Toxicology studies with novel supplement formulations

In this guide

148. The increased bioavailability of supplements formulated in novel ways as discussed above may have important toxicological implications. As EFSA (2018) state in their ‘guidance for risk assessment on nanotechnologies’: “If nanoencapsulates function as intended…there will be increased bioavailability (systemic exposure) of the encapsulated material. This represents a potential

concern since health-based guidance values are currently set based on the external rather than the internal dose and may no longer provide an appropriate level of protection to the consumer." HBGVs, therefore, may need reassessing in light of specific formulations and relative bioavailabilities. This also suggests that exposure assessments based on standard formulations may potentially underestimate exposure from novel formulations.

149. In addition to increases in bioavailability, novel supplement formulations may alter toxicological profiles through other toxicokinetic parameters such as alterations in tissue distribution, or via changes in physiology not reported in the studies reviewed above. Furthermore, there may be formulation specific toxicological effects that cannot be extrapolated from toxicology studies using standard formulations of a given compound / supplement. For instance, it is reported that administration of vitamin C may enhance iron absorption which may be of concern for individuals with hemochromatosis or heterozygous for this disorder (EVM, 2008). However, the mechanism of vitamin C enhanced iron absorption occurs through the chelation of ferric iron which increases the solubility of the latter (Lynch and Cook, 1980), an effect which may be less relevant for encapsulated vitamin C formulations which are less able to physically interact with iron (example formulated by the Secretariat). These considerations suggest the need for case-by-case evaluation of specific formulations with respect to their potential toxicological implications beyond effects on bioavailability.

150. An initial review of toxicological effects which may be related to increased bioavailability, or specific to novel formulations of the compounds reviewed above was conducted using a literature search. The full results of this literature search, including the search strings used, number of retrieved results, and brief summaries of *in vitro* and *in vivo* studies are presented in appendix 1. This was not designed as a comprehensive review of toxicological effects related to novel formulations but intended as a preliminary scoping exercise to guide future assessments and/or to identify data gaps.

151. No studies investigating toxicological effects of novel formulations of vitamin C or CBD were identified. This reflects the lack of knowledge regarding the safety of novel formulations in general. Thus, although toxicological data exists on these active compounds, there are significant data gaps as to how these toxicological profiles may be altered when they are formulated in the ways discussed above.

152. Several studies investigating the toxicology of novel curcumin formulations in experimental systems and/or human subjects were retrieved. The majority of these studies investigated the toxicity of novel curcumin formulation in *in vitro* and/or *in vivo* systems, and three studies investigated effects in human subjects. Owing to the scope and aims of the current discussion paper, the *in vitro* and *in vivo* preclinical studies are summarised briefly in appendix 1 and include cytotoxicity assessments in a number of primary cells and cell lines and toxicological studies in rats and mice. A number of these studies might be of interest for inclusion in future discussion papers. The studies in human subjects are summarised below.

Curcumin

Human studies

153. Storka *et al*. (2015) investigated the safety and tolerability of a liposomal curcumin in healthy human subjects in a randomised dose escalation study. Subjects were administered liposomal curcumin intravenously at either 10, 20, 40, 80, 120, 180, 240, 320 or 400 mg/m^2. Because of adverse reactions to mean red blood cellular volume and the formation of echinocytes only two subjects received 400 mg/m^2. Red blood cell echinocyte formation was dose-dependent, detectable at a threshold dose of 120 mg/m^2, without clinical symptoms, transient, and fully recoverable at 6 hours post infusion. Increases in mean red blood cellular volume were observed in two subjects administered 400 mg/m^2 and did not associate with markers of haemolysis but did associate with increased venous serum lactate concentrations (maximum of 3.7 mmol/L versus normal range of ≤ 2.2 mmol/L). Twenty five subjects also experienced at least 1 adverse event; there were 49 adverse events in total, 11 of which were moderate and 38 of which were mild. Based on the dose-dependent, transient, and reversible effects on echinocyte formation and increases in mean red blood cell volume, the authors concluded that “a single intravenous dose of liposomal curcumin is considered safe up to a dose of 120 mg/m^2 when infused over a period of 2 hours.”

154. In another dose escalation study, Greil *et al*. (2018) assessed the safety, tolerability, and efficacy of liposomal curcumin in 32 patients with locally advanced or metastatic cancer. Liposomal curcumin was administered intravenously weekly for 8 weeks, and the dose was increased from 100 mg/m^2 over 8 hours to 300 mg/m^2 over 6 hours. Twenty-six patients successfully completed dose-escalation without dose-limiting toxicity. However, the number of

adverse events related to the treatment increased with doses at 300 mg/m^2. One patient receiving 300 mg/m^2 developed haemolysis, three patients treated with this dose displayed haemoglobin decreases without signs of haemolysis, whilst one patient exhibited definite haemolysis. Out of a total of 143 adverse events, 34 were considered related to the treatment and the remainder to underlying disease. Two of these events, facial oedema, and anaemia, were considered serious. Echinocytes were also observed in one patient. Although adverse events were observed, this was a study population with advanced cancer disease, many of whom had exhausted other lines of treatment and all of whom were taking other medication at the time of the study, and therefore these events may not be generalisable to other populations.

155. It should be noted that the two above studies (Storka *et al*., 2015 and Greil *et al*., 2018) administered liposomal curcumin via intravenous infusion. This may have resulted in plasma levels that would not be achieved by oral dosing alone.

156. Kocher *et al*. (2016) investigated the safety of micellar curcuminoids in moderately hyperlipidaemic individuals. Subjects consumed 294 mg of micellar curcuminoids or placebo per day for 5 weeks. Neither blood lipids, nor markers of inflammation, glucose and iron homeostasis, or liver enzymes differed between curcuminoid and placebo interventions. The authors concluded the intervention was safe.