Phytomedicine 21 (2013) 98–99

Contents lists available at ScienceDirect

Phytomedicine

journal homepage: www.elsevier.de/phymed

Editorial

# Now *Ginkgo* – 10 years after *Cimicifuga*?

Approximately 10 years ago, after publication of very poorly investigated case reports, a discussion began on a "hepatotoxic risk" associated with the ingestion of *Cimicifuga* preparations. After several years and thorough evaluations by numerous research groups and responsible authorities (e.g. the HMPC) this risk was not substantiated.

A similar discussion has now been started following a publication in March 2013 of a report by the National Toxicology Program (NTP) of the U.S. Department of Health and Human Services on "Toxicology and Carcinogenesis: Studies of *Ginkgo biloba* Extract in Rats and Mice" (NIH Publication No. 13-5920). According to the authors the reason for the experiments was the widespread use of *Ginkgo* products and the content of quercetin in such preparations. From the results of the 2-year carcinogenicity studies it is concluded that the investigated *Ginkgo biloba* extract caused cancers of the thyroid gland in male and female rats and male mice and cancers of the liver in male and female mice. The discrepancies between the results presented in this report and the proof of safety, together with long-standing experiences in Europe with herbal medicinal products (HMPs) containing a specific *Ginkgo* extract (EGb 761) forced ESCOP to examine the report in greater detail, especially with regard to the quality and analytical data.

## Analytical data

In the NTP studies two different lots of an extract of *Ginkgo biloba* leaves from China (extraction solvent not specified) were used, one for method development and the other one for the animal experiments. The experimental part (p. 167) gives the impression that both extracts contain exactly the same quantities of analytes:

- flavonol glycosides (31.2%) composed of 16.71% quercetin, 12.20% kaempferol and 2.37% isorhamnetin glycosides;
- terpene lactones (15.4%) composed of 6.94% bilobalide, 3.74% ginkgolide A and 1.62% ginkgolide B, 3.06% ginkgolide C;
- ginkgolic acids I and II, 9.17 and 1.28 ppm, respectively.

That two different batches of a herbal preparation result in completely congruent amounts of analytes, up to the second decimal place even of ppm, is amazing! The figures of the HPLC-analyses do not identify of the studied lot.

The authors emphasize the comparability of the tested extract with the standardized extract most frequently used in herbal medicinal products in Europe and worldwide (EGb 761). This is simply not correct! The specifications of EGb 761 are as follows:

- ginkgo flavone glycosides (22.0-27.0%);
- terpene lactones (5.0-7.0%), consisting of 2.6-3.2% bilobalide, 2.8-3.4% ginkgolides A, B and C;
- ginkgolic acids less than 5 ppm.

As shown, the extract tested by the NTP contains about one third more flavone glycosides and nearly twice the amount of terpene lactones and ginkgolic acids.

## Data on quality regarding (i) the vehicle, (ii) the long storage of the dose formulations under room temperature, and (iii) the mode of stability testing

Corn oil is frequently used as the vehicle for lipophilic test substances but why this vehicle was used for the polar *Ginkgo* extract is not clear. As expected, this obviously resulted in poor solubility of the large amounts of the extract in the corn oil. Consequently, homogeneity studies were performed and based on the 'satisfying' results, homogeneity was the only parameter examined throughout the storage of the prepared dosage forms. One must argue and underline the fact that during storage over several weeks at room temperature not only changes in the composition of the active ingredients might occur but also reaction between the vehicle and the test item may happen, and this need to be controlled. The description of periodic control of the bulk material (without showing any data), and the statement *Results of these assays were inconclusive due to high amounts of variability in the measurements* cannot be accepted as adequate.

## Administration of pesticides to the animals with the extract and feed

Carbendazime (0.24 ppm) was detected in the *Ginkgo* extract. In the European legal framework on pesticides residues (Regulation 396/2005 and amendments) the limits for this substance in different foods are between 0.01 and 0.2 ppm with very few exemptions only. In addition, all animals received methylchlorpyrifos (on average $0.109 \pm 0.098$ ppm; range 0.020–0.356 ppm) and malathion (on average $0.245 \pm 0.240$ ppm; range 0.020–0.997 ppm) via the feed. The Pharm. Eur. limits for these two pesticides are 0.1 ppm and 1 ppm, respectively. As recent results in mice have shown, synergistic hepatotoxic effects may occur if carbendazime is applied in combination with other pesticides (e.g., imazalil and cypermethrin) (Dikic et al., 2012a,b). Therefore, the question has to be asked if similar synergistic toxic effects may have contributed to the observed

0944-7113/$ – see front matter 
http://dx.doi.org/10.1016/j.phymed.2013.10.001

pathologic findings during in long-term ingestion of carbendazime with methylchlorpyrifos and malathion at such doses.

## A query regarding doses

In the NTP study in all animals a dose-dependent increase of incidence and severity of hepatocellular hypertrophy and focal hepatocellular necrosis was observed. Such effects might be considered as rather unspecific toxic effects at high doses. The recommended doses of HMPs with the standardized *Ginkgo* extract on the European market usually are up to 240 mg/day, equivalent to 4 mg/kg/day for an adult human with a body weight of 60 kg. Thus, the lower dosages administered to mice and rats in the NTP study were, respectively, 50 and 25 times higher than the human dose. Based on the weight/body surface area, a dose of 200 mg/kg in mice corresponds to 600 mg/m$^2$ and, a dose of 100 mg/kg for rats also corresponds to 600 mg/m$^2$. The dose of 4 mg/kg (for a 60 kg adult human receiving 240 mg) corresponds to 148 mg/m$^2$. Accordingly, by comparison, the lower doses administered to the animals were still 4 times higher than the highest recommended human dose.

In summary, the NTP study contains several deficiencies which depreciate the relevance of the findings and relativize the conclusions drawn. ESCOP, in agreement with several other comments of stakeholders in the field of phytotherapy, wants to underline that the data must not be misinterpreted as an indication of a potential risk for humans. In this respect, ESCOP appreciates the thorough assessment of the NTP study by the HMPC and their conclusion which indicates "that at present there is no proof for an increased cancer risk identified for patients taking *Ginkgo* medicinal products at their approved posology" (HMPC 2013).

This highlights yet again, the urgent need of sound proofs of quality for extracts used in food supplements.

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