection of daidzein and genistein in beer using GC–MS. Six years later, we have confirmed his findings and also observed 4'-methoxy derivatives of the above mentioned isoflavones, i.e., formononetin and biochanin A, respectively (Lapcik et al., 1998). The concentrations of individual isoflavones in beer ranged from 0.1 to 15 nmol/L and their sum barely reached 30 nmol/L. These observations were later verified by others

Table 2
Occurrence of isoflavonoids in non-legumes: families not listed in previous reviews

Family	Species	Number of aglycones	Reference
<i>Monocotyledons</i>			
Asphodelaceae	<i>Aloe vera</i>	1	Saxena and Sangeeta (2000)
Cyperaceae	<i>Eriophorum scheuzeri</i>	6	Maver et al. (2005)
Juncaceae	<i>Juncus acutus</i> , <i>J. rigidus</i>	1	Abdel-Mogib (2001)
Smilacaceae	<i>Smilax glabra</i>	1	Yi et al. (1998)
<i>Dicotyledons</i>			
Apiaceae	<i>Bupleurum scorzonnerifolium</i> , <i>B. chinense</i> , <i>Carum carvi</i>	4	Tan et al. (1998), Liang et al. (2000), and Mazur and Adlercreutz (1998)
Asclepiadaceae	<i>Sarcobolus globosus</i>	12	Wangensteen et al. (2004, 2005)
Brassicaceae	<i>Arabidopsis thaliana</i> , <i>Brassica oleracea</i> , <i>Lepidium sativa</i>	6	Lapcik et al. (2005), Davidova (2002), and Mazur and Adlercreutz (1998)
Connaraceae	<i>Cnestis ferruginea</i>	1	Parvez and Rahman (1992)
Convolvulaceae	<i>Erycibe expansa</i>	20	Matsuda et al. (2004)
Crassulaceae	<i>Sedum alfredi</i>	1	Men (1986)
Erythroxylaceae	<i>Erythroxylum ulei</i> , <i>E. australe</i>	8	Johnson and Schmidt (1999, 2004)
Melastomataceae	<i>Henriettella fascicularis</i> ,	1	Calderon et al. (2002)
Myrtaceae	<i>Acca sellowiana</i> , <i>Psidium guajava</i> , <i>P. littorale</i>	6	Lapcik et al. (2005)
Nymphaeaceae	<i>Nymphaea ampla</i> , <i>N. pulchella</i>	1	Marquina et al. (2005)
Sapotaceae	<i>Madhuca latifolia</i>	2	Siddiqui et al. (2004)
Urticaceae	<i>Pouzolzia indica</i>	1	Sayeed et al. (2003)
Verbenaceae	<i>Premna microphylla</i>	1	Zhong and Wang (2002)

Table 3
Occurrence of isoflavonoids in non-legumes: structures not listed in previous reviews

Isoflavonoid	Family	Reference
2-Hydroxyorobol (2,5,7,3',4'-pentahydroxyisoflavone)	Erythroxylaceae	Johnson and Schmidt (1999)
2-Methyldihydrogenistein (5,7,4'-trihydroxy-2-methylisoflavanone)	Erythroxylaceae	Johnson and Schmidt (1999)
2-Methyldihydroorobol (5,7,3',4'-tetrahydroxy-2-methylisoflavanone)	Erythroxylaceae	Johnson and Schmidt (1999)
3',4',5,7-Tetrahydroxyisoflavone	Nymphaeaceae	Marquina et al. (2005)
4',5,7-Trihydroxy-6,8-dimethylisoflavone	Melastomataceae	Calderon et al. (2002)
5,2',4'-Trihydroxy-2'',2''-dimethylpyrano(3'',4'',6,7) isoflavone 1	Cyperaceae	Maver et al. (2005)
5,2',4'-Trihydroxy-2'',2''-dimethylpyrano(3'',4'',7,8) isoflavone 2	Cyperaceae	Maver et al. (2005)
5,2',4'-Trihydroxy-7-methoxy-3'-methylisoflavone	Cyperaceae	Maver et al. (2005)
5,2',4'-Trihydroxy-7-methoxyisoflavone	Cyperaceae	Maver et al. (2005)
5,4'-Dihydroxy-7,2'-dimethoxyisoflavone	Cyperaceae	Maver et al. (2005)
5,4'-Dihydroxy-7,2'-dimethoxy-3'-methylisoflavone	Cyperaceae	Maver et al. (2005)
5,7,4'-Trihydroxy-3'-methoxyisoflavone	Convolvulaceae	Matsuda et al. (2004)
5-Hydroxy-3',5'-dimethoxyisoflavone	Crassulaceae	Men (1986)
5-Hydroxy-6-methoxy dihydropseudobaptisin 3	Erythroxylaceae	Johnson and Schmidt (2004)
5-Methoxy-4'-hydroxy-2'',2''-dimethylpyrano(3'',4'',7,8) isoflavone 4	Urticaceae	Sayed et al. (2003)
6,3'-Dihydroxy-7-methoxy-4',5'-methylenedioxyisoflavone 5	Verbenaceae	Zhong and Wang (2002)
7,2'-Dihydroxy-4',5'-dimethoxyisoflavone	Convolvulaceae	Matsuda et al. (2004)
7,3',4'-Trimethoxyisoflavone	Convolvulaceae	Matsuda et al. (2004)
7,5 -Dihydroxy-6,3',4'-trimethoxyisoflavone	Asphodeliaceae	Saxena and Sangeeta (2000)
Afrormosin (7-hydroxy-6,4'-dimethoxyisoflavone)	Connaraceae	Parvez and Rahman (1992)
Alpinumisoflavone 6	Convolvulaceae	Matsuda et al. (2004)
Baptigenin (2,5,7,3',4',5'-hexahydroxyisoflavone)	Erythroxylaceae	Johnson and Schmidt (1999)
Barbigerone 7	Asclepiadaceae	Wangensteen et al. (2004, 2005)
Calycosin (7,3'-dihydroxy-4'-methoxyisoflavone)	Convolvulaceae	Matsuda et al. (2004)
Derrone 8	Convolvulaceae	Matsuda et al. (2004)
Dihydroorobol (5,7,3',4'-tetrahydroxyisoflavanone)	Erythroxylaceae	Johnson and Schmidt (2004)
Erycibenin A 9	Convolvulaceae	Matsuda et al. (2004)
Erycibenin B 10	Convolvulaceae	Matsuda et al. (2004)
Erythrinin B 11	Convolvulaceae	Matsuda et al. (2004)
Madhusalmone 12	Sapotaceae	Siddiqui et al. (2004)
Madhushazone 13	Sapotaceae	Siddiqui et al. (2004)
Puerarin (7,4'-dihydroxyisoflavone 8-C-glucopyranoside)	Apiaceae	Liang et al. (2000)
Sarcolobone 14	Asclepiadaceae	Wangensteen et al. (2004, 2005)
P: 11b-Hydroxy-11b,1-dihydromaackiain 15	Convolvulaceae	Matsuda et al. (2004)
P: 11b-Hydroxy-11b,1-dihydromedicarpin 16	Convolvulaceae	Matsuda et al. (2004)
P: Erycibenin C 17	Convolvulaceae	Matsuda et al. (2004)
P: Homopterocarpin (3,9-dimethoxypterocarpan)	Convolvulaceae	Matsuda et al. (2004)
P: Medicarpin (3-hydroxy-9-methoxypterocarpan)	Convolvulaceae	Matsuda et al. (2004)
P: Pterocarpin (3-methoxy-8,9-methylenedioxypterocarpan)	Convolvulaceae	Matsuda et al. (2004)
P: Secundiflorol I (3-hydroxy-8,9-dimethoxypterocarpan)	Convolvulaceae	Matsuda et al. (2004)
R: 11-Hydroxytephrosin 18	Asclepiadaceae	Wangensteen et al. (2004, 2005)
R: 12a-Hydroxyrotenone 19	Asclepiadaceae	Wangensteen et al. (2004, 2005)
R: 12a α -Hydroxyrotenone 20	Asclepiadaceae	Wangensteen et al. (2004, 2005)
R: 12a α -Hydroxydeguelin 21	Asclepiadaceae	Wangensteen et al. (2004, 2005)
R: 13-Homo-13oxa-6a,12a-dehydrodeguelin 22	Asclepiadaceae	Wangensteen et al. (2004, 2005)
R: 6a,12a-Dehydrodeguelin 23	Asclepiadaceae	Wangensteen et al. (2004, 2005)
R: 6a α ,12a α ,12-Hydroxyelliptone 24	Asclepiadaceae	Wangensteen et al. (2004, 2005)
R: Sarcolobin 25	Asclepiadaceae	Wangensteen et al. (2004, 2005)
R: Tephrosin 26	Asclepiadaceae	Wangensteen et al. (2004, 2005)

P, pterocarpane; R, rotenoid.

Except for puerarin only aglycones are listed. Number of compounds would be higher if different glycosides of the same aglycone were counted as individual structures.

(Clarke et al., 2004). As the pilsner type beers are made of barley (*Hordeum vulgare*, Poaceae) or rarely of wheat (*Triticum aestivum*, Poaceae) and of hops (*Humulus lupulus*, Cannabaceae), these plants represent candidate sources of isoflavones. There are several reports on isoflavones in barley, wheat and triticale in the literature based on GC–MS (Mazur and Adlercreutz, 1998; Mazur, 2000) and HPLC–

DAD (Ciepiela et al., 1997). It is worthy of note that the content of daidzein and genistein measured in beer corresponded to that found in barley. To our knowledge, no report has been published on the occurrence of isoflavones in the Cannabaceae so far.

The detection of isoflavonoids in bourbon (Gavaler et al., 1987) remains enigmatic. Even if there were certain

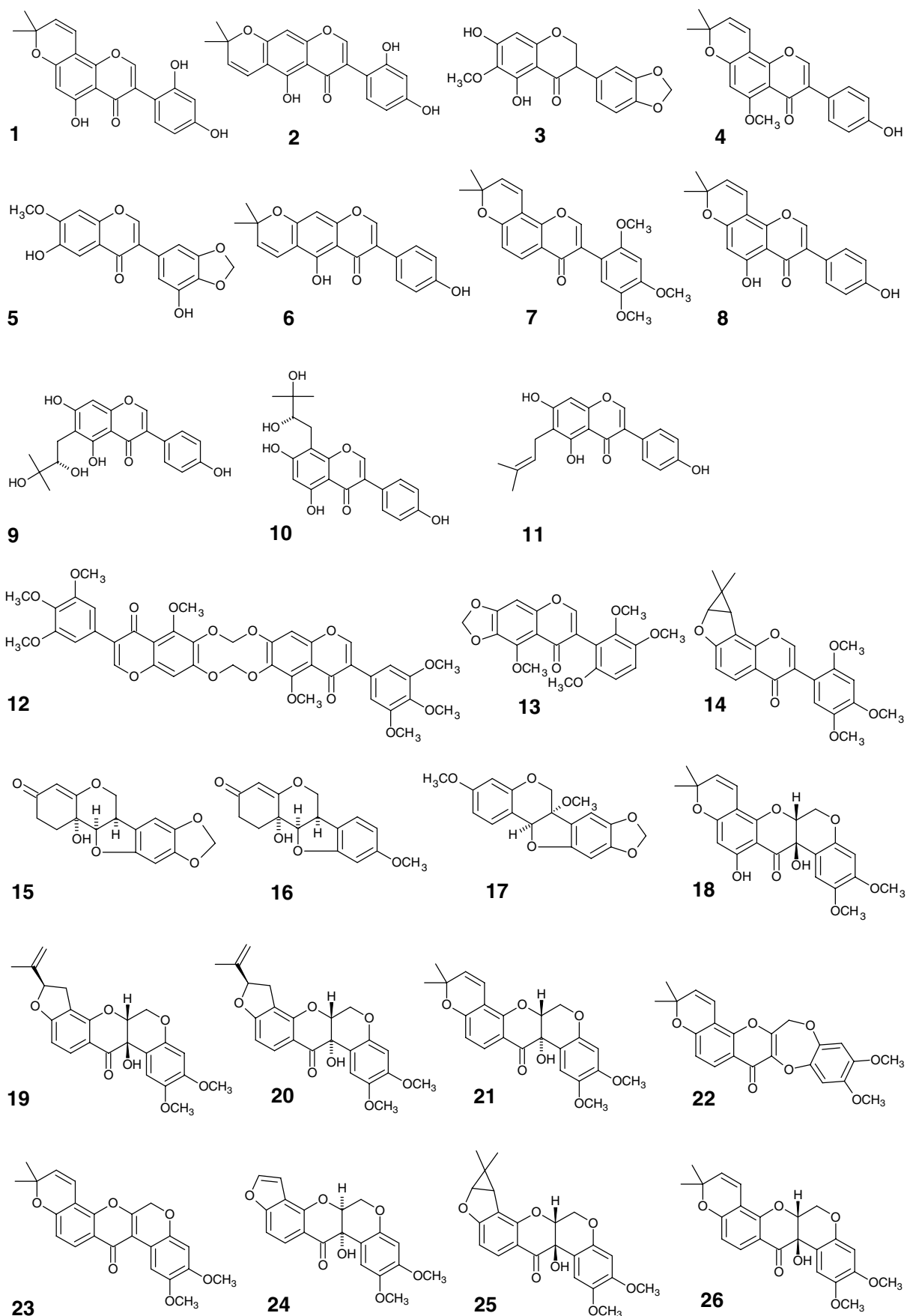


Fig. 1. Isoflavonoids found in non-leguminous taxa: a selection of structures listed in Table 3.

amounts of isoflavones in the maize bowl, they are unlikely to distill together with alcohol and other volatiles. However, bourbon matures in oak barrels, which enrich it with numerous compounds extracted from wood and although no representative of the Fagaceae family has been reported to contain isoflavones yet, we think that oak wood cannot be automatically excluded as a potential source of isoflavones in alcoholic beverages.

4. Miscellaneous

Ferrugin from *Aglaia ferruginea* (Meliaceae) was originally considered to be an isoflavonoid (Dean et al., 1993), later on, its structure was revised to a benzofurane (Mulholland and Naidoo, 1998). The occurrence of 5,7-dimethoxy-4'-hydroxyisoflavone and its 4'-*O*-glucoside was reported in nectar of two *Fremontia* species (Sterculiaceae), but detailed description of the identification was missing (Scogin, 1997). Two articles mentioned the detection of isoflavones in avocado (*Persea americana*, Lauraceae), however in both cases the identification was made by two-dimensional paper chromatography only (Ramirez-Martinez and Bor, 1973; Prabha and Patwardhan, 1980). Mazur detected low levels of daidzein and genistein, sometimes accompanied with the corresponding 4'-methoxyisoflavones, in edible parts of several non-legumes during his extensive HPLC–MS screening for dietary sources of phytoestrogens (Mazur and Adlercreutz, 1998; Mazur et al., 1998; Mazur, 2000). His list included also the representatives of families not known to produce isoflavones by that time, e.g., broccoli, cauliflower and red cabbage (*Brassica oleracea*, Brassicaceae), walnuts (*Juglans nigra*, Juglandaceae), sesame seeds (*Sesamum indicum*, Pedaliaceae), coffee (*Coffea arabica*, Rubiaceae), tea (*Camellia sinensis*, Theaceae), caraway seed (*Carum carvi*, Apiaceae). It is worthy of note that several other species on the list were additional representatives of families only once reported to contain isoflavones, e.g., the Papaveraceae (Jain et al., 1996) and the Poaceae (Casabuono and Pomilio, 1990). Although this research was not aimed at phytochemistry and so the analyzed material was not sufficiently identified from this point of view, it is likely that mono-species raw food materials were classified correctly. Later on, the presence of isoflavones has been verified in the Apiaceae (Tan et al., 1998; Liang et al., 2000) and the Brassicaceae (Davidova, 2002; Lapcik et al., 2006). We have also noted one reference to the role of isoflavones in resistance of *C. arabica* to pathogens, but the type of isoflavone was not clearly stated in the abstract and full text of that paper was unattainable to us (Mazzafera and Magalhaes, 1989).

5. How many isoflavones are there in non-legumes?

Reports on the occurrence of isoflavones result from two different research approaches. The first one primarily

emphasizes the identification of new compounds. The other approach is focused on revelation of new sources of already known substances. While new compounds appear to be attractive per se, publishing an observation of known compounds is often difficult, despite the novelty of the source, without an additional commentary embedding it into wider consequences. Such relationships are apparent in biochemical pathways.

Individual metabolites cannot appear in organisms without biosynthesis (or without uptake – which is unlikely in the case of isoflavones in plants). In other words, the identification of particular metabolite indicates the presence of a whole metabolic pathway leading to that substance, i.e., enzymatic apparatus and the cascade of metabolic precursors, in the respective species. From this point of view, in a plant, in which at least one isoflavonoid has been reliably detected, the occurrence of a whole isoflavonoid pathway may be presumed. Needless to say, the concentrations of individual intermediates may differ by orders of magnitude, depending on the anatomical part and the developmental stage in which they are found. Yet, we may expect them to be detectable using sufficiently sensitive methods. Vice versa, sufficiently sensitive screening for the early metabolites can reveal the presence of the isoflavonoid pathway despite the possibility that the detected compounds does not have to represent major isoflavonoids in the respective species of interest.

The key step at the very beginning of the isoflavonoid metabolic pathway is the oxidation of flavanone connected with the migration of aryl moiety from C2 to C3 (Akashi et al., 1999; Steele et al., 1999). The enzyme responsible for this reaction, designated as isoflavone synthase (IFS), was identified and cloned in several legumes, e.g., *Glycyrrhiza echinata*, *Glycine max*, *Cicer arietinum*, *Lotus japonicus* and in sugar beet (*Beta vulgaris*, Chenopodiaceae) (Akashi et al., 1999; Steele et al., 1999; Overkamp et al., 2000; Shimada et al., 2000; Jung et al., 2000). Native substrates of IFS are two flavanones, namely liquiritigenin and naringenin. The very early metabolites of this pathway are daidzein, genistein, formononetin and biochanin A (Liu and Dixon, 2001). Hence these substances appear to be good candidates for indicators of presence of the isoflavonoid metabolic pathway. Coincidentally, owing to the fact that the same four isoflavones have been attracting the attention of biomedical science for several decades due to their possible impact on health, numerous sensitive and specific methods are available for their analysis (Bennetau-Pelissero et al., 2003; Wu et al., 2004; Umphress et al., 2005). Recently, we have applied specific immunoassays for screening of isoflavonoids in several non-leguminous families, using HPLC–MS as the confirmatory method. This approach revealed whole spectra of isoflavonoids in the Rutaceae (Lapcik et al., 2004), the Myrtaceae (Lapcik et al., 2005) and the Brassicaceae (Lapcik et al., 2006). While the only isoflavonoid described in the Solanaceae up to now is torvanol from *Solanum torvum* (Arthan et al., 2002), our most recent data show, that low levels (from micrograms

to milligrams per kilogram, dry weight) of daidzein and genistein and their methoxy-derivatives and glycosides are regularly found in leaves and flowers of several *Nicotiana* species as well as in *Solanum dulcamara* and *Lycopersicon esculentum* (Macková et al., manuscript in preparation).

6. Conclusion

The occurrence of isoflavonoids has been reported in above fifty families (Tables 1 and 2). This number includes the representatives of both classes of flowering plants, three gymnosperm families (Sethi et al., 1983; Briggs and Cebald, 1963; Fonseca et al., 2000) and one representative of the Bryopsida (Anhut et al., 1984; Geiger et al., 1987). While most of the reports come from the Leguminosae and few others (i.e., Asteraceae, Iridaceae, Chenopodiaceae, Moraceae, Myricaceae, and Rosaceae) in majority of families the knowledge does not exceed one or two reports on one or a few isoflavones in rather limited number of species (Reynaud et al., 2005; Harborne and Baxter, 1999; Tahara and Ibrahim, 1995). We conclude that screening for intermediates of this metabolic pathway could reveal higher number of isoflavonoids in each species where at least one isoflavonoid has been reliably detected, provided that sufficiently sensitive methods are used. In addition, the availability of sophisticated methodology for the detection of simple isoflavones (Bennetau-Pelissero et al., 2003; Wu et al., 2004; Umphress et al., 2005) gives the opportunity to systematically seek out the presence of isoflavonoid pathway in taxa not studied from this point of view so far.

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