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2,3-Dehydroderivatives of Silymarin Flavonolignans: Prospective Natural Compounds for the Prevention of Chronic Diseases †

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Abstract: *Silybum marianum* fruit extract silymarin displays various biological activities, which are attributed mostly to its major component silybin. However, silymarin contain several other isomeric flavonolignans (isosilybin, silychristin, silydianin) and their oxidation products, the 2,3-dehydroflavonolignans (2,3-dehydrosilybin, 2,3-dehydrosilychristin, 2,3-dehydrosilydianin). The latter compounds were found to be 1-2 orders of magnitude more efficient radical scavengers, reducing, chelating, cytoprotective, anti-aging, anti-cancer and anti-angiogenic agents than the parent flavonolignans. Although 2,3-dehydroflavonolignans occur in silymarin as minorities, they seem to be responsible for the majority of the biological activity and therefore have potential for the prevention of chronic diseases.

Keywords: silymarin; flavonolignan; chemoprevention

1. Introduction

Food supplements containing *Silybum marianum* (L.) Gaertn. (Asteraceae) fruit extract, silymarin, are used especially by the elderly population. Silymarin displays many biological activities such as antioxidant, anti-inflammatory, immunomodulatory and hepatoprotective. Depending on the plant cultivar and extraction method used [1], silymarin contains a mixture of structurally closely related flavonolignans (flavonoids with fused lignan part). The main silymarin constituents are silybin A, silybin B, isosilybin A, isosilybin B, silychristin A, silydianin, the flavonol taxifolin (Figure 1) [2] together with minor flavonolignans and approximately 30% of undefined polymeric fraction. The most abundant and hence easily isolated silymarin component [3] is silybin (mixture of diastereomers A and B) and thus, it is considered as the major active principle of silymarin. Therefore, the literature mostly focuses on silybin and ignores all other components. However, other flavonolignans are likely to contribute to, or even be responsible for distinct beneficial effects of silymarin. In particular, minor flavonolignans of the silymarin complex, e.g., 2,3-dehydroflavonolignans, which occur as a result of bio-oxidation in the plant itself or due to oxidation during extraction and processing [4] have been neglected for a long time. The aim of the present study was to evaluate the chemopreventive potential of all silymarin constituents including the minorities and their potential metabolites.

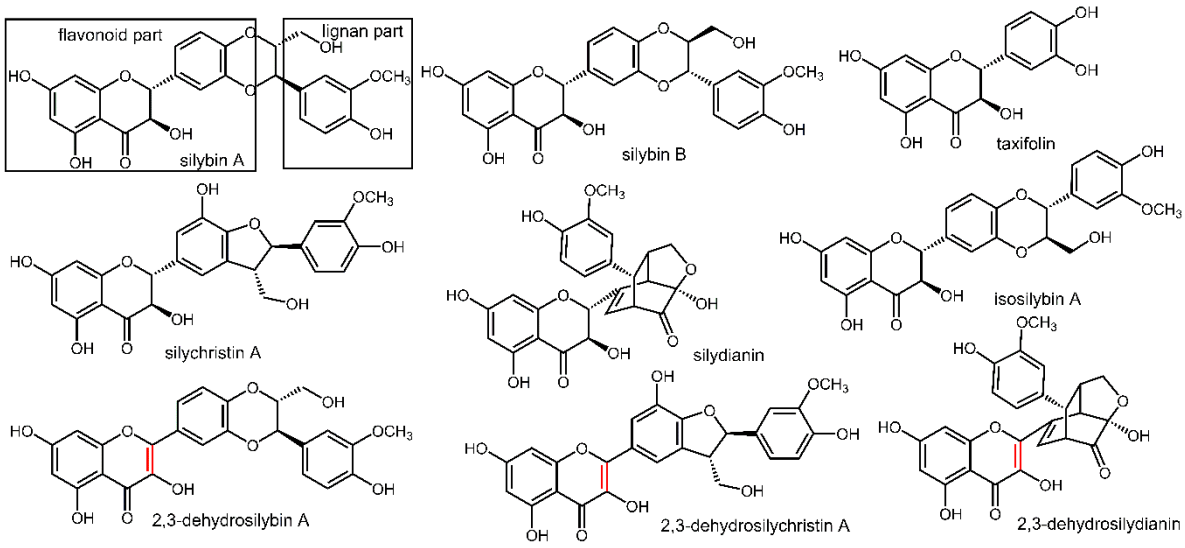

Figure 1. Selected silymarin components.

2. Materials and Methods

Silybin A, silybin B, silychristin A, and silydianin were isolated from silymarin (Liaoning Senrong Pharmaceutical, Panjin, China; batch No. 120501) as described previously [5,6]. 2,3-Dehydrosilybin, 2,3-dehydrosilychristin and 2,3-dehydrosilydianin were prepared from respective flavonolignans by optimized oxidative methods [4,7,8] in the presence of organic bases. 2,3-Dehydroanhydrosilychristin was prepared by treatment of 2,3-dehydrosilychristin by HCl [7]. Sulfated metabolites of (2,3-dehydro)flavonolignans were prepared using arylsulfotransferase from *Desulfitobacterium hafniense* heterologously expressed in *E. coli* [9]. Reducing, radical-scavenging [4], metal chelating [10], cytotoxic [7] and cytoprotective [4] activities of all compounds were measured and compared to those of parent flavonolignans.

3. Results

All the compounds were successfully prepared in multimiligram up to gram amounts, enabling a detailed study of their properties. The 2,3-dehydroflavonolignans proved to be 1-2 orders of magnitude more active radical scavengers, reducing and cytoprotective agents than their parent compounds [4,7,8]. Significant reducing and antioxidant activity remained even after sulfate conjugation of the 2,3-dehydroderivatives [9]. While silybin A, silybin B and silychristin A were quite weak chelators, 2,3-dehydrosilybin was found to be a potent iron and copper chelating agent [10]. Moreover, 2,3-dehydrosilydianin (but not silybin, silychristin, silydianin, 2,3-dehydrosilybin, or 2,3-dehydrosilychristin) was found to activate Nrf2 and upregulate NAD(P)H:quinone oxidoreductase 1 in Hepa1c1c7 cells [11]. 2,3-Dehydrosilybin and its gallates were also more efficient inhibitors of angiogenesis that silybin and silybin derived gallates [12,13]. In a more complex model, 2,3-dehydrosilybin (and especially the A enantiomer) displayed lifespan-extension effect superior to that of silybin, isosilybin, silychristin and silydianin in *Caenorhabditis elegans* [14]. Moreover, 2,3-dehydrosilybin inhibited basal cell carcinoma allograft tumor growth more than silybin in mice [15].

4. Conclusions

Although the 2,3-dehydroflavonolignans occur as minorities in silymarin, their biological activity is superior to that of other silymarin constituents and they therefore may be responsible for certain biological activities of this complex plant extract. These natural compounds have therefore a great potential for the prevention of chronic diseases.

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