Statement on the potential risk to human health of turmeric and curcumin supplements

Annex A

In this guide

In this guide

Introduction

1. The information presented in this Annex should be read in conjunction with the main draft statement on the potential risk to human health of turmeric and curcumin supplements. It contains further detailed information on curcuminoids in relation to toxicity covering both animal and human studies.

Curcuminoids and hepatotoxicity: Animal studies

Short-term studies of toxicity

Mice

2. Dietary turmeric powder (0%, 0.2%, 1.0%, 5.0%, equivalent to 0, 400, 2000, 10000 mg/kg bw/day) or ethanolic turmeric extract (ETE, 0%, 0.05 %, 0.25 %, equivalent to 0, 100, 500 mg/kg bw/day) for 14 days, at doses reported to be cancer preventive in model systems, were found to be hepatotoxic in mice. The curcumin contents of the turmeric powder and ETE were not reported. Exposure of mice to dietary turmeric or ETE did not have any significant effect on body weight/ liver weight or liver to body weight ratios. Animals exposed to 0.2 % turmeric showed coagulative necrosis in liver. Livers from mice receiving 1.0 % and 5.0 % turmeric showed extreme degenerative changes with necrosis. Coagulative necrotic foci surrounded by a zone of regeneration were also evident. Similar changes were also seen in animals treated with 0.05 % and 0.25 % ETE. Necrotic changes in liver, the principal effect, was seen in 6/6 animals from 5.0 % turmeric, 3/6 from 1.0 % turmeric and 3/6 animals from 0.2 % turmeric. Similarly, 3/6 animals from 0.05 % as well as 0.25 % ETE also showed these alterations. Liver from mice receiving control diet showed normal ultrastructure. In the liver of the animals receiving 5.0 % or 1.0 % turmeric diet or 0.25 % or 0.05 % ETE, some of the parenchymal cells had round nuclei consisting of clumped or densely marginated chromatin. The cytoplasm consisted of numerous pleomorphic vacuolated mitochondria filled with dense bodies, surrounded by rough or smooth endoplasmic reticulum and free ribosomes, and an increased number of glycogen particles and Golgi complexes with vesicles (Kandarkar et al., 1998).

3. Sub-chronic oral toxicity of turmeric and ETE was studied in female Swiss mice and Wistar rats fed turmeric powder (0, 1 and 5 %) and ethanolic turmeric extract (0, 0.05 and 0.25 %) through the diet for 14 and/or 90 days. The curcuminoid content of the ETE was approximately 98%. The administration of a high dose of turmeric (5 %) for longer duration (90 days) resulted in a significant reduction in body weight gain, alterations in absolute and / or relative liver weights, and hepatotoxicity i.e., focal necrosis or focal necrosis with regeneration both in mice and rats. In mice, lower doses of turmeric (i.e., 0.2 or 1 % for 14 days) also showed hepatotoxicity and mice were found to be more susceptible to turmeric-induced hepatotoxicity than rats (Deshpande et al., 1998).

4. In a 13-week study, groups of 10 male and 10 female B6C3F1 mice were fed diets containing 0, 1000, 5000, 10000, 25000, or 50000 ppm turmeric oleoresin, which were estimated to deliver average daily doses of 0, 150, 750, 1700, 3850, or 7700 mg/kg bw to males and 0, 200, 1000, 1800, 4700 or 9300 mg/kg bw to females (NTP 1993). The major component of the oleoresin was identified as curcumin (79 to 85 %). The percent composition was monitored periodically at the study laboratory with free-acid titration and high-performance

liquid chromatography methods, and no change in composition was observed. There were no deaths attributed to turmeric oleoresin and the final mean body weight gains and final mean body weights of all exposed groups of male and female mice were similar to those of the controls. Feed consumption by exposed male and female mice was similar to that by the controls. Absolute and relative liver weights of male mice that received 5000 ppm and male and female mice that received 10000, 25000 and 50000 ppm were significantly greater than those of the controls. These changes were not considered adverse. Clinical findings in mice included stained fur, and discoloured faeces and urine. According to NTP, there were no biologically significant differences in hematologic, clinical chemistry, or urinalysis parameters, and there were no chemical related histopathologic lesions. JECFA concluded that the no observed adverse effect level (NOAEL) in mice was 7700 and 9280 mg/kg bw/day in males and females, respectively, which were the highest doses tested (cited in EFSA, 2010). The EFSA ANS Panel (2010) agreed with the NOAEL concluded by JECFA.

Rats

5. Liju *et al.* (2013) investigated the acute and sub-chronic toxicity of turmeric essential oil (TEO) from *Curcuma longa* L in male and female Wistar rats. Acute administration of TEO was as a single oral gavage dose of up to 5 g/kg bw, and a 13-week sub-chronic toxicity study was performed at gavage doses of 0, 0.1, 0.25 and 0.5 g/kg bw/day. There were no substance-related mortalities, adverse clinical signs or changes in body weight, water and food consumption during the acute and sub-chronic studies. Indicators of hepatic function such as aspartate aminotransferase (AST), ALT and alkaline phosphatase (ALP) were unchanged in treated animals compared to untreated animals. Oral administration of TEO for 13 weeks did not alter total cholesterol, triglycerides, markers of renal function, serum electrolyte parameters and histopathology of tissues.

6. In a 28-day study, curcumin (purity > 98 %) was administered to male Sprague Dawley rats through the diet at dose levels of 0, 26.1, 84.8, 224,8, 459.7 and 1117.8 mg/kg bw/day. Clinical chemistry did not reveal major signs of liver damage associated with administration of the compound. cDNA microarray experiments were performed on hepatic RNA. Curcumin altered the expression of 12 genes. Three of these were related to peroxisomes (phytanoyl-CoA dioxygenase, enoyl-CoA hydratase; CYP4A3). Increased cyanide insensitive palmitoyl-CoA oxidation was observed. The authors concluded that these data suggest that curcumin is a weak peroxisome proliferator in rats (Stierum et al.,

2008).

7. In a 13-week study, groups of 10 male and 10 female F344/N rats were fed diets containing 0, 1000, 5000, 10000, 25000, or 50000 ppm turmeric oleoresin (79 to 85 % curcumin), estimated to deliver average daily doses of 0, 50, 250, 480, 1300, or 2600 mg/kg bw to males and 0, 60, 300, 550, 1450, or 2800 mg/kg bw to females. All rats survived until the end of the study. The final mean body weight of males receiving 50000 ppm was 5 % lower than that of the controls. Feed consumption by exposed male and female rats was similar to that of controls. The absolute and relative liver weights of female rats and the relative liver weights of male rats receiving ≥ 5000 ppm were significantly greater than those of the controls. According to the NTP, these increases may have been due to mild hepatocellular swelling or hypertrophy. In the clinical chemistry, urinalysis, and hematologic assessment, no differences were observed that were considered by the NTP to be biologically significant. Clinical findings included stained fur, and discoloured faeces and urine of exposed animals. Mild to moderate hyperplasia of the mucosal epithelium was observed in the cecum and colon of male and female rats that received 50000 ppm. The authors did not attempt to identify a NOAEL, as this was a dose range-finding study (NTP 1993).

8. A six-month toxicity study of curcuminoids extracted from the powdered dried rhizome of *Curcuma longa* was performed in six groups of 15 Wistar rats of each sex. The extract was reported to contain 58-67 % curcuminoids. The water control group received 5 ml of water/kg bw/day, while the tragacanth (plant-derived gum) control group received 5 ml of 0.5 % tragacanth suspension/ kg bw/day orally. Three treatment groups were given the suspension of curcuminoids powder at doses of 10, 50 and 250 mg/kg bw/day. The fourth treatment group, or the recovery group, also received 250 mg/kg bw/day of curcuminoids for six months, but two weeks of no curcuminoids treatment elapsed before the time of termination. The exact method of administration was not described. The growth rate of male rats receiving curcuminoids at 50 mg/kg bw/day was significantly higher than that of the tragacanth control group. Curcuminoids did not produce any significant dose-related changes of haematological parameters. In male animals receiving 250 mg/kg bw/day of curcuminoids, actual and relative liver weights and the level of serum alkaline phosphatase (ALP) were significantly higher than those of the two controls, but the ALP level was still within a normal range. There appeared to be a higher incidence of mild degree of liver fatty degeneration and adrenocortical fatty degeneration in this group of animals, however the incidence was not significantly different from that of the two controls. The authors concluded that at higher doses, curcumin may affect the

function and morphology of the liver in a reversible manner. The EFSA ANS Panel (2010) concluded that due to the lack of knowledge on the curcumin content and nature of the extract tested, the study could not be used to assess the safety of curcumin (Chavalittumrong et al., 2002).

9. Majeed *et al*., (2023) performed a battery of toxicological studies with "AC 3®", a BDMC enriched *C. longa* extract, obtained through solvent extraction, standardized to contain a minimum of 85% w/w of total curcuminoids by HPLC and with BDMC in the range of 30 – 35%.

10. An acute toxicity study at a gavage dose of up to 2000 mg/kg bw (i.e., maximum recommended by the test guidelines) did not result in any clinical signs of toxicity in female adult Wistar rats. A subacute 28-day oral toxicity study in males and females was undertaken with low, mid, and high gavage doses of 125, 250, and 500 mg/kg/bw per day. No changes were observed in body weight gain and feed consumption. No treatment-related changes were observed in haematological, biochemical, or urine parameters, nor in organ weights and gross and histopathological examination. A sub chronic oral toxicity study was performed in male and female Wistar rats at gavage doses 125, 250, and 500 mg/kg bw/day for 90 days. The authors state that "administration [of the test substance] showed no mortality and clinical signs of toxicity. No significant difference was observed in feed consumption, body weight, as well as percentage body weight change. Similarly, there were no significant changes in neurological, ophthalmological, haematological, and biochemical parameters as well as urine analysis. The necropsy did not reveal any treatment-related pathological significance. There were no treatment-related changes observed in absolute and relative organ weights. Majeed *et al*., (2023)"

11. Nirvanashetty *et al*., (2022) conducted a series of safety studies on a proprietary oleoresin-based turmeric extract (CURCUGEN®). These studies were designed as a preclinical safety assessment. The test substance was referred to by the authors as "a standardized curcuminoid extract with 50% curcuminoid, 1.5% essential oils, and other constituents of turmeric." In a repeat dose 90-day oral toxicity test the extract was administered to male and female Sprague-Dawley rats at a gavage dose level of 2000 mg/kg bw/day. This study omitted a low and mid dose because, based on their interpretation of the OECD guidelines, the authors argued that "The limit test precludes the need for full study of three dose levels." The authors reported that "There was no treatment-related variation in the mean body weight or percent body weight change with respect to day 1 in the limit dose of 2000 mg/kg/day. No treatment-related changes in feed

consumption were noted in both the main group and recovery group animals." Moreover, there were no opthalmological abnormalities, no treatment-related effects on neurological examination, and "no adverse treatment-related changes in haematology parameters were noted." There were some changes in haematological parameters that the authors considered to not be test item related." There were no treatment-related adverse effects in the other parameters measured, including blood biochemistry, gross necropsy, and histopathology. Overall, the authors concluded that based on the observed results in rats receiving the test item at 2000 mg/kg bw/day by gavage (orally) over 90 consecutive days, a no observed adverse effect level (NOAEL) of 2000 mg/kg bw/day was identified.

Chronic toxicity

Two years study in mice

12. Groups of 60 male and 60 female B6C3F1 mice were fed diets containing 0, 2000, 10000, or 50000 ppm turmeric oleoresin (79 to 85 % curcumin) for 103 weeks, which were estimated to deliver average daily doses of 0, 220, 520, or 6000 mg/kg bw per day to males and 0, 320, 1620, or 8400 mg/kg bw per day to females (NTP 1993). Nine or ten mice from each exposure group were evaluated after 15 months. Survival of exposed male and female mice was similar to that of the controls. The mean body weight of female mice receiving 50000 ppm was slightly lower (up to 12 %) than that of the controls from about week 25. The final mean body weights of males that received 50000 ppm and females that received 10000 and 50000 ppm were significantly lower than those of controls. The final mean body weights of other exposed groups of male and female mice were similar to those of the controls. Feed consumption by exposed male and female mice was similar to that of the controls throughout the study. The absolute and relative liver weights of male and female mice receiving 10000 and 50000 ppm were significantly greater than those of the controls at the 15-month interim evaluation. There were no clinical findings related to toxicity. No biologically significant differences were observed in hematologic parameters. The alkaline phosphatase (ALP) values of male and female mice that received 10000 and 50000 ppm were significantly higher than those of controls at the 15-month interim evaluation. The incidence of thyroid gland follicular cell hyperplasia was statistically significantly increased in high dose female mice. The incidences of hepatocellular adenoma in male and female mice receiving 10000 ppm (the mid dose group), but not those in mice receiving 2000 or 50000 ppm, were

significantly increased (male: 25/50, 28/50, 35/50, 30/50; female: 7/50, 8/50, 19/51, 14/50). The number of male and female mice in the 10000 and 50000 ppm groups with multiple hepatocellular neoplasms compared with controls was statistically significantly increased. However, JECFA concluded that these effects were not dose-related and that curcumin is not a carcinogen in mice (FAO/WHO 1995). In addition, the EFSA ANS Panel (2010) noted that all statistically significant effects noted by the NTP refer to benign neoplastic lesions (adenomas). The EFSA Panel noted that the effects observed were not dose-dependent, were in line with historical control values and were not consistent across sexes and/or species. The Panel noted moreover that “hepatocellular tumours occurring in untreated and treated B6C3F1 mice are not relevant for humans”.

Two years study in rats

13. Groups of 60 male and 60 female F344/N rats were fed diets containing 0, 2000, 10000, or 50000 ppm turmeric oleoresin (79 to 85 % curcumin) for 104 (males) or 103 (females) weeks, which were estimated to deliver average daily doses of 0, 80, 460, or 2000 mg/kg bw per day to males and 0, 90, 440, or 2400 mg/kg bw per day to females. Nine or 10 rats from each exposure group were evaluated after 15 months. Survival of exposed male and female rats was similar to that of the controls. The final mean body weights of all exposed male rats and female rats receiving 2000 and 10000 ppm were similar to those of the controls. The final mean body weights of male and female rats that received 50000 ppm were slightly lower (up to 10 %) than those of the controls throughout much of the study. Feed consumption by exposed male and female rats was similar to that of controls throughout the study. The absolute and relative liver weights of female rats receiving 10000 or 50000 ppm were significantly greater than those of controls at the 15-month interim evaluation. There were no clinical findings related to toxicity. In male and female rats receiving 50000 ppm the haematocrit values, haemoglobin concentrations and erythrocyte counts at the 15-month interim evaluation were significantly lower than those in the controls. In addition, platelet counts in male and female rats that received 50000 ppm and reticulocyte counts in male rats that received 50000 ppm were significantly higher than those in the controls. No biologically significant differences were observed in clinical chemistry parameters. Gastrointestinal irritation (ulcers, hyperplasia and inflammation) was observed in high dose males and females. The incidence of clitoral gland adenomas (females) was statistically significantly increased in treated animals (NTP, 1993), JECFA concluded that these effects were not dose-related and that curcumin was not a carcinogen in rats. The authors did not

attempt to identify a NOAEL for this study (NTP 1993), but JECFA identified a NOAEL for the gastrointestinal effects of 440 mg/kg bw/day.

The EFSA Panel noted that all statistically significant tumourigenic effects observed in the NTP studies in rats and mice related to benign lesions (adenomas) and that the incidences for carcinomas did not reach statistical significance. The Panel also noted that the effects observed were not dose-dependent, were in line with historical control values and were not consistent across sex and/or species. The Panel also noted that the hepatocellular tumours observed in B6C3F1 mice are not relevant for humans. Therefore, the Panel agreed with the overall conclusion of JECFA that curcumin is not carcinogenic.

Human Studies

14. Twenty-five patients with conditions indicating a high risk of malignancy were administered diferuloylmethane (curcumin) (purity 99.3 %) in tablet form for 3 months. The starting dose was 500 mg/day, which was increased stepwise to 1000, 2000, 4000, 8000 and finally 12000 mg/day (equivalent to 7, 14, 29, 57, 114, and 171 mg/kg bw/day). The patients received regular follow-up, including physical examination, weekly haemogram, and “biweekly” blood electrolytes and biochemistry study (though blood results not presented). No adverse effects were reported at doses up to 8000 mg/day. The highest dose of 12000 mg/day was not acceptable to the patients because of the bulky volume of the tablets. The serum concentration of curcumin usually peaked at 1 to 2 hours after oral intake of diferuloylmethane and gradually declined within 12 hours. The average peak serum concentrations after taking 4000 mg, 6000 mg and 8000 mg of curcumin were 0.51 ± 0.11 µM, 0.63 ± 0.06 µM, and 1.77 ± 1.87 µM, respectively. Urinary excretion of diferuloylmethane was undetectable. The study authors noted that these results suggest that diferuloylmethane is not adequately absorbed from the gastrointestinal tract. (Cheng *et al*., 2001).

15. In a dose escalation study fifteen patients with advanced colorectal cancer received an extract of Curcuma (containing 20 mg of curcuminoids, 18 mg of diferuloylmethane and 2 mg of demethoxycurcumin suspended in 200 mg of essential oils derived from *Curcuma spp*.) daily for up to 4 months. Doses were equivalent to 36, 72, 108, 144 and 180 mg diferuloylmethane/day. There were three patients per dose. Neither diferuloylmethane, nor its metabolites (glucuronide or sulfate conjugates, hexahydrocurcumin or hexahydrocurcuminol) were detected in plasma or urine over up to 29 days of daily treatment. However, diferuloylmethane was detected in the faeces of all patients. Diferuloylmethane

sulfate was also detected in the faeces of one of the patients receiving diferuloylmethane at a dose of 180 mg/day, which may have been a result of biotransformation in the gut. The study authors therefore concluded that diferuloylmethane has low oral bioavailability in humans and may undergo intestinal metabolism. Oral Curcuma extract was well tolerated, and no dose-limiting toxicity was observed. However, gastrointestinal effects were reported, with one patient (receiving 108 mg diferuloylmethane/day) experiencing nausea and two patients (receiving 72 and 180 mg diferuloylmethane /day) experiencing diarrhoea. (Sharma *et al*., 2001).

16. In a subsequent phase I study, Sharma *et al.,* (2004) evaluated another formulation, which was a 500 mg curcuminoid capsule containing 450 mg diferuloylmethane, 40 mg demethoxycurcumin, and 10 mg bisdemethoxycurcumin. The dose levels of diferuloylmethane were 450, 900, 1800, or 3600 mg/day for up to 4 months (equivalent to 6, 13, 26, and 51 mg/kg bw/day). A total of 15 patients with refractory colorectal cancer were enrolled. Full blood cell count and urea, electrolytes, liver and bone function were measured in venous samples, and physical examination was performed, before treatment and on treatment days 1, 2, 8, 29, and monthly thereafter. The drug was well tolerated and dose-limiting toxicity was not observed. However, one patient consuming 450 mg diferuloylmethane daily and one patient consuming 3600 mg diferuloylmethane daily developed diarrhoea 1 month and 4 months into treatment, respectively. One patient consuming 900 mg diferuloylmethane daily experienced nausea, which resolved spontaneously despite continuation of treatment. In addition, a rise in serum alkaline phosphatase (ALP) level was observed in 4 patients, and serum lactate dehydrogenase (LDH) rose to > 150 % of pre-treatment values in 3 patients. Diferuloylmethane was detected in the plasma (at 1-hour post dosing) from three out of the six patients consuming 3600 mg of diferuloylmethane daily, but not from patients receiving the lower doses. The mean plasma concentration of diferuloylmethane in these three patients was 11.1 ± 0.6 nmol/L. Moreover, glucuronides and sulfates of diferuloylmethane and desmethoxycurcumin were found in the plasma from all 6 of the patients consuming 3600 mg of curcumin daily at all the time points studied. Analysis of urine suggested the presence of diferuloylmethane (0.1 - 1.3 µmol/L) and its sulfate (19 - 45 nmol/L) and glucuronide (210 - 510 nmol/L) conjugates in all of the samples from patients consuming 3600 mg of diferuloylmethane daily. Diferuloylmethane was not detected in plasma or urine from patients who received lower doses of diferuloylmethane. Abundant amounts of diferuloylmethane were recovered from the faeces at all of the dose levels. Diferuloylmethane levels in day 8 faecal samples from patients consuming 3600

mg of diferuloylmethane daily were between 25 and 116 nmol/g dried faeces.

17. A dose escalation study was performed with 24 healthy human volunteers using single doses of between 500-12000 mg of a turmeric extract (containing a minimum of 95 % of diferuloylmethane (75 %), bisdemethoxycurcumin (2 %), and demethoxycurcumin (23 %), equivalent to 7-171 mg/kg bw). Seven volunteers experienced minimal toxicity at doses > 14 mg/kg bw (diarrhoea, headache, rash and/or yellow stool) that did not appear to be dose-related. The remaining volunteers did not experience adverse effects (Lao *et al.,* 2006).

18. Nine healthy volunteers between 20 and 33 years of age were tested for haemoglobin, blood counts, liver and kidney functions, bleeding and clotting time and serum electrolytes initially and at 1 and 3 months of treatment. They were administered 0.6 ml of turmeric oil extract (TO; 59 % turmerone and Ar-turmerone) three times a day for 1 month and 1 ml in 3 divided doses for 2 months. One volunteer discontinued on day three because of allergic skin rashes which, on discontinuation of TO, gradually disappeared by two weeks. Another discontinued on day seven for intercurrent fever requiring antibiotic treatment. Seven volunteers completed the study. There was no effect of TO intake on bodyweight, blood pressure, symptoms and signs up to 12 weeks. There was no clinical, haematological, renal or hepatotoxicity of TO at 1 month and 3 months. Serum levels of AST, ALT, ALP, albumin, direct and indirect bilirubin were normal initially and remained within normal limits during treatment. One volunteer showed normal serum triglycerides and low-density lipoprotein (LDL) initially and at 4 weeks but elevated levels at 12 weeks. This volunteer was followed up for 1 month after discontinuation of TO and serum lipids returned to normal. (Joshi *et al.,* 2003).

19. Kanai *et al.,* (2011) evaluated the safety and feasibility of combination therapy using “curcumin” with gemcitabine-based chemotherapy. The ”curcumin” used was a mixture of diferuloylmethane (73 %), demethoxycurcumin (22 %), and bisdemethoxycurcumin (4 %), provided in microbead form. Gemcitabine-resistant patients with pancreatic cancer received 8 g oral curcumin daily (equivalent to 114 mg/kg bw) in combination with gemcitabine-based chemotherapy. The primary endpoint was safety for phase I and feasibility of oral curcumin for phase II study. Twenty-one patients were enrolled. No dose-limiting toxicities were observed in the phase I study and oral curcumin 8 g/day was selected as the recommended dose for 17 patients in the phase II study. No patients were withdrawn from this study because of the intolerability of curcumin, which met

the primary endpoint of the phase II study, and the median compliance rate of oral curcumin was 100 % (range 79-100 %). Plasma curcumin levels ranged from 29 to 412 ng/ml in five patients tested. Adverse effects were reported (neutropenia, fatigue, drowsiness, anorexia, obstruction of the gastrointestinal tract, and oedema) all of which were also attributed to disease progression or gemcitabine-based chemotherapy, and considered to be irrelevant to curcumin. Kanai *et al*. (2011) concluded that combination therapy using 8 g oral curcumin daily with gemcitabine-based chemotherapy was safe and feasible in patients with pancreatic cancer and warrants further investigation into its efficacy.

20. In September 2018, the Committee on Herbal Medicinal Products (HMPC) of the European Medicines Agency (EMA) considered several clinical studies with turmeric preparations involving patients with digestive disorders. Although a possible effect in improving symptoms was seen, firm conclusions could not be drawn due to limitations in the study design, for example only one study compared turmeric with a control group. However, the HMPC considered the effectiveness of these preparations for relief of mild digestive problems to be plausible and there is evidence that they have been used safely in this way for at least 30 years (including at least 15 years within the European Union). Thus, the HMPC concluded that turmeric preparations (which cover turmeric ground powders and extracts) can be used (only by adults) for relief of mild problems with digestion, such as feelings of fullness, slow digestion and flatulence. This conclusion was based on long-standing use and not on clear evidence of efficacy. The EMA considered that side effects of dry mouth, flatulence and stomach irritation may occur. If symptoms continue for longer than two weeks or worsen while taking the medicine, a qualified healthcare practitioner should be consulted.

Curcuminoids and hepatotoxicity: Case studies in humans

21. In addition to the reported cases in Italy, several cases of liver toxicity associated with intake of curcumin supplements have been reported in the literature.

22. In Australia, from Luber et al., (2019) a 52 year old woman presented to her general practitioner with a one-week history of nausea, pruritus, and painless jaundice with associated pale stools and dark urine. This occurred approximately one month following commencement of one tablet per day of a ‘High Potency’ turmeric supplement (375 mg curcuminoids and 4 mg black pepper per tablet), along with a flaxseed oil supplement and occasional diclofenac use for arthritic pain. (N.B. this supplement leads to a curcumin intake of 204.6 % of the upper

bound of the ADI of 3 mg/kg bw). She had no prior history of liver disease and had normal liver function tests (LFTs) three months before. Her medical history was notable only for oligoarticular osteoarthritis.

23. Upon admission, all oral medications and supplements were ceased. She was found to have a bilirubin concentration of 162 µmol/L with a hepatocellular injury profile on liver function tests (ALT 2591 U/L, AST 1770 U/L, ALP 263 U/L, and Gamma-Glutamyl Transferase (GGT) 370 U/L). With progressive jaundice over the subsequent days she was referred to the emergency department, at which point her bilirubin peaked at 536 □□mol/L.

24. Due to lack of significant improvement by day four of admission, a liver biopsy was performed. Histology showed nonspecific inflammatory changes with generally preserved hepatic architecture and no fibrosis. She was discharged on day 12 of admission (bilirubin 260 □□mol/L, ALT 1232 U/L) with the presumptive diagnosis of diclofenac induced liver injury. By two months after admission her liver function tests had normalised (bilirubin 21 □□mol/L, ALT 33 U/L) and she was discharged from the clinic. At this point she recommenced the turmeric supplement as sole therapy for her arthritis. Three weeks later her nausea recurred and repeat liver function tests showed an acute hepatitis (ALT 2093 U/L, AST 1030 U/L, and bilirubin 60 □□mol/L). She was advised to cease the turmeric supplement, and two months later her liver function tests had again normalised.

25. The turmeric supplements were sent for analysis by a validated liquid chromatography mass spectrometry method. Results were compared to a toxicology library containing approximately 1400 compounds, including medications, illicit drugs, and over-the-counter medicines. A further sample was analysed by inductively coupled plasma mass spectrometry for the presence of trace elements. The turmeric supplement tested negative for drugs, adulterants and toxic heavy metals. (Luber et al. 2019).

26. A 55-year-old man was found to have an asymptomatic transaminitis at a routine check-up. His background history included idiopathic thrombocytopenic purpura, hypertension, gout, and osteoarthritis, with regular medications including long-term telmisartan, atenolol, and lercanidipine. He had no known liver history, with normal liver function tests one year prior. His only new medication was commencement of a turmeric supplement five months prior to testing. He was referred to a hepatologist and underwent a screen for causes of acute hepatitis. Abdominal ultrasonography showed diffuse steatosis. A drug reaction was suspected, and the turmeric supplement was ceased. Follow-ups occurred over the subsequent four months. Near normalisation of liver function

tests occurred by one month (ALT 96 U/L, bilirubin 10 □□mol/L) with further improvement by four months after cessation (ALT 46 U/L, bilirubin 11 □□mol/L). The turmeric supplement was the presumed cause of the hepatitis. The turmeric supplement was not identified, thus further analysis on it could not be performed and hence dose information is unknown. (Luber et al. 2019).

27. In the US, reported by Suhail et al., (2020) a 61 year old female with polycystic liver disease presented with fatigue, dark urine and polyarthralgias for one week. She denied alcohol use. Physical examination demonstrated right upper quadrant abdominal tenderness. Laboratory findings were notable for elevations of aspartate aminotransferase (AST) 1553 mg/dL, alanine aminotransferase (ALT) 2607 mg/dL, alkaline phosphatase (ALP) 246 mg/dL and total bilirubin 1.6 mg/dL with a direct component of 1 mg/dL. Hepatic synthetic function was intact. Medications included naproxen and ergocalciferol (vitamin D) with no changes over the four years prior to the onset of transaminitis. Viral infections were ruled out. Autoimmune work-up yielded positive antinuclear antibody (1:250) with normal anti-smooth muscle antibody and serum IgG levels. Abdominal Doppler was negative for portal or hepatic vein thrombosis. Magnetic resonance cholangiopancreatography did not show any biliary duct pathology. It was discovered that the patient had been taking turmeric supplements for 6 months. A liver biopsy demonstrated pan lobular hepatitis with early parenchymal collapse suggestive of a morphologic counterpart of acute hepatitis and hepatocellular pattern of injury. The patient was thought to have drug-induced liver injury from turmeric pills that were discontinued and she was discharged with prednisone. Her LFTs normalized after 3 weeks, after which, her prednisone was tapered off.

28. Suhail et al. (2020) noted that the temporal association of liver injury in the patient, normalisation of LFTs upon withdrawal and improvement with steroids implicate the turmeric supplement as the likely causative agent of liver injury. The Roussel-Uclaf Causality Assessment Method (RUCAM) scale, which attempts to codify causality of drug toxicity into objective criteria, was eight in the patient indicating a probable adverse drug reaction. A negative serological workup and normalisation of LFTs following the discontinuation of steroids further solidify this conclusion.

29. In another case in the US reported by Lukefahr et al., (2018) a 71 year old woman was found to have an asymptomatic transaminitis at routine check-up. Her medical history included hypothyroidism, Raynaud’s syndrome, osteoarthritis, hypertension, dyslipidaemia, irritable bowel syndrome, and diverticulosis, with

regular medications including amlodipine, metoprolol, and atenolol. A ‘low cost’ turmeric supplement (of unknown product identification) was taken according to label recommendations for a period of 8 months prior to transaminitis testing. The patient was referred to a gastroenterologist. Laboratory and biopsy findings led to a diagnosis of autoimmune hepatitis. Treatment was limited to withdrawing use of turmeric supplements. AST and ALT decreased significantly within 30 days of discontinuation and normalised by 13 months. In this case, the patient (not the clinicians) hypothesised that the turmeric may have been the cause of the elevated liver transaminases and elected to cease its use.

30. Subsequent reassessment of the liver biopsy by Lukefahr et al., (2018) revealed auto-fluorescent inclusions in the pigment-laden histiocytes, with an excitation/emission spectrum consistent with curcumin, or possibly lipofuscin. Histiocyte fluorescence, which was not noted in liver biopsy specimens from patients with unrelated disorders, was quenchable by treatment with Sudan Black B (SBB), as has been reported for lipofuscin. However, because the authors also documented complete SBB quenching of curcumin autofluorescence in fixed cultured cells specifically loaded with curcuminoids, the authors could not ascertain with certainty whether the histiocyte inclusions were composed of lipofuscin, a lysosomal degradation product and/or curcuminoids derived from the turmeric supplement that the patient was still consuming at the time of the biopsy.

31. Smati *et al*., (2023) report a case report of turmeric-induced liver injury in a 36-year-old woman with minimal alcohol use and no use of prescription or over-the-counter medications, acetaminophen, tobacco, or recreational drugs who had been taking approximately 2g turmeric per day. The patient showed increased liver enzymes and bilirubin. Liver biopsy was notable for chronic portal triaditis, bile ductular injury, and patchy hepatocellular necrosis consistent with hepatocellular liver injury. Her liver functions showed gradual but steady improvement. Post-discharge, liver tests (aspartate aminotransferase (AST)/alanine aminotransferase (ALT) > alkaline phosphatase (ALP)) slowly decreased to relatively normal values 31 days after stopping turmeric intake. By the point that she presented as an outpatient for her 6-month post-discharge follow-up, her liver function test results were all normal. The discussion of the article suggests that “Despite significant underreporting of HDS [herbal and dietary supplements]-associated adverse events, approximately 20% of documented drug-induced liver injuries are secondary to HDS.” Through reference to the wider literature, the authors go on to argue that there is a “a dose-dependent relationship between curcumin absorption and liver injury” and

that “further complicating safe dosage of [curcumin] are factors such as form of delivery and inclusion of bioavailability-increasing substances like piperine.”

32. A case study of potential drug-induced liver injury related to turmeric consumption was reported by Ajitkumar *et al*., 2023. The patient was a 55-year-old woman with progressive jaundice and elevated bilirubin and liver enzymes but without evidence of acute liver failure. The patient had been taking 2g turmeric once daily for wrist pain for approximately one month prior to presentation. Her liver function tests normalized 2 months after her initial presentation and cessation of turmeric supplement intake.

List of Abbreviations and Technical terms

ADI	Acceptable Daily Intake
ALP	Alkaline Phosphatase
ALT	Alanine Aminotransferase
ANS Panel	Scientific Panel on Food Additives and Nutrient Sources Added to Food
AST	Aspartate Aminotransferase
cDNA	complementary Deoxyribonucleic acid
CYP	P450
dL	Decilitre (100 millilitres)
EFSA	European Food Safety Authority
EMA	European Medicines Agency

ETE	Ethanolic Turmeric Extract
FAO	Food and Agricultural Organisations of the United Nations
g	gram
GGT	Gamma-Glutamyl Transferase
HDS	Herbal and Dietary Supplements
HMPC	Committee on Herbal Medicinal Products
IgG	Immunoglobulin G
JECFA	Joint FAO-WHO Expert Committee on Food Additives
kg bw/day	Kilogram Bodyweight per Day
L	Litre
LDL	Low-Density Lipoprotein
LFTs	Liver Function Tests
mg	Milligram
mg/kg bw/day	Milligrams per Kilogram Bodyweight per Day
ml	Millilitre

ng	Nanogram
nMol	Nanomole
NOAEL	No-Observed-Adverse-Effect Level
NTP	National Toxicology Program
OECD	Organisation for Economic Co-operation and Development
ppm	Parts per million
RNA	Ribonucleic acid
RUCAM	Roussel-Uclaf Causality Assessment Method
SBB	Sudan Black B
TEO	Turmeric Essential Oil
TO	Turmeric Oil extract
U	Enzyme unit
µg	Microgram
µM / µMol	Micromole
WHO	World Health Organisation

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