TOX/2017/02

COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT

First draft statement on potential risks from nickel in the diet of infants aged 0 to 12 months and children aged 1 to 5 years

Introduction

1. The Committee on Toxicity (COT) has been asked to consider the toxicity of chemicals in the diets of infants (0 to 12 months) and young children (1 to 5 years), in support of a review by the Scientific Advisory Committee on Nutrition (SACN) of Government recommendations on complementary and young child feeding. A scoping paper (TOX/2015/32), highlighting some of the chemicals for possible consideration was discussed by the COT in October 2015. Members concluded that a full review of the exposures from nickel should be completed.

2. A discussion paper on nickel (TOX/2016/41) was presented to the COT in December 2016. At the December meeting the Committee requested that information be provided to establish a TDI. The TDI established by the European Food Safety Authority (EFSA) was based on an embryo-fetal toxicology endpoint and as such may not be sufficiently protective of the infant and toddler population groups. Relevant toxicological information and a provisional TDI are presented in Annex A.

3. Members also requested that data be provided on nickel sensitisation in the infant and young child age groups. This should include any estimates of prevalence within the general population in order to establish whether acute dietary exposures should be considered in addition to chronic dietary exposures. This information is provided in Annex B.

4. Members requested that a range of breast milk concentrations be considered to reflect the low and high concentrations of nickel found in breast milk. A first draft statement is provided in Annex C.

Questions on which the views of the Committee are sought

5. Members are invited to consider the information provided in Annexes A and B and the first draft statement in Annex C and to answer the following questions:

Annex A

i. Has enough information been provided on the bioavailability of nickel?

ii. Do Members agree with the proposed TDI?

iii. If not, what information do Members want to establish a TDI?

Annex B

iv. Has enough information been provided, in relation to the prevalence of sensitised individuals in the infant and young children populations, to determine whether acute exposure needs to be addressed in this statement?

v. Do Members agree with the reference point ($BMDL_{10}$ of 1.1 µg/kg bw/day) established by EFSA to be suitable for assessing acute exposures in these age groups?

Annex C

vi. Members are asked to comment on the layout of the first draft statement.

vii. Do Members agree with the concentrations of nickel used to assess exposures from breast milk?

viii. Taking into account the prevalence of sensitisation in infants and young children aged 1 to 5, does a more in-depth acute assessment need to be carried out?

ix. With dietary exposures to nickel more than 10-fold greater than nickel exposure from environmental sources do Members consider that an assessment of aggregate exposures be carried out?

x. Do Members wish the risk characterisation to consider acute and chronic exposures?

xi. Do Members have any further comments?

Secretariat
January 2017

COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT

First draft statement on potential risks from nickel in the diet of infants aged 0 to 12 months and children aged 1 to 5 years

de novo tolerable daily intake (TDI) for chronic exposure to nickel

Introduction

1. At the December 2016 meeting the Committee concluded that the tolerable daily intake (TDI) that had been established by EFSA was not applicable to these age groups because it was based on embryo/fetal toxicity, an effect that is possible only when exposure occurs prior to birth. In order to establish a TDI specific to these age groups, Members requested further information on the results of an existing 3-generation toxicity study and other available multigeneration toxicity studies, particularly on any adverse effects reported in offspring. If there were no adverse effects in offspring observed in these studies then the TDI could be based on systemic toxicity in older animals.

2. This paper sets out the information available for establishing a TDI using multi generation reproductive studies. The process undergone by EFSA to establish a TDI for chronic dietary exposure to nickel is presented first followed by details from the 2 studies used by EFSA to calculate the TDI. Additional 2- or multigeneration and systemic studies for possible consideration in determining a TDI are also presented. A summary of these data has also been provided in Table 1.

3. Data from these studies have been used to propose a TDI.

EFSA

4. EFSA's CONTAM panel (2015) established the TDI for chronic exposure to nickel based on a dose response assessment. Reproductive and developmental toxicity was identified as the critical effect for the risk characterisation of chronic oral exposure to nickel. A one-generation dose-range finding study (DRF) and a subsequent main 2-generation study (2-GEN) (both described in more detail below) were identified as the most suitable and reliable dose-response information for reproductive and developmental effects. Five dose groups (2.2, 4.4, 6.6, 11 and 17 mg nickel/kg bw) and a control with 7 – 8 animals/group were used in the DRF study and 4 dose groups (0.2, 0.6, 1.1 and 2.2 mg nickel/kg bw) and a control with 25 – 28 animals/group were used in the 2-GEN study. A benchmark dose

(BMD) approach was used in the dose-response assessment and the incidence of litters with post implantation loss per treatment group was used as the endpoint.

5. The DRF and 2-GEN studies were carried out under identical conditions, and therefore the CONTAM panel were able to combine the data from the 2 studies, on the incidence of litters with post-implantation loss per treatment group, to derive a reference point. This was statistically more suitable than the data from the individual studies. The quality of the data from the combined DRF and 2-GEN studies met the criteria developed by EFSA (EFSA, 2009, 2011e) for the BMD approach using the Nlogistic, the so-called NCTR and the Rai-van-Ryzin (RvR) models of the US EPA BMDS software.

6. The $BMDL_{10}$ value of 0.28 mg nickel/kg bw/day was selected by the CONTAM panel as a reference point for chronic exposure to nickel. BMD analysis was also carried out on studies by Smith *et al.* (1993) and Panday and Srivastava (2000) and the resultant $BMDL_{10}$ values confirmed the $BMDL_{10}$ value of 0.28 mg nickel/kg bw derived from the DRF and 2-GEN studies, which were of a better quality. (EFSA, 2015).

7. From the selected $BMDL_{10}$ of 0.28 mg/kg bw/day the CONTAM Panel applied a default uncertainty factor of 100, to account for extrapolation from experimental animals to humans and for inter-individual variability, and derived a TDI of 2.8 µg/kg bw.

2- or multi-generation studies

SLI, 2000a (DRF)

8. This range-finding study was conducted to evaluate the potential effects of nickel sulphate hexahydrate when administered orally to rats over the course of one generation. Data from this study were used to select dosage levels for a 2-generation reproduction study in rats.

9. Nickel sulphate hexahydrate, in reverse osmosis-deionized water, was administered once daily by gavage to 6 groups of Sprague-Dawley rats. The exposure concentrations were 0, 10, 20, 30, 50 and 75 mg/kg/day (equivalent to 0, 2.2, 4.4, 6.7, 11 and 17 mg elemental nickel/kg bw). Each dose group contained 8 animals of each sex. F0 parental animals and the F1 offspring were dosed for 14 days prior to mating and on postpartum day 22, respectively. Dosing was continued until the day prior to or the day of scheduled euthanasia. After a minimum of 14 days of treatment each female was cohabited with a single randomly selected male from the same treatment group (1:1 pairings).

10. Unless relevant to findings in the F1 generation, only results from the F1 generation and details on post implantation loss as this is the endpoint used by EFSA for their BMD analysis are provided in this paper.

11. Mean live litter size was significantly decreased and the incidence of dead pups was significantly increased on day 0 at the 17 mg/kg bw level. Significant increases in the incidence of dead pups on lactation day 0 were also observed at the 2.2, 4.4 and 6.7 mg/kg bw levels, but not at 11 mg/kg bw. Pup viability at the 17 mg/kg bw level continued to decline and was significantly lower than controls on lactation day 4, prior to culling. After culling on day 4, pup viability appeared to stabilise at 17 mg/kg bw.

12. Pup observations were generally unremarkable and body weight differences were not toxicologically meaningful during lactation. All animals survived to scheduled euthanasia and no remarkable signs of toxicity were noted during the dosing phase or during gross necropsy.

SLI, 2000b (2-GEN)

13. This study was undertaken to determine the potential effects of nickel sulphate hexahydrate when it is administered orally to rats over 2 generations.

14. Nickel sulphate hexahydrate, in reverse osmosis-deionized water, was administered once daily by gavage to 5 groups of Sprague-Dawley rats. The exposure concentrations were 0, 1, 2.5, 5 and 10 mg/kg/day (equivalent to 0, 0.22, 0.56, 1.1 and 2.2 mg elemental nickel/kg bw). Each dose group contained 28 animals of each sex. F0 parental animals and the F1 offspring were dosed for 70 days prior to mating and on postpartum day 22, respectively. Dosing was continued until the day prior to scheduled euthanasia. After a minimum of 70 days of treatment each female was cohabited with a single randomly selected male from the same treatment group (1:1 pairings). During the breeding phase of the F1 generation, only non-siblings were mated.

15. Unless relevant to findings in the F1 generation, only results from the F1 generation and details on post implantation loss as this is the endpoint used by EFSA for their BMD analysis are provided in this paper. No toxicologically meaningful differences were noted in F1 pup viability data, pup survival, pup bodyweights during lactation or in body weight, body weight gain, food consumption, during the growth phase of the F1 generation. No treatment-related differences were noted in the onset of vaginal opening or the completion of preputial separation.

16. There were no statistically significant differences in F1 copulation and fertitlity indices, estrous cycling, precoital intervals, or gestation lengths. Whilst neither the number of F1 females with evidence of estrous cycling nor mean estrous cycle lengths were significantly different between the F1 groups, the number of cycling F1 females was lower and the mean cycle lengths were longer in each group of the F1 females compared to the F0 females. The authors put this down to the likely inadvertent induction of pseudopregnancy.

17. No test article-related mortality or clinical signs of toxicity were noted during the growth phase of the F1 animals selected to produce the F2 generation. There was no evidence of treatment-related changes upon gross necropsy of the F1 parental animals. Nor were there any test article-related microscopic changes.

18. There were no toxicologically meaningful differences in mean implantation scar counts, mean number of live pups on lactation day 0, mean post-implantation loss in F1 females or in sperm parameters in F1 males.

19. There were statistically significant differences in some of the F1 organ weight data. These consisted of lower absolute pituitary weight in group 2 males; higher relative adrenal weight and lower relative liver weight in group 4 and 5 males; and lower relative liver weight in group 3 and 5 females. Of these only the decreased relative liver weights in the group 4 and 5 males were considered toxicologically significant. Although relative not absolute liver weights were significantly decreased in F1 males of groups 4 and 5, both the absolute and relative liver weights were significantly decreased in males in group 5 of the F0 generation. This suggests that these findings may be test article related.

20. Nickel sulphate hexahydrate was administered to rats by oral gavage over 2 generations. At elemental nickel concentrations of up to 2.2 mg/kg bw/day there was no toxicity or adverse reproductive effects. There was a slight reduction in adult male liver weight in F0 males dosed with 2.2 mg/kg bw/day and in the F1 males dosed with 1.1 and 2.2 mg/kg bw/day. There were no treatment-related histopathological finding in the liver or other tissues from rats dosed with 2.2 mg.kg bw/day. Therefore the dose level of 2.2 mg/kg bw/day was considered the no observed adverse effect level (NOAEL) for oral administration of nickel sulphate hexahydrate over 2 generations.

Rush, (2002); SLI, (2002)

21. In a 90-day range finding study, nickel sulphate hexahydrate was administered daily by oral gavage to F344 rats at levels of 0, 50, 75, 100, 125 and 150 mg nickel sulphate hexahydrate/kg bw/day (corresponding to 0, 11, 17, 22, 28 and 33 mg nickel/kg bw/day). Bodyweight gain was reduced in an exposure-related manner in all treated groups. Males exhibited a significant reduction in body weight gain within the first 4 weeks of treatment at the 2 highest doses. Exposures of males in these 2 groups were subsequently reduced to 30 and 15 mg nickel sulphate hexahydrate/kg bw/day (corresponding to 7 and 3 mg nickel /kg bw/day), respectively to ensure survival of the animals for the duration of the study. Following the reduction in exposure levels, bodyweight gains were nearly comparable to the control group. Decreases in bodyweight were observed at doses ≥ 50 mg nickel sulphate hexahydrate/kg bw/day. Histopathological analysis showed no treatment-related effects. The NOAEL was 30 mg nickel sulphate hexahydrate/kg bw/day, corresponding to 7 mg nickel/kg bw/day. (Rush, 2002; SLI, 2002).

Ambrose et al. (1976)

22. A 3-generation study was undertaken in albino Wistar rats. Rats that were 28 days old were separated into 4 groups of 30 rats of each sex, to constitute the parent F/0 generation. Mean bodyweight and weight range, in so far as possible were similar for all groups. One group was placed on each of the following dietary concentrations of nickel sulphate hexahydrate: 0, 250, 500 and 1000 ppm (0, 56, 111 and 222 mg elemental nickel/kg bw).

23. Finely ground laboratory chow served as the basic diet and nickel solutions were thoroughly blended into the diet. Rats were individually caged and had free access to water and diet.

24. After 11 weeks on the above dietary regimen, 20 females from each diet were transferred to individual breeding cages and each was mated with a male of the same dietary level of nickel for the F/1a generation. Male rats within each group were rotated to a different female on each of three successive 7-day periods. On the 20th mating day all males were removed. Records were maintained of mating, number of pregnancies, litters cast (alive and dead), pups in litter at 1, 5 and 21 days weaning, and total weight of the litter at weaning. Litters containing more than 10 offsprings were randomly reduced to 10 on day 5. All surviving F/1a siblings were sacrificed and autopsied at weaning. Approximately 10 days after weaning of F/1a litters, F/0 parent generation rats were remated for F1/b litters. Procedures and observations recorded were the same as those described for F/1a litters. Following weaning of F/1b litters, surviving F/0 rats were sacrificed and autopsied.

25. For the F/2 generation 30 male and 30 female F/1b offsprings from each diet level were continued on their respective parents' diet for 11 weeks at which time 20 of each sex within each group were mated and the same procedure followed as with the F/0 generation through production and weaning of F/2a and F/2b litters. At weaning of F/2b rats, F/1b parents were sacrificed and autopsied. For the F/3 generation, the same procedure as with the previous generations was followed through the production of F/3a and F/3b litters. All matings in each generation were made with rats from different litters.

26. Bodyweights for parent generation rats (F0) on 56 and 111 mg/kg bw diets, before mating and at weaning of respective litters, were not adversely affected, but rats on 222 mg/kg bw exhibited slightly lower bodyweights. The average decrease in bodyweight did not exceed 8 % for females and 13 % for males.

27. On fertility, gestation, viability and lactation indices, no adverse effects were noted at any of the dietary levels of nickel. Data on the number of pups born dead showed higher incidence of stillborn in the first generation at all levels of nickel, but this was not observed to any extent in subsequent

generations. The number of siblings (alive and dead) cast per litter averaged 10.3, 10.6, 9.8, and 9.0 for 0, 56, 111 and 222 mg/kg bw diets respectively. The number of siblings weaned per litter were progressively fewer with increasing dietary level of nickel, averaging 8.1, 7.2, 6.8 and 6.4 for 0, 56, 111 and 222 mg/kg bw diets, respectively. On average weaning bodyweight, a clear-cut adverse effect is only apparent in weanlings of females on 222 mg/kg bw diet, averaging 73 % of control. However, offsprings maintained on 222 mg/kg bw diet from weaning to mating of succeeding generations recovered considerably from this deficit, averaging 92 % of controls.

28. Gross observations on siblings cast, at all dietary levels of nickel through 3 generations showed no teratogenic effects. Histopathologic findings on F/3b weanlings, 10 of each sex on each dietary level, were entirely negative.

RTI, (1988)

29. A 2-generation reproduction and fertility study was used to evaluate the potential reproductive toxicity of nickel chloride. Only report III of III, which documented the treatment of the F1 generation and the F2 generation, was available to include in this paper.

30. Male and female CD rats in the F1 generation remained assigned to the same dose group as their parents i.e., 0, 50, 250 or 500 mg nickel (Ni^{++})/L filtered/deionised water. Exposure was continuous through 21 to 24 weeks of age for F1 males (until the end of the second cohabitation period), or 27 to 30 weeks of age for F1 females (until sacrifice at gestational day 20 of eh F2b litter). Treatment of the F1b pups started on postnatal day 21.

31. Males and females were 27 - 32 days old at the time of arrival at RTI for the P0 generation. At weaning (postnatal day 21 of the F1b litter) 218 F1 males and 204 F1 females were assigned to the study. During the exposure period animals were given ad libitum access to control or nickel-treated water.

32. On postnatal day 42, a total of 112 male and 109 female breeder animals (F1 generation) were randomly assigned to 3 cohorts. Breeding pairs were then sequentially assigned to the 3 cohorts (10 breeding pairs per cohort per dose group). For the 500 ppm group (22 males and 19 females) animals were assigned to breeding pairs in 2 cohorts (10 and 9 pairs, respectively). Three untreated females were assigned as mates to the three remaining males, and these pairs were assigned to the 2 cohorts so that each cohort had 11 breeding pairs.

33. Average pup bodyweight for F1b litters was significantly below controls on postnatal day 21 at 250 ppm (87 % of control weight, $p < 0.05$) and at 500 ppm (75 % of control weights: $P < 0.01$). At week 32 (of the whole study) body weight for the 500 ppm animals was still significantly decreased (81 % of control weight for females and 72 % for males: both $p < 0.01$), but minor

reductions in body weight at 250 ppm (95 % of control weights for both sexes) were no longer statistically significant.

34. During the period of juvenile development (days 22 - 24) for selected F1 pups, 23 males and 22 females died or were sacrificed *in extremis* (11 % of the total population of 218 males and 204 females). The incidence of deaths was significantly increased at 250 ppm ($p < 0.05$) and 500 ppm ($p < 0.01$) for males (2/60, 4/67, 8/60 and 9/31 animals, in the control through high-dose groups) and at 500 ppm ($p < 0.01$) for females (4/58, 5/59, 4/59 and 9/28).

35. As adults (after 42 days of age) all F1 males survived to scheduled sacrifice: among females, 4 deaths occurred at parturition of the F2a litter (3/30 females at 250 ppm and 1/19 at 500 ppm). Although the incidence of maternal deaths was not strictly dose-related, deaths had been observed under similar circumstances for nickel-exposed females in the P0 generation, but not in the P0 or F1 control groups. In addition death during delivery is an unexpected finding based upon historical data. Collectively, the incidence of these deaths, suggest that nickel exposure was associated with compromised status of pregnant females around the time of parturition. Clinical signs observed for both males and females included rough hair coat, piloerection and dental problems and were not very different from those observed for the P0 generation.

36. During the non-reproductive portions of this study, average daily intake of nickel tended to decrease across weeks due to decreased relative fluid intake in all groups. This effect was also noted for male rats and appeared to be age-related. Average nickel intake for F1 females in the low- through high-concentration groups, respectively was 8.7, 35, and 67 mg/kg/day during week 33; 5.8, 23 and 44 mg/kg/day during week 38 and 4.6, 18 and 35 mg/kg /day during week 49.

37. Water intake (g/kg/day) for females during the non-reproductive portion of the study was not affected at 50 ppm (99 – 107 % of control intake), but was significantly reduced ($p < 0.01$) at 250 ppm (76-84 % of control) and 500 ppm (74 – 87 %) of control during each week of the exposure period).

38. Average body weight for F1 females in the 50 and 250 ppm groups were not significantly different from the control group. The 500 ppm group exhibited average body weights which were significantly below the control group (81 – 93 % of control weights; $p < 0.05$ or 0.01) for each week during the non-reproductive portions of the study.

39. Average daily water intake and therefore of nickel intake (for exposed groups) for each group of F1 males tended to decrease across weeks. Average nickel intake for the low- though high- concentrations, respectively was 7.6, 36 and 63 mg/kg/day during week 33 and 3.0, 14 and 28 mg/kg/day for week 49. Water intake showed a significant decreasing trend across groups ($p < 0.05$, 0.01 or 0.001) within each week of the study, except for

weeks 37, 45 – 47 and 49. The predominant effects on water intake occurred at 250 and 500 ppm prior to study week 45.

40. Food intake for nickel-exposed males did not differ significantly from the control animals for weeks 33 – 37. For the remaining weeks (38 and 41 – 49), no effects were observed on food intake at 50 or 250 ppm. At 500 ppm, relative food intake was significantly increased (108 – 121 % of control intake ($p < 0.01$). The absolute amount of food consumed was significantly decreased ($p < 0.05$ or 0.01) in the 500 ppm group throughout the study due to the persistence of decreased body weight for males in that group.

41. Throughout weeks 32 – 49 only males treated with 500 ppm had average body weights significantly different from the controls (71 – 86 %; $p < 0.01$).

42. On gestational days 0, 6 and 13 gestation body weight for timed-mated females with litters showed significant trends ($p <0.05$) due to non-significant decreases at 500 ppm. On gestational day 20 the 500 ppm group was significantly below controls (87 % of control weight; $p < 0.01$).

43. Relative food consumption for F1 dams during gestation showed no adverse treatment-related effects on gestational days 0 – 6 and 6 – 13. However, food consumption was significantly decreased (93 % of control; $p < 0.01$) at 500 ppm on gestational days 13 – 20.

44. Water consumption during gestation was not affected by nickel exposure at 50 ppm. However, water consumption in the 250 and 500 ppm groups was significantly below controls ($p < 0.01$) during gestational days 0 – 6, 6 – 13 and 13 – 20 (71 – 77 % of control intake at 250 ppm and 58 – 73 % of control at 500 ppm). Within individual groups, average daily fluid intake did not vary greatly across different periods of gestation. Thus average daily nickel consumption was relatively stable throughout gestation e.g., 5.8 – 6.4, 21 – 25 and 39 – 45 mg/kg/day at 50, 250 and 500 ppm, respectively.

45. During late gestation and lactation the female body weight of the 500 ppm group was significantly below controls (82 – 92 %: $p < 0.05$ or 0.01). Maternal food consumption was significantly decreased only at 500 ppm ($p < 0.01$) to 45 % of the control during gestation day 20 to postnatal day 1.

46. Maternal water intake was not affected at 50 ppm throughout late gestation and lactation. At 250 ppm, water intake was significantly decreased to 66 % of control ($p < 0.01$) on gestational day 20 to postnatal day 1. The 500 ppm group showed significant reduction in water consumption ($p < 0.05$ or 0.01) for all periods of measurement from gestational day 20 to postnatal day 21. The characteristic slight reduction in fluid intake during late gestation and the large systematic increase during lactation was observed, in addition to the treatment-related decreases. Nickel average consumption during this time was: the 50 ppm group increased from 5 to 13 mg/kg/day; the 250 ppm group

increased from 16 to 55 mg/kg/day; and the 500 ppm group increased from 15 to 90 mg/kg/day.

47. The F2a litters were evaluated on postnatal days 1 to 21. On postnatal days (pnd) 1, 4, 14 and 21 significant decreasing trends ($p < 0.01$ or 0.001) for live litter size were observed. The low dose was a NOAEL (104 – 110 % of control litter size), the mid-dose litter size was not significantly reduced (91 – 97 % of control), and the high-dose group was significantly below the controls (67 – 84 % of control). Average pup bodyweight per litter was not affected at 50 or 250 ppm. At 500 ppm, average pup bodyweight per litter was reduced to 91, 90, 87, 84 and 86 % of the average control weights on pnd 1, 4, 7, 14 and 21, respectively. At scheduled necropsy, no treatment-related findings were observed.

48. During the gestation period of the F2b litters maternal body weight, food consumption, water consumption, and consequently, exposure to nickel was comparable to that of the F2a litters.

49. At scheduled sacrifice (gd 20 of the F2b litters), time-mated F1 females exhibited reduced body weight at 500 ppm. Absolute liver weight exhibited a significant decreasing trend ($p < 0.001$), which was 103, 94 and 80 (significant: $p < 0.01$) % of control at 50, 250 and 500 ppm, respectively. Relative liver weight (% body weight) also exhibited a decreasing trend across all groups ($p < 0.001$), with reductions of 98, 96 and 88 % ($p < 0.01$) of control weights at 50, 250 and 500 ppm, respectively. Relative kidney weight (% body weight) exhibited a significant increasing trend across all groups ($p < 0.001$), showing 101, 105 and 113 ($p < 0.01$) of control values for at 50, 250 and 500 ppm, respectively. There was also suggestive evidence of increased relative adrenal and relative lung weights also at 500 ppm. Treatment related microscopic findings were limited to an apparent increase in histiocytic cellular infiltration of the lungs at the high dose (7, 7, 7 and 44 % of females examined from the control through high dose groups, respectively).

50. As scheduled, F1 breeder males were sacrificed following the second cohabitation period. Bodyweights were 106, 98 and 86 ($p < 0.01$) % of controls for 50, 250 and 500 ppm, respectively. No differences were observed for adrenal weights. For all other weighed organs, statistically significant tests were obtained for either the absolute or relative weights. For prostate and heart weights the group averages generally followed the pattern of bodyweight, and no differences among groups were observed for relative organ weights. Liver weight generally followed the pattern for body weight: relative weights at the low- and mid-dose levels were 106 % of the controls ($p < 0.05$), but the absence of a clear dose-related pattern suggests a spurious result.

51. Kidney weight was significantly higher at 50 ppm (112 % of control: $p < 0.01$), but did not differ from controls at 250 or 500 ppm; relative kidney weight was significantly higher in all groups ($p < 0.05$ or 0.01), but a clear association with dose was not observed (106, 111 and 108 % of control values in the low

through high dose groups, respectively). Relative lung weight was increased (114 % of control; $p < 0.05$) at 500 ppm. Pituitary weight did not differ among groups; relative pituitary weight showed an increasing trend ($p < 0.001$) for which 50 ppm was a NOAEL (95 % of control), 250 ppm was not significantly increased (109 % of control) and 500 ppm was above the control group (123 % of the control group; $p < 0.01$). Thus, differences in organ weights among groups failed to show a clear association with nickel exposure, with the exception of increased relative lung and pituitary weights at 500 ppm. By comparison P0 generation males showed increased pituitary weight (both absolute and relative) at 250 and 500 ppm, but other relative organ weights were not affected. No treatment-related pathology was noted upon gross examination at necropsy. Treatment-related microscopic findings were limited to an apparent increase in histiocytic cellular infiltration of the lungs at the high dose (0, 3, 3 and 18 % of males examined from the control through high dose groups, respectively).

Smith *et al*. (1993)

52. Four groups of 34 female Long-Evans rats (40 - 43 days) each were given nickel chloride ($NiCl_2 \cdot 6H_20$) drinking water solutions at 0, 10, 50 and 250 ppm for 11 weeks prior to breeding and then throughout the study[1]. Males had prior breeding experience and were not dosed with nickel chloride. After the first breeding, females were bred a second time, only if they were sperm positive in the first breeding. Dams were rested for 2 weeks after weaning of the first litters before the second breeding was initiated.

53. There were no overt clinical signs of toxicity in any of the groups. Per kilogram bodyweight, the food and water consumption were significantly reduced in lactation period 2 compared to lactation period 1. The overall average daily nickel dose was 1.3, 6.8 and 31.6 mg/kg bw/day for the groups dosed with 10, 50 and 250 ppm, respectively.

54. Pup sex ratios were comparable across dose groups in gestation period 1 and gestation period 2. The only significant reduction in pup growth was seen in L1 male pups in the 50 ppm dose group. Pups in lactation period 2 (including controls) weighed significantly less than those in lactation period 1. This difference persisted until weaning. No significant differences in litter size were found between L1 and L2.

55. There was a dose-related increase in both the number and proportion per litter of pups either born dead or dying shortly afterwards (trend analysis: G1, $P < 0.001$, 0.04; G2, $P < 0.03$, 0.02). In G1 pup death occurred at statistically significant levels only in the litters from females drinking 250 ppm nickel. In the second gestation period the number of dead pups per litter was

[1] Except when males joined the females for copulation. On these occasions overnight the females were given water without nickel chloride so that the untreated males would not be inadvertently treated.

significantly increased at each dose. The proportion of dead pups per litter was statistically significant at 10 and 250 ppm nickel, while the increase observed at 50 ppm approached statistical significance (P, 0.075).

56. Additional pups died during both lactation periods in all dose groups, mostly before PD7. During L1, one control female lost an entire litter (13 pups) and 11 of 14 pups were lost in a second control litter; in the high-dose group 31 of the 65 deaths were from three litters (10/15, 12/15, 9/14). During L2, the deaths were dose related (P = 0.03), and there were no large losses from single litters in controls or in females drinking 10 ppm. At 50 ppm, 24 of the pups lost were from 2 litters (11/14, 13/13), and at 250 ppm, 3 entire litters were lost (34 pups). The survival rate during L2 in the 50 and 250 ppm groups was significantly decreased among male pups at all time points measured, and this effect became more pronounced toward weaning (Day 21) (50 ppm, P = 0.023; 250 ppm, P = 0.016).

Systemic toxicity studies

57. In the study by Gathwan, Al-Karkhi and Al-Mulla (2013), 50 day old male Balb/c mice (5 animals per dose group) were exposed to nickel chloride by gavage for 40 days at 0, 0.5, 2 or 4 mg nickel/kg bw/day. There was a dose-dependent decrease in intake of feed and water. Significant decreases in body and liver weight were found in animals treated with 2 or 4 mg nickel/kg bw/day. In addition, hepatocyte degeneration, nuclear pycnosis, cellular swelling and congestion of blood vessels, cellular hypertrophy, increases in apoptosis and severity of necrosis were observed. The LOAEL for hepatotoxicity was 2 mg nickel/kg bw/day and the NOAEL was 0.5 mg nickel/kg bw/day. (Gathwan, Al-Karkhi and Jaffar Al-Mulla, 2013).

58. EFSA (2015) summarised data from a report by the American Biogenics Corporation, (1988). Sprague Dawley (SD) rats were exposed to nickel chloride hexahydrate by oral gavage at doses of 0, 5, 35 and 100 mg nickel/kg bw/day for 91 days. Clinical signs of toxicity were observed at the highest dose. A dose-related increase in mortality was observed (0, 2, 14 and 60/60 animals, respectively). Mortality at high dose and in 3/6 males and 3/8 females at the mid dose was attributed to treatment. Lower bodyweight and food consumption were noted at the 2 highest doses. At the interim sacrifice significant increases in white blood cells were seen at low and mid doses (not measured at the high dose due to the decreased survival in that treatment group) as well as dose-related increases in platelet count in females, increases in differential count in neutrophils and decreases in lymphocytes at medium dose in females. There was also a dose-related decrease in glucose at the mid-dose. Decreases in kidney, liver, spleen, brain and heart weights were observed in males at mid dose and decreases in kidney weight in females at the mid dose. Gastrointestinal tract (discoloured contents, distension, stomach discolouration, ulceration and smooth mucosa) and lung abnormalities (pneumonitis in 6/19 males and 9/17 females in medium dose) were observed in treated animals. Macroscopic ulcerative gastritis and

enteritis was observed at high dose. No NOAEL was identified in this study. The LOAEL was 5 mg Ni/kg bw/day. (EFSA, 2015).

59. Adult male SD rats were given 0, 4, 10 and 20 mg nickel/kg bw/day nickel sulphate hexahydrate in their drinking water for 13 weeks. Slight decreases in bodyweight were noted at the high dose. Changes in several organ weights were also noted. Decreases in both relative and absolute liver weights were observed at the 2 highest doses. Decreases in absolute weight of the testes and heart were observed in treated animals and increases in absolute kidney, brain and spleen weights at high dose. There were also increases in relative spleen weigh tin all treated groups, in relative kidney weights at low and high dose, relative brain weight at high dose, absolute lung weights at low and high dose and relative lung weights at low dose. Total plasma proteins were decreased at the 2 highest doses and plasma albumin and globulins as well as plasma glutamic pyruvic transaminase activity at high dose. Lymphocyte subpopulations (T and B cells) were induced at lower dose levels but suppressed at the highest dose group. A significant decrease in urine volume and an increase in blood urea nitrogen were observed at the highest dose. Biochemical analysis of bronchoalveolar lavage fluid and lung tissue showed some lung damage. No damage to the testes was observed. No gross or microscopic changes were seen in any of the tissues examined. The NOAEL was 4 mg nickel/kg bw/day. (Obone *et al.*, 1999).

60. Four groups of albino Wistar rats (25 each sex) were fed diets containing 0, 100, 1000 or 2500 ppm nickel (corresponding to 0, 5, 50 or 125 mg nickel/kg bw/day) for 2 years. Bodyweight was recorded weekly and food consumption was measured over 3-day periods at the end of 1, 3, 6, 12, and 24 months. Two-year survival was poor, particularly amongst control animals of both sexes and males in the high dose group, but there was no indication that this was an effect of nickel. Nickel had a depressant effect on body weight in both sexes at the high dose and sporadically for rats in the mid dose group. Food consumption indicated no consistent trends, but it appeared that the lesser weight gains, particularly in the high dose group, may be in part a result of lower food consumption. Hematologic values for haemoglobin, haematocrit and differential leukocyte counts, obtained at 3-month intervals, for rats of all dietary levels of nickel did not depart significantly from those of the controls. Results of tests for urinary reducing substance at three month intervals were negative. Results of semiquantitative tests for urinary protein at the same time intervals were quite variable and inconsistent, with no clear trends. A tendency toward increased heart-to-body weight ratios and decreased liver-to-body weight ratios appears in female rats in the mid- and high-dose groups. Gross pathologic and histologic findings on rats sacrificed at term were negative. The NOAEL was 5 mg nickel/kg bw/day. (Ambrose *et al.*, 1976)

61. Groups of 3 male and 3 female purebred beagle dogs of about 6-months of age, individually housed, were maintained for two years on diets providing 0, 100, 1000 and 2500 mg nickel/kg food (0, 1.8, 18, and 45 mg nickel/kg bw/day). All dogs survived the two year experimental period. During

the first 3 days, all 6 dogs on the high dose diet vomited, usually within an hour. On the 4th day they were returned to the control diet. All but one dog readjusted within 3 days. The one dog readjusted after parenteral feeding and i.v. fluids. At the start of the second week 5 of the dogs were placed on 1500 ppm nickel and the sixth dog was included at the start of the sixth week. This level of nickel apparently was well tolerated, as no emesis, salivation or gastro-intestinal irritation was observed. At two-week intervals the diet level of nickel was raised to 1700, 2100 and 2500 ppm, respectively, with no further evidence of emesis, salivation, or gastrointestinal irritation. After 2 years decreased bodyweight was observed at the highest dose. Haematologic values obtained at 3 month intervals were quite variable but within normal range. There was a tendency toward lower haematocrit and haemoglobin values in dogs in the highest dose group, suggestive of a simple hypochromic anaemia. Marked polyuria was noted in 2 dogs at the highest dose. Relative kidney and liver weights were higher at the highest dose. At the highest dose, all dogs showed lung lesions and 2 dogs had granulocytic hyperplasia of the bone marrow. The NOAEL was 18 mg nickel/kg bw/day. (Ambrose *et al.*, 1976)

62. Nickel sulphate hexahydrate was administered by daily oral gavage to Fischer 344 rats at levels of 0,10, 30 and 50 mg nickel sulphate hexahydrate/kg bw/day (corresponding to 0, 2.2, 6.7, and 11.2 mg nickel/kg bw/day) (60 animals per sex per group). There was no apparent treatment-related effect on mortality in treated males (60, 48, 50 and 57 % in control, low, mid and high dose, respectively). Not all mortalities were related to treatment: a higher rate of mortality was observed in treated animals during the first 24 weeks of the study that were secondary to aspiration of nickel sulphate solution. Starting during week 24 and continuing through the remainder of the study, oral exposure was delayed in the morning, in order to allow time for gastric emptying to occur. The change in exposure time was effective in increasing survival. Bodyweight decreased in an exposure-dependent manner, with statistical significance at the 2 highest doses. No treatment-related effects were observed on clinical signs, hematology, biochemistry, urinalysis parameters, gross pathology or histopathology. The NOAEL was 2.2 mg nickel/kg bw/day. (Heim *et al.*, 2007).

63. Pandey *et al.* (1999) reported an accumulation of nickel in the epididymis, testes, seminal vesicles and prostate gland in male mice exposed by gavage to 5 or 10 mg nickel sulphate/kg bw/day (corresponding to 1.1 or 2.2 mg nickel/kg bw/day) (5 days/week) for 35 days. There was no change in bodyweight, but a decrease in weights of testes, epididymis, seminal vesicles and prostate gland was observed. The accumulation of nickel in male reproductive tissues resulted in histopathological damages in these tissues (at 2.2 mg nickel/kg bw/day atrophy of centrally located tubules and disturbed spermatogenesis (decrease in sperm motility and total sperm count), damages in epididymis were observed) and sperm damages. In addition, male mice from the control group and exposed to 2.2 mg nickel/kg bw/day for 35 days were mated with untreated females. A decrease in the fertility index was observed in the treated group. In females mated with treated males a

decrease in the number of pre- and post-implantations and an increase in resorptions were observed. A decrease in weight was also observed in foetuses from dams mated with treated males. The authors concluded that the testicular and spermatotoxic changes may be responsible for observed male mediated developmental toxic effects. (Pandey *et al.*, 1999).

64. Pandey and Srivastava (2000) reported dose-related decreases in weights of reproductive organs (testes, epididymis, seminal vesicles and prostate gland), in mice exposed by gavage to 20 mg nickel sulphate or nickel chloride/kg bw/day for 35 days. Decreases in sperm motility and count and increases in abnormal sperm were observed at 10 and 20 mg nickel sulphate or nickel chloride/kg bw (corresponding to 2.2/2.5 and 4.5/5 mg nickel/kg bw). At comparable doses, the spermatotoxic effects were of higher severity for nickel chloride than for nickel sulphate. The NOAEL was 5 mg nickel sulphate or nickel chloride/kg bw/day (1.1/1.3 mg nickel/kg bw/day). The authors concluded that the abnormal and non-motile sperm may reduce the fertilising capacity of spermatozoa and adversely affects the fertilisation of the ovum. The CONTAM Panel noted that in this study only a limited number of parameters have been investigated – bodyweight gain, male reproductive organ weights and sperm parameters- and that only 6 males were tested per group. (Pandey and Srivastava, 2000; EFSA, 2015).

65. Young male Swiss albino mice were given a daily oral dose of 0 (0.9 % NaCl) or 20 mg nickel sulphate/kg bw/day (corresponding to 0 or 4.5 mg nickel/kg bw/day) for 5 days/week for 6 months. There was no sign of toxicity in any of the treated animals, but after 6 months of exposure, mean bodyweight was reduced in treated animals. The urinary excretion of protein (testosterone-dependent) was lower in treated mice compared with controls. Testicular weight and histology did not differ in the 2 groups. Lower weight and smaller size (diameter) of the seminal vesicles was observed in exposed males. There was also a lower secretory activity of the cells of the vesicular epithelium. Nickel accumulated in the interstitial tissue of the testes. These effects are similar to those expected when the seminal vesicle is subjected to decrease testosterone levels. The authors concluded that the decreased production of testosterone may therefore be an early effect of long-term nickel exposure. (Pandey and Singh, 2001).

66. Toman *et al.* (2012) demonstrated the adverse effect of nickel on the mouse testis structure from 3 to 12 weeks of administration in feed of 10 mg nickel chloride/kg bw/day. The most vulnerable site is the seminiferous epithelium which undergoes degeneration and the germ cells desquamate from the Sertoli cells connections in the tubule lumen creating empty spaces in the epithelium and die. The interstitial tissue was also significantly affected. The changes of the testis become more visible the longer are the periods of nickel exposure. This study shows that the oral administration of nickel causes serious damage to the spermatogenesis and development of the testis structure, when administered for long-term to young mice at the beginning of their sexual maturity. (Toman *et al.*, 2012)

Table 1. A summary of toxicity studies

Study (Doses in mg nickel/kg bw/day)	NOAEL (mg nickel/kg bw/day)	LOAEL (mg nickel/kg bw/day)	Reference
Repeat dose toxicity studies with nickel compounds			
40-day oral M mouse Nickel chloride (0, 0.5, 2 and 4)	-	0.5	Gathwan *et al.*, (2013)
13-week oral (drinking water) Male rat Nickel sulphate hexahydrate (0, 4, 10 and 20)	4	10	Obone *et al.*, (1999)
91-day oral (gavage) Rat Nickel chloride hexahydrate (0, 5, 35 and 100)	-	5	American Biogenics Corporation (1988)
90-day oral (gavage) Rat Nickel sulphate 0, 11, 17, 22, 28(7), 33(3) Reduction of dose in 2 high dose groups on day 28	7	11	Rush (2002) SLI (2002)
2-year study oral (diet) Rat Nickel sulphate hexahydrate (0, 5, 50 and 125)	5	50	Ambrose *et al.*, (1976)
2-year study oral (diet) Dog Nickel sulphate hexahydrate (0, 1.8, 18 and 45)	18	45	Ambrose *et al.*, (1976)
2-year oral (gavage) Rat Nickel sulphate hexahydrate (0, 2.2, 6.7 and 11.2)	2.2	6.7	Heim *et al.*, (2007)
Developmental and reproductive studies with nickel compounds: Reproductive toxicity: 1-3 generations studies			
2-GEN study oral (drinking water) Rat Nickel chloride hexahydrate (0, 6.0/6.2, 25/23, and 42/42 Average exposure premating/mating period: Males (0, 4, 19 and 31) Females (0, 3, 12 and 22)	Parental toxicity: 25 Reproduction toxicity: 42 Offspring toxicity: 6	Parental toxicity: 42 Reproduction toxicity: - Offspring toxicity: 25	RTI (1988)

Exposure ranges gestation period: (5-6, 22-26, 33-44) Exposure ranges post natal period(GD20 – PND21) (4-13, 12-58, 14-98)			
3-generation study oral (diet) Rat 30M and 30F/group (F0, F1b, F2b) → after 11 wk: 20F mated with 20M Nickel sulphate hexahydrate (0, 5, 50 and 125)	Parental toxicity: 50 Reproductive toxicity: 125 Offspring toxicity: -	Parental toxicity: 125 Reproductive toxicity: - Offspring toxicity: 5	Ambrose *et al.*, (1976)
1-generation oral (gavage) Rat Nickel sulphate hexahydrate (0, 2.2, 4.4, 6.6, 11 and 17)	Parental and reproductive toxicity: 17 Offspring toxicity: -	Parental and reproductive toxicity: - Offspring toxicity: 2.2	SLI, (2000a)
2-GEN oral (gavage) Rat Nickel sulphate hexahydrate (0, 0.2, 0.6, 1.1 and 2.2)	Parental, reproductive and offspring toxicity: 2.2	Parental, reproductive and offspring toxicity: -	SLI, (2000b)
2-litter study 11-week prior to mating + during 2 successive gestation + lactation periods Oral (drinking water) F Rat Nickel chloride (0, 1.3, 6.8 and 31.6) Mated with untreated M	Maternal toxicity: 1.3 Offspring toxicity: -	Maternal toxicity: 6.8 Offspring toxicity: 1.3	Smith *et al.*, (1993)
Developmental and reproductive studies with nickel compounds: Reproductive organs toxicity			
2-year study oral (diet) Rat Nickel sulphate hexahydrate (0, 5, 50 and 125)	Systemic toxicity: 5 Reproductive toxicity: 125	Systemic toxicity: 50 Reproductive toxicity: -	Ambrose *et al.*, (1976)
2-year study oral (diet) Dog Nickel sulphate hexahydrate (0, 1.8, 18 and 45)	Systemic toxicity: 18 Reproductive	Systemic toxicity: 45 Reproductive	Ambrose *et al.*, (1976)

	toxicity: 45	toxicity: -	
13-week oral (drinking water) M Rat Nickel sulphate (0, 4, 10 and 20)	Systemic toxicity: 4 Reproductive toxicity: 20	Systemic toxicity: 10 Reproductive toxicity: -	Obone *et al.*, (1999)
91-day oral (gavage) Rat Nickel chloride hexahydrate (0, 5, 35 or 100)	Systemic toxicity: - Reproductive toxicity: 100	Systemic toxicity: 5 Reproductive toxicity: -	American Biogenic Corporation, (1988)
35-day study oral (gavage) (5 days/week) M mouse Nickel sulphate 0, 1.1, or 2.2)	Systemic toxicity: 1.1 Reproductive toxicity: -	Systemic toxicity: 2.2 Reproductive toxicity: 1.1	Pandey *et al.*, (1999)
35-day study oral (gavage) (5 days/week) M mouse Nickel sulphate (0, 1.1, or 2.2) Nickel chloride (0, 1.3, 2.5 or 5)	Systemic toxicity: 1.1 (sulphate) 1.3 (chloride Reproductive toxicity: -	Systemic toxicity: 2.2 (sulphate) 2.5 (chloride) Reproductive toxicity: 1.1 (sulphate) 1.3 (chloride)	Pandey and Srivastava, (2000)
6-month study oral (gavage) (5 days/week) M mouse Nickel sulphate (0 or 4.5)	Systemic and reproductive toxicity: -	Systemic and reproductive toxicity: 4.5	Pandey and Singh, (2001)
3-, 6-, 9- and 12-week oral (pellets) M mouse Nickel chloride (0 or 2.5)	-	2.5	Toman *et al.*, (2012)
Developmental and reproductive studies with nickel compounds: Reproductive organs toxicity			
35-day study oral (gavage) (5 days/week) M mouse Nickel sulphate (0 or 2.2) Mated with untreated females	-	2.2	Pandey *et al.*, (1999)

(15 dams/dose)			

2-GEN: 2-generation; bw: bodyweight; F: female; GD: gestation day; M: male; PND; post-natal day

Proposed TDI

67. In the SLI 2-generation study (2000b) nickel sulphate hexahydrate was administered to rats by oral gavage over 2 generations. There was a slight reduction in adult male liver weight in F0 males dosed with 2.2 mg/kg bw/day and in the F1 males dosed with 1.1 and 2.2 mg/kg bw/day. There were no treatment-related histopathological finding in the liver or other tissues from rats dosed with 2.2 mg.kg bw/day. The dose level of 2.2 mg/kg bw/day was therefore considered the no observed adverse effect level (NOAEL).

68. From the selected NOAEL of 2.2 mg/kg bw/day a TDI of 22 µg/kg bw was derived by applying a default uncertainty factor of 100 to account for extrapolation from experimental animals to humans and for inter-individual variability.

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Secretariat

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COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT

First draft statement on potential risks from nickel in the diet of infants aged 0 to 12 months and children aged 1 to 5 years

Prevalence of nickel sensitisation in infants and young children aged 1 to 5 years

Introduction

1. Following a request from Members at the December 2016 meeting a literature search was undertaken specifically to look for information regarding nickel sensitisation in infants and young children.

2. Some studies provided information about the prevalence of nickel sensitisation in the general population of infants and young children. However, most studies reported on nickel sensitisation in infants and young children who were patients at dermatology clinics. Table 1 provides a summary of the references discussed in this annex. The patch testing used in the studies described below tests for a number of allergens simultaneously. Where possible, the information on nickel has been drawn out but on occasions, responses to any allergen have been described.

3. Nickel can cause allergic contact dermatitis (ACD), i.e. type IV hypersensitivity, in the general population. There has been an increase in nickel sensitisation which represents an increase in nickel exposure, especially ear/body piercings (even in young children), toys and clothing fasteners (Heim and McKean, 2009; Mortz *et al.*, 2002; Jensen *et al.*, 2014). The consumption of nickel-rich foods may elicit eczematous flare-up reactions in the skin in sensitised individuals. However, experimental studies have also shown that repeated oral exposure to nickel may reduce or prevent sensitisation. (EFSA, 2015)

Nickel sensitisation

4. A study by Jensen *et al.* (2003) investigated reactions to oral nickel stimuli in healthy and nickel sensitised adults. Sixty volunteers took part in a double-blind oral nickel exposure study. Of these volunteers 40 were nickel sensitive and 20 were healthy controls. All enrolled volunteers were patch tested with 3 concentrations (equivalent to 5, 1.66 and 0.55 % Ni) of nickel and a vehicle control. One month after patch testing to confirm the nickel sensitisation status, volunteers were orally exposed to nickel. The 40 nickel

sensitive volunteers were exposed to 0.3, 1, or 4 mg nickel or placebo. The healthy controls were exposed to placebo or 4 mg nickel.

5. Of the nickel sensitive volunteers 7/10 had cutaneous reactions to the 4 mg nickel, 4/10 reacted to the 1 mg nickel, 4/10 had a reaction to the 0.3 mg nickel and 1/10 reacted to the placebo. None of the volunteers in the control group had a cutaneous reaction to either of the oral exposures.

6. The authors found dose dependency in the nickel content of both serum and urine after oral nickel exposure. Higher oral exposure correlated with higher levels of nickel in urine and serum. The differences between the nickel content in the urine of clinically cutaneous reacting and non-reacting nickel-sensitive individuals were also significant.

7. The nickel content in the serum of nickel-sensitive volunteers who reacted to the 4.0 mg or 1.0 mg nickel was higher than the content in serum of nickel-sensitive volunteers who did not react. The opposite was true when 0.3 mg nickel or a placebo was given. No similar shift was observed in the nickel content of urine. (Jensen *et al.*, 2003).

Nickel sensitisation in infants and young children

General population studies

8. Eleven out of 85 (12.9 %) asymptomatic infants and children (attending routine well-child care at Denver area paediatric practices), aged 6 months to 5 years, tested positive when challenged with nickel sulphate. The youngest responder was 6 months old. Seven of the children were less than 18 months old. A single patient had a history of rash over the earlobes that occurred after wearing costume jewellery. This patient was sensitised to nickel but had discontinued earring use (Bruckner, Weston and Morelli, 2000).

9. Weston tested 314 children (129 of which were aged 6 months to 5 years) with 20 allergens for 48 hours and read at 72 hours. Twenty four (7.6 %) children reacted to nickel sulphate. The number of children (20 %) with positive patch tests to any allergen in the 6 months to 5 years age group was very similar to the numbers in other age groups (% children reacted): 5 to 12 years (20 %) and adolescents (21 %) (Weston *et al.*, 1986).

10. In a study by Barros 562 children aged between 5 and 14 years from the Oporto area were patch tested with 25 allergens. Five children, who had no previous history of allergy to jewellery or metallic clothing accessories, tested positive to nickel sulphate (0.9 % of the total population). The highest percent of positive reactions (20.2 %) to any allergen was in the youngest age group (5 to 6 years old) (Barros *et al.*, 1991).

Studies in patient groups

11. Between 1995 and 2004 five hundred children were referred to the Department of Dermatology at Leeds General Infirmary for patch test investigations and patch tested with a number of different chemicals. Of the 133 (27 %) positive tests 44 (8.8 %) were to nickel sulphate. The mean ± SD age of patients with a positive patch test was 12 ± 3.8 years (median 16, range 0-16). This was significantly higher than that of patients with a negative result (mean ± SD 10 ± 4.5 years, median 11, range 0-16). Girls were significantly more likely to have a positive patch test result than boys. Of the children with positive reactions a personal history of atopy was found in 81 and 55 had a diagnosis of atopic dermatitis. Ear piercing appears to be an important cause for nickel allergy. Girls are more likely to have their ears pierced and this was reflected in the results for positive tests to nickel. (Clayton *et al.*, 2006).

12. In a study by Seidenari (2005) 1094 patients, aged from 7 months to 12 years, with suspected allergic contact dermatitis, were patch tested with 30 or 46 allergens depending on age. 570 had positive patch test results, 119 of which were sensitised to nickel. The sensitisation rate was significantly higher in children less than 3 years of age (63.4 % compared to 48.4 % and 45.6 % for children aged 4 – 8 and 9 – 12 years, respectively). Girls (13.5 %) were significantly more likely to be sensitised to nickel than boys (7.9 %). This was also observed in small children. As only 17% of this age group had piercings the authors surmised that ear piercing did not seem to be a risk factor for nickel allergy in this population. There was a significant increase in sensitisation prevalence in comparison with the findings for 1988-1994, published previously (Manzini, 1998). No differences in the sensitisation rate between atopic dermatitis patients and nonatopic ones were observed. (Seidenari *et al*., 2005).

13. Fortina (2011) patch tested 321 children under 3 years of age, with suspected contact dermatitis, with 30 allergens. Of the children tested 200 responded with at least one positive result. The most common allergen was nickel sulphate (26.8 %). The number of positive reactions showed that sensitisation rates in children aged up to 2 years were similar to those aged between 2 and 3 years. Males (68.1 %) were found to have a statistically higher sensitisation rate than females (57.5 %) ($P < 0.05$) in contrast to the results from most other studies, and with what happens in older children. Children less than 3 years of age were found to have the same prevalence of contact dermatitis irrespective of whether or not they had allergic dermatitis. (Fortina *et al.*, 2011).

14. A study by Hammonds, Hall and Yiannias (2009) retrospectively reviewed patch test data from 2000-2006. Patch tests were carried out on 136 children aged 3 to 18 years. Out of 135 patients tested for nickel allergy, 30 were positive (22 %). A positive reaction to nickel was observed in 35 % of the population of 3 to 10 year olds (n = 26) compared to 20 and 18 %, respectively of 11 to 15 (n = 55) and 16 to 18 (n = 55) year olds. The authors observed that more males tested positive to any allergen than females in the 3 to 10 year olds group but the percentage of females testing positive was

greater than the number of males in the 11 to 15 and 16 to 18 year old groups. (Hammonds, Hall and Yiannias, 2009).

15. Seventy nine patients aged 1-18 years were patch tested in a study by de Waard-van der Spek and Oranje (2009). Of the 40 children that had a positive result to one or more allergens, 17 had a positive reaction to nickel sulphate, 2 of whom were less than 5 years old. This study reported that allergic contact dermatitis (ACD) increased with age. (de Waard-van der Spek and Oranje, 2009).

Table 1. Summary of the percentage of study populations testing positive to nickel

Number of participants	Age (years)	Sensitised to nickel (%)	Reference
General population			
85	0.5 to 5	13	Bruckner, Weston and Morelli, 2000
314	0.5 to 5	7.6	Weston et al., 1986
562	5 to 14	0.9	Barros *et al.*, 1991
Studies in patient groups			
500	0 to 16	8.8	Clayton *et al.*, 2006
1094	0.5 to 12	11	Seidenari *et al*., 2005
321	≤3	27	Fortina *et al.*, 2011
136	3 to 18	22	Hammonds, Hall and Yiannias, 2009
79	1 to 18	22	de Waard-van der Spek and Oranje, 2009

Oral desensitisation

16. 2176 patients entered the study and filled in the questionnaire. Patients who had received a dental brace within 1 year before or after ear piercing were excluded, as were patients who had worn a brace for less than 6 months and/or had not answered the question of ear piercing and/or the questions of orthodontic treatment in the interview. When wearing of braces preceded ear piercing, nickel allergic contact hypersensitivity was significantly reduced (in females 25.0 % versus 39.3 % and in males 7.7 % versus 22.5 %) (Van Hoogstraten *et al.*,1991).

17. A cohort of 1,501 12-16 year old children in Odense, Denmark were patch tested and 8.6 % tested positive for nickel sensitisation. This is similar to other studies in which the prevalence of nickel sensitisation in children and adolescents in the general population varies from 0.9 to 14.9 %. The application of dental braces prior to ear piercing was associated with a significantly reduced frequency of nickel allergy in girls compared to the use of dental braces after ear piercing. There were no significant associations between nickel allergy and atopic dermatitis or inhalant allergy (Mortz et al., 2002).

Effect of nickel in ambient air on nickel sensitisation

18. A study by Kasper-Sonnenberg (2011) measured nickel in ambient air in four study areas (Krefeld, Witten, Bochum and Siegen) near stainless steel mills in North Rhine-Westphalia, Germany. Average annual nickel concentration for all study areas together was 11.3 ng/m^3 (mean) and ranged from 4.0 to 140.4 ng/m^3. (Ranges of median and 99th percentile nickel values of 0.27 to 6.80 and 2.23 to 56.23 ng/m3, respectively, were measured in particulate matter in the UK (TOX/2016/41)). A total of 749 children eligible to start school (aged 5 to 6 years) and 720 mothers took part in the study and had urinary nickel concentrations measured and were patch tested for nickel sensitisation. 594 and 539 samples of urine from children and mothers, respectively were analysed for nickel. The rounded range from the 5th to the 95th percentile of the distributions in the whole study group were used as the unit of exposure change for nickel in urine (7.1 µg/L). The unit of exposure change in ambient air was 10 ng/m^3. An increase of nickel in ambient air of 10 ng/m3 yielded a significant elevation of urinary nickel concentrations in both mothers (9.1% ; 95% CI: 6.8-11.4%) and children (2.4%; 95% CI: 0.4-4.4%).

19. The prevalence of nickel sensitisation in children in the standard patch test was 10.9% (N=504). There was a higher prevalence for boys than for girls (12.2% versus 9.7%). An increase of the nickel concentration in ambient air by 18 ng/m^3 led to an increased prevalence of nickel sensitisation (OR: 2.4; 95% CI: 1.25-1.32). Higher urinary nickel concentration was also associated with increased prevalence of nickel sensitisation (OR: 2.4; 95% CI: 1.28-4.48). (Kasper-Sonnenberg *et al.*, 2011).

20. Studies by Mann (2010) and Wilhelm (2007) measured nickel in ambient air in 3 locations in North Rhine Westphalia. A total of 309 children of school starter age from Duisburg (coking plant, a steel mill, a sintering plant and a blast furnace) (N=103), Dortmund (a steel mill) (N=101) and Borken (rural town) (N=105) were included in the analysis and patch tested with 24 allergens including nickel (II) sulphate. The prevalence rates of nickel sensitisation were 13.3% (Borken), 30.7% (Dortmund) and 12.6% (Duisburg). Nickel was measured in urine in 210 of these children. There was a statistically significant association between nickel sensitisation and internal nickel exposure as urinary nickel values of sensitised children were significantly higher than that of non-sensitised children. There was also a

significant association between mean annual exposure to nickel in ambient air and nickel sensitisation. (Mann *et al.*, 2010; Wilhelm *et al.*, 2007).

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COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT

First draft statement on potential risks from nickel in the diet of infants aged 0 to 12 months and children aged 1 to 5 years

Introduction

1. The Scientific Advisory Committee on Nutrition (SACN) is undertaking a review of scientific evidence that will influence the Government's dietary recommendations for infants and young children. The SACN is examining the nutritional basis of the advice. The Committee on Toxicity in Food, Consumer Products and the Environment (COT) was asked to review the risks of toxicity from chemicals in the diet of infants, most of which has been completed, and young children. The reviews will identify new evidence that has emerged since the Government's recommendations were formulated, and will appraise that evidence to determine whether the advice should be revised. The recommendations cover diet from birth to age five years.

2. This statement gives an overview of the potential risks from nickel in the diets of infants and young children in the UK aged 0 to 12 months and 1 to 5 years, respectively.

Background

3. Nickel is a metal that exists in various mineral forms and is present throughout the environment. It is used in a wide variety of processes including electroplating and alloy production, and is present in a wide range of consumer products. Nickel concentrations in the environment reflect both natural and anthropogenic contributions. Although it can exist in various oxidation states, the divalent form of nickel (Ni(II)) generally occurs in food and water as this is its most stable oxidation state (EFSA, 2015).

4. The general population is primarily exposed to nickel via food and drinking water, with inhalation from ambient air and percutaneous exposure acting as generally minor sources of exposure. Food is generally considered to be a more important source of oral exposure to nickel than drinking water (EFSA, 2015; EVM, 2003; WHO, 2005).

5. Following oral exposure in humans, nickel is bioavailable at levels from 1% up to 40%. Bioavailability appears to be lower when exposure to nickel occurs in the presence of food or under non-fasted conditions, than when nickel is dosed in drinking water alone (EFSA, 2015). It has been reported

that typically <10% of nickel ingested with food is absorbed, while nickel from drinking water alone is more highly absorbed at ~20-25% (EVM, 2002 and 2003). In rats nickel is rapidly but poorly absorbed following ingestion; absorption has been reported to range from 0.01 to 33.3% depending on the solubility of the nickel compound that had been ingested. Absorbed nickel can bind to serum proteins and is widely distributed in the organism. Absorbed nickel is mainly excreted via urine; it is excreted to a lower extent in breast milk. An estimated elimination half-life of 28 ± 9 hours was calculated in human volunteers (EFSA, 2015).

6. Although nickel is an essential micronutrient for higher plants and some animal species, there are currently no data proving that it is essential for humans (EFSA, 2015).

7. In humans, the non-carcinogenic effects of oral exposure to nickel include effects on the gastrointestinal, haematological, neurological, and immune systems. Gastrointestinal (i.e. vomiting, abdominal cramps, and diarrhoea) and neurological (i.e. giddiness, headache, and weariness) symptoms are the most reported effects after acute exposure. Exposure to nickel through skin or by inhalation may lead to nickel sensitisation; although oral exposure is not known to lead to sensitisation, it may be able to elicit eczematous flare-up reactions in the skin of nickel-sensitised individuals (EFSA, 2015).

8. Oral ingestion of nickel salts in experimental animals has resulted in a wide range of adverse effects including nephrotoxicity, hepatotoxicity and metabolic effects. Nickel is able to cross the placental barrier and exerts its primary toxic effects in experimental animals by affecting the developing embryo or fetus. Increases in pre- and perinatal mortality have been reported in the offspring of female rats ingesting nickel salts. The currently available epidemiological data do not support an association between dietary nickel exposure and reproductive and developmental effects in humans (EFSA, 2015).

9. The COT has commented on nickel in food a number of times in the past; the general conclusion has been that dietary exposures to nickel were unlikely to be of toxicological concern. The Committee has also concluded that although nickel may exacerbate contact dermatitis/eczema in sensitised individuals, pre-school children are less likely than adults to be sensitised and would therefore not be considered to be a sensitive sub-group (COT, 2008).

Expert opinions

10. An expert opinion on exposure to nickel in food and drinking water has been published by the European Food Safety Authority's (EFSA) Panel on Contaminants in the Food Chain (CONTAM) (EFSA, 2015). The Expert Group on Vitamins and Minerals (EVM) reviewed nickel in their report on the 'Safe Upper Levels of Vitamins and Minerals' (EVM, 2003). The World Health Organization (WHO) has reviewed exposures to nickel via drinking water as

part of the development of their 'Guidelines for Drinking Water Quality' (WHO, 2005 and 2011). The International Agency for Research on Cancer (IARC) has published an evaluation of the carcinogenicity of nickel and nickel compounds (IARC, 2012).

11. At the previous meeting, Members considered that COT should review the relevance of the EFSA TDI based on developmental toxicity for infants and young children. Text reflecting the outcome of this discussion will be included in a subsequent draft.

Nickel exposures in infants aged 0 to 12 months and young children aged 1 to 5 years

Sources of nickel exposure

Human breast milk

12. In general, low levels of nickel are found in breast milk (EFSA, 2015).

13. Two concentrations of nickel in breast milk were selected from the literature. A minimum value of 0.13 µg/L (Krachler *et al.*, 2000) and a maximum value of 47 µg/L (Bjorklund *et al.*, 2012) have been used to represent the low and high levels of nickel measured in samples of breast milk. These values are taken from studies in European countries but the minimum and maximum nickel concentrations, from the UK data (Woolridge *et al.*, 2004), are within this range of values.

Infant formulae and food

14. Concentrations of nickel have recently been measured in an FSA survey of metals and other elements in infant formulae and foods (e.g. commercial infant foods) (referred to as the Infant Metals Survey), and in the composite food samples of the 2014 Total Diet Study (TDS).

Food contact materials

15. The migration of nickel from food contact materials could represent an additional source for the presence of nickel in food and drinking water. In general, nickel-containing food contact materials are made of highly corrosion resistant stainless steel so that the metal should not migrate into food in quantities that would endanger human health. Stainless steel products are used in food transportation, for food processing equipment and containers, for cooking utensils and tableware, and for electric kettles and other kitchen appliances. Nickel may also be released from nickel-plated kitchenware; although nickel-plating is less resistant to corrosion than stainless steel so nickel-plated articles are not normally used for materials that are meant to come into contact with food (EFSA, 2015).

16. At present, as recommended by the Council of Europe, manufacturers of food preparation and handling tools and equipment made of stainless steel should respect the migration of nickel compliant with a specific release limit (SRL) of 0.14 mg/kg food (EDQM, 2013; EFSA, 2015).

17. The EFSA CONTAM Panel concluded that the extent of nickel migration due to the use of good quality stainless steel in food contact materials has likely little or no relevance compared to the dietary exposure determined by the intrinsic presence of nickel in the diet. However, leaching of nickel may not be negligible for food contact materials made of poor quality stainless steel, or of other nickel-containing metal alloys (EFSA, 2015).

18. The EVM reported that the quantity of nickel released from food cooked in “already used” stainless steel pans was low to negligible (< 0.07 µg/g), and although release from pans on their first use was higher (up to 0.27 µg/g), the amounts released were still considered relatively small (EVM, 2002).

19. The EFSA stated that the potential leaching of nickel into food from food contact materials was not covered by the occurrence dataset that was used to estimate dietary exposure (EFSA, 2015). The 2014 TDS food samples were prepared ‘as consumed’ prior to analysis and thus any potential levels of nickel leached into food from food contact materials will be reflected in the overall concentration. However, this is not the case for the samples of the infant metals survey.

Drinking water

20. The primary source of nickel in drinking water is leaching from metals in contact with drinking water, such as pipes and taps. Although the nickel concentration in drinking water is normally less than 20 µg/L, release from such metal fittings could contribute up to 1 mg/L. Nickel may also be present in some groundwater as a consequence of dissolution from nickel ore-bearing rocks (WHO, 2005 and 2011).

21. EU legislation sets a value of 20 µg/L for nickel in water intended for human consumption (Directive 98/83/EC), and a maximum level of 20 µg/L in natural mineral waters (Directive 2003/40/EC). The WHO has established a guidance level of 70 µg/L for nickel in drinking water, but has stated that a concentration of 20 µg/L should be achievable by conventional water treatment (WHO, 2011).

22. Levels of nickel in drinking water in 2014/2015 from England and Wales, Northern Ireland and Scotland were provided by the Drinking Water Inspectorate (DWI), Northern Ireland Water and the Drinking Water Quality Regulator (DWQR) for Scotland, respectively. Median and 97.5th percentile values calculated from this data are shown in Table 1. These values have been used to calculate exposures to nickel from drinking water in combination with exposures from food.

Table 1. Median and 97.5th percentile concentrations (µg/L) of nickel in water across the UK for 2014/2015.

Country	Number of samples	Limit of Detection (µg/L)	Median concentration (µg/L)	97.5th Percentile concentration (µg/L)
England and Wales	14708	0.8-2.0*	1.36	4.63
Northern Ireland	392	0.4	1.14	4.47
Scotland	1500	0.2	0.30	1.95

* The DWI noted that the water companies had reported a range of LODs that varied with the analytical method used, and clarified that the relevant drinking water regulations specify that the LOD must not be more than 10% of the prescribed value (20 µg/L for nickel)

Environmental

Dust

23. The Agency for Toxic Substances and Disease Registry (ATSDR) of the US Department of Health and Human Services advises that nickel concentrations in household dust can be high and therefore pose an increased risk to young children who have greater contact with floors (ATSDR, 2005).

24. The median and maximum nickel concentrations of 53.3 and 97.1 mg/kg respectively, reported by Turner and Simmonds (2006), have been used in this assessment to estimate exposures to nickel via dust for UK infants and young children.

Soil

25. Nickel is present at about 20 mg/kg in the Earth's upper continental crust (Rawlins *et* al., 2012). It occurs naturally at high levels in some types of rock, and is released to soils from anthropogenic activities such as smelting, disposal of sewage sludge, and emissions from motor vehicles and electric power utilities, and from natural activities such as weathering and erosion of geological materials. The EFSA have estimated that soil ingestion by children would make a low contribution to their nickel exposure (EFSA, 2015).

26. In 2012 and 2013, Defra published normal background concentrations (NBCs) for nickel in soil in England and Wales (Defra, 2012 and 2013). An NBC is the 95th percentile upper confidence interval of the available data; it is defined as a contaminant concentration that is seen as typical and widespread in topsoils (depth 0 - 15 cm). In order to establish meaningful NBCs, the available soil data were grouped in domains (e.g. principal, urban, and ultrabasic) that were defined by the most significant controls on a contaminant's high concentrations and distribution. The NBCs for each

domain in England and Wales were published following a Defra-commissioned BGS project to define the typical background concentrations for soil contaminants.

27. As part of the BGS project, summary statistics were derived from topsoil data from 2 or 3 core datasets held for England and Wales (Ander *et al*., 2012 and 2013). Although the NBCs and summary statistics were derived for several domains for England and Wales, the most significant domain for each country was the principal domain. The principal domains are areas which do not contain significantly elevated levels of nickel. Overall, for England and Wales, the area covered by the principal domains constitutes approximately 99% and 94% of each country respectively. The summary statistics reported for the principal domain in England were a median of 23 mg/kg and a 95th percentile of 42 mg/kg (n = 41,768 samples). The statistics reported for the same domain in Wales were a median of 22 mg/kg and a 95th percentile of 38 mg/kg (n = 1,327 samples).

28. The highest median and 95th percentile concentrations for nickel in soil from the Defra-commissioned BGS project on NBCs (23 and 42 mg/kg respectively) have been used to estimate exposures to soil in this assessment. These data have been used as they are recent, and represent a relevant domain for estimating exposure for the general population.

Air

29. In the atmosphere nickel occurs mainly as fine respirable particles (<2 µm) and is eventually suspended onto particulate matter. Anthropogenic sources account for more than 80% of the atmospheric nickel burden, with the remainder accounted for by natural sources such as soil dust, volcanoes and forest fires (EFSA, 2015).

30. EU legislation sets a target value of 20 ng/m^3 for nickel in air (Directive 2004/107/EC). Annual mean ambient particulate phase concentrations of nickel in the urban environment are typically of the order of 1 ng/m^3 with the exception of a few industrial areas, where higher annual means may occur, in some locations exceeding the target value of 20 ng/m^3 (Defra, 2015).

31. Nickel in particulate matter less than 10 µm (PM_{10}) was measured at 23 sites and as metal deposition was measured at 5 sites across the UK in 2014/2015. Median values from these sites ranged from 0.27 to 6.80 ng/m^3 and 99th percentile values ranged from 2.23 to 56.23 ng/m^3. One site in Wales was excluded from the analysis as it regularly measured much higher values than any other site (Defra, 2015).

Exposure assessment

32. Consumption data (on a bodyweight basis) from the Diet and Nutrition Survey of Infants and Young Children (DNSIYC) (DH, 2013), and the National Diet and Nutrition Survey Rolling Programme (NDNS) (Bates *et al.*,

2014) have been used for the estimation of dietary exposures for ages 4 to 18 months, and 18 to 60 months respectively. Bodyweight data used in the estimation of other nickel exposures are shown in Table 2 below.

33. Thorough exposure assessments have been performed for the dietary sources of exposure to nickel. The assessments for the non-dietary sources of exposure (i.e. dust, soil and air) have been included to give a more holistic view of exposures, but are not as thorough, as the focus of this statement is the diet of infants and young children.

34. It is possible for infants and young children to be sensitised to nickel through an increased exposure to nickel in the environment, especially costume jewellery (especially ear piercings), clothing fasteners and toys (Heim and McKean, 2009; Mortz *et al.*, 2002; Jensen *et al.*, 2014). Possible flare-up reactions may be caused by exposure to high levels of nickel in food. Therefore acute exposures have been considered. For each exposure source there is a comment to describe the possible increase for an acute exposure. All of the estimated acute exposures are less than 2-fold higher than the corresponding chronic exposures.

Table 2. Average bodyweights used in the estimation of nickel exposures

Age group (months)	Bodyweight (kg)
0 to <4	5.9[a]
4 to <6	7.8[b]
6 to <9	8.7[b]
9 to <12	9.6[b]
12 to <15	10.6[b]
15 to <18	11.2[b]
18 to <24	12.0[c]
24 to <60	16.1[c]

[a] DH, 1994
[b] DH, 2013
[c] Bates *et al.*, 2014

Infants (0 to 12 months)

Breast milk

Chronic

35. No consumption data were available for exclusive breastfeeding in infants aged 0 to 6 months. Therefore, the default consumption values used by the COT in other evaluations of the infant diet of 800 and 1200 mL for average and high level consumption have been used to estimate exposures to nickel from breastmilk. These estimates were based on low and high nickel

concentrations of 0.13 and 47 µg/L, respectively (Paragraph 13). The ranges of exposure to nickel in exclusively breastfed 0 to 6 month olds were 0.01 to 0.02 and 0.02 to 0.03 µg/kg bw/day in average and high level consumers respectively with a nickel concentration of 0.13 µg/L. For breast milk with a nickel concentration of 47 µg/L the ranges of exposures to nickel were 4.8 to 6.4 and 7.2 to 9.6 µg/kg bw/day in average and high level consumers respectively (Table 3).

Table 3. Estimated nickel exposure from exclusive breastfeeding in 0 to 6 month old infants, with breast milk containing nickel at 0.13 and 47 µg/L.

Nickel concentration (µg/L)	Exposure (µg/kg bw/day)			
	Average consumer (800 mL/day)		High consumer (1200 mL/day)	
	0 to <4 months	4 to <6 months	0 to <4 months	4 to <6 months
0.13	0.02	0.01	0.03	0.02
47	6.4	4.8	9.6	7.2

Values rounded to 2 significant figures (SF)

36. Data on breast milk consumption for infants aged 4 to 18 months were available from the DNSIYC and the NDNS, and have been used to estimate exposures at these ages (Table 6), based on low and high nickel concentrations of 0.13 and 47 µg/L. There were too few records of breast milk consumption for children older than 18 months in the NDNS to allow a reliable exposure assessment, and breast milk is expected to contribute minimally in this age group.

37. Mean exposures to nickel for 4 to 18 month olds with a breast milk nickel concentration of 0.13 µg/L were 0.003 to 0.01 µg/kg bw/day, and 97.5th percentile exposures were 0.01 to 0.02 µg/kg bw/day. A nickel concentration in breast milk of 47 could lead to mean nickel exposures of 1.2 to 4.3 µg/kg bw/day and 97.5th percentile exposures of 2.4 to 7.3 µg/kg bw/day (Table 4).

Table 4. Estimated chronic nickel exposure in 4 to 18 month old infants from breast milk

Nickel concentration in breast milk (µg/L)	4 to <6 months (n=116)		6 to <9 months (n=606)		9 to <12 months (n=686)		12 to <15 months (n=670)		15 to <18 months (n=605)	
	Mean	97.5th	Mean	97.5th	Mean	97.5th	Mean	97.5th	Mean	97.5th
0.13	0.01	0.02	0.01	0.02	0.005	0.02	0.004	0.01	0.003	0.01
47	4.3	7.3	3.1	7.5	1.8	5.4	1.4	3.5	`1.2	2.4

Values rounded to 2 SF

Acute

38. Acute mean and high-level exposure to nickel from breast milk for infants and young children were up to 1.7-fold higher than corresponding chronic exposures from this source.

Infant formulae and complementary foods

Chronic

39. Nickel exposure estimates for this category were derived using occurrence data from the Infant Metals Survey (FSA, 2016a), based on both lower bound (LB) and upper bound (UB) concentrations. Exposure estimates for 0 to 6 month olds were calculated for exclusive feeding on infant formulae using the default consumption values of 800 and 1200 mL (Table 7). Consumption data from the DNSIYC were used to estimate exposures for 4 to 12 month olds (DH, 2013) (Table 8).

40. In 0 to 6 month olds, exposures to nickel from ready-to-feed formula were 0 to 1.2 µg/kg bw/day in average consumers, and 0 to 1.8 µg/kg bw/day in high level consumers. Exposures to nickel calculated for reconstituted formula incorporating the water concentration from the TDS, and the highest median and 97.5th percentile concentrations for nickel in water reported in Table 3 were 0.40 to 2.0 µg/kg bw/day in average consumers, and of 0.60 to 3.0 µg/kg bw/day in high level consumers (Table 5).

Table 5. Estimated average and high level exposures to nickel from exclusive feeding on infant formulae for 0 to 6 month olds.

Infant Formula	Nickel exposure (LB-UB Range) (µg/kg bw/day)			
	0 to <4 months		4 to <6 months	
	Average consumer	High level consumer	Average consumer	High level consumer

	(800 mL/day)	(1200 mL/day)	(800 mL/day)	(1200 mL/day)
Ready-to-Feed [a]	0 - 1.2	0 - 1.8	0 - 0.92	0 - 1.4
Dry Powder [b, c]	0.37 - 1.1	0.55 - 1.6	0.28 - 0.83	0.42 - 1.2
Dry Powder [c] + TDS water of <8 µg/L [d]	1.3 - 2.0	1.9 - 3.0	0.98 - 1.5	1.5 - 2.2
Dry Powder [c] + median water of 1.36 µg/L [d]	0.53 - 1.3	0.79 - 1.8	0.40 - 0.94	0.60 - 1.4
Dry Powder [c] + 97.5th percentile water of 4.63 µg/L [d]	0.90 - 1.6	1.4 - 2.4	0.68 - 1.2	1.0 - 1.8

Values rounded to 2 SF
[a] Exposure based on first milk infant formula using LB to UB nickel concentrations of 0-9 µg/L
[b] Exposure does not include the contribution from water
[c] Exposure based on first milk infant formula using LB to UB nickel concentrations of 18-54 µg/kg
[d] Calculated assuming reconstituted formula comprises 85% water

41. Total mean exposures (excluding water) to nickel from infant formulae, commercial infant foods, and other foods, for 4 to 12 month olds were 1.2 to 2.9 µg/kg bw/day, and 97.5th percentile exposures were 3.9 to 5.9 µg/kg bw/day. Detailed exposure assessments for 4 to 18 month old infants and young children are provided in Annex B. Total mean and 97.5th percentile exposures were also calculated using the highest median and 97.5th percentile concentrations for nickel in water reported in Table 1. The resulting total mean and 97.5th percentile exposures indicated that levels of nickel in water made a negligible contribution to total exposure (Table 6).

Table 6. Estimated chronic exposures to nickel from infant formulae, commercial infant foods and other foods for infants aged 4 to 12 months

Food	Nickel Exposure (LB-UB Range) (µg/kg bw/d)					
	4 to <6 Months (n=116)		6 to <9 Months (n=606)		9 to <12 Months (n=686)	
	Mean	97.5th	Mean	97.5th	Mean	97.5th
Infant formula	0.0044 - 0.59	0.030 - 1.3	0.0031 - 0.43	0 - 0.98	0.0021 - 0.31	0 - 0.68
Commercial infant foods	0.64 - 0.84	2.3 - 3.0	0.89 - 1.2	3.0 - 4.0	0.80 - 1.1	3.0 - 4.2
Other foods	0.46 - 0.57	2.8 - 3.0	0.82 - 1.1	2.9 - 3.6	0.96 - 1.5	3.0 - 4.0
Total (excl. water)	1.2 - 2.1	4.1 - 5.7*	1.8 - 2.8	3.9 - 5.7*	1.8 - 2.9	4.4 - 5.9*

Values rounded to 2 SF
* Determined from a distribution of consumption of any combination of categories rather than by summation of the respective individual 97.5th percentile consumption value for each of the three food categories

Acute

42. Mean and 97.5th percentile acute exposures to total nickel were estimated to be between 1.2- and 1.4-fold higher than corresponding total chronic exposures.

Children aged 12 to 18 months

43. Estimated exposures to nickel from food for children aged 12 to 18 months were calculated using occurrence data from both the Infant Metals Survey (FSA, 2016a), and the 2014 TDS (FSA, 2016b). The exposure data derived from the Infant Metals Survey allow estimation of nickel exposure in infant formula, commercial infant foods and the most commonly consumed adult foods ('other foods') as sold, whereas the results from the TDS are based on analysis of food that is prepared as for consumption. In addition, the Infant Metals Survey included analysis of infant formulae and commercial infant foods which are not included in the TDS. Exposure estimates based on both LB and UB concentrations are provided.

44. The consumption data from the DNSIYC were used for the estimation of exposure for children aged 12 to 18 months (DH, 2013).

Exposure estimates based on the Infant Metals Survey

Chronic

45. The ranges of chronic total mean and 97.5th percentile exposures (excluding water) to nickel from infant formula, commercial infant foods and other foods were 1.3 to 2.5 and 2.8 to 5.2 µg/kg bw/day, respectively. As for infants, the total mean and 97.5th percentile exposures including water (calculated using the highest median and 97.5th percentile values in Table 3) were equal to those estimated for the total mean exposures excluding water (Table 7).

Table 7. Estimated chronic exposures to nickel from infant formulae, commercial infant foods and other foods for children aged 12 to 18 months

Food	Nickel Exposure (LB-UB Range) (µg/kg bw/d)			
	12 to <15 Months (n=670)		15 to <18 Months (n=605)	
	Mean	97.5th	Mean	97.5th
Infant formula	0.00050 - 0.13	0 - 0.57	0.00030 - 0.070	0 - 0.42
Commercial infant foods	0.45 - 0.60	2.0 - 2.8	0.24 - 0.32	1.2 - 1.7
Other Foods	0.96 - 1.7	2.6 - 3.8	1.0 - 1.8	2.5 - 3.5
Total (excl. water)	1.4 - 2.5	3.6 - 5.2*	1.3 - 2.2	2.8 - 4.3*

Values rounded to 2 SF
* Determined from a distribution of consumption of any combination of categories rather than by summation of the respective individual 97.5th percentile consumption value for each of the three food categories

Acute

46. Mean and 97.5th percentile acute exposures to total nickel exposure were estimated to be between 1.4- and 1.5-fold higher than corresponding total chronic exposures.

Exposure estimates based on the TDS

Chronic

47. Table 8 shows the estimated nickel exposures calculated using the TDS data for children aged 12 to 18 months. The nickel concentration for the tap water group in the TDS was reported to be below the limit of detection (LOD) of 8 µg/L. This LOD is higher than that reported for nickel in tap water by the water authorities across the UK (Table 1). The calculation was therefore also performed using the highest median (1.36 µg/L) and 97.5th percentile (4.63 µg/L) nickel concentration in tap water reported in Table 1.

48. Total mean and 97.5th percentile exposures to nickel from a combination of all food groups are in the region of 3.7 to 5.0 and 7.7 to 8.8 µg/kg bw/day, respectively (Table 8). These are higher than those estimated from the Infant Metals Survey due to the inclusion of a greater number of foods in the exposure estimate for the TDS. Overall the figures in Table 8 demonstrate that the nickel content of water has a negligible impact on total dietary exposure to nickel of young children in the UK.

Table 8. Estimated chronic dietary exposure to nickel based on the TDS data in children aged 12 to 18 months.

Nickel concentration in the water	**Nickel Exposure (LB-UB Range) (µg/kg bw/day)**			
	12 to <15 Months (n=670)		**15 to <18 Months (n=605)**	
	Mean	**97.5th**	**Mean**	**97.5th**
1.36 µg/L	3.7 - 4.5	7.7 - 8.6	4.0 - 4.9	7.8 - 8.7
4.63 µg/L	3.7 - 4.5	7.7 - 8.6	4.0 - 4.9	7.9 - 8.7

Values rounded to 2 SF

49. In general, the food groups making the highest contribution to nickel exposure were miscellaneous cereals (includes pasta and rice products), poultry and potatoes groups (FSA, 2016b).

Acute

50. Mean and 97.5th percentile acute exposures to total nickel exposure were estimated to be between 1.3- and 1.4-fold higher than corresponding total chronic exposures.

Children aged 18 months to 5 years

51. Exposure estimates for these age groups were derived using occurrence data from the 2014 TDS, and consumption data from the NDNS (Bates *et al*., 2014).

Chronic

52. Table 9 shows the nickel exposures that were calculated using the TDS data for children aged 18 months to 5 years. Detailed exposure assessments are presented in Annex C. As described in paragraph 80, the exposures have been estimated using the TDS water concentration (8 µg/L, the LOD), and the highest median (1.36 µg/L) and 97.5th percentile (4.63 µg/L) nickel concentrations in water reported in Table 1. This results in total mean and 97.5th percentile exposures to nickel from a combination of all food groups of 4.3 to 5.7 and 7.1 to 8.7 µg/kg bw/day, respectively (Table 9). Overall the figures in Table 9 demonstrate that the nickel content of water has

a negligible impact on total dietary exposure to nickel of young children in the UK.

Table 9. Estimated chronic dietary exposure to nickel in children aged 18 months to 5 years.

Nickel concentration in water	Nickel Exposure (LB-UB Range) (µg/kg bw/day)			
	18 to <24 Months (n=70)		24 to <60 Months (n=429)	
	Mean	97.5th	Mean	97.5th
1.36 µg/L	4.7 - 5.6	7.5 - 8.7	4.3 - 5.0	7.1 - 8.0
4.63 µg/L	4.8 - 5.6	7.5 - 8.7	4.4 - 5.0	7.1 - 8.0

Values rounded to 2 SF

53. As with the younger children, the food groups making the highest contribution to nickel exposure in the TDS were miscellaneous cereals (includes pasta and rice products), poultry and potatoes groups (FSA, 2016b).

Acute

54. Mean and 97.5th percentile acute exposures to total nickel exposure were estimated to be between 1.3- and 1.5-fold higher than corresponding total chronic exposures.

Dust

55. Potential exposures of UK infants aged 6 to 12 months and young children aged 1 to 5 years to nickel in dust were calculated assuming ingestion of 30 or 60 mg/day, respectively (US EPA, 2011a). Younger infants, who are less able to move around and come into contact with dust, are likely to consume less dust than children of these age groups. Median and maximum nickel concentrations in dust of 53.3 and 97.1 mg/kg, respectively, were used to estimate average and high level exposures (paragraph 24) (Table 10).

Table 10. Possible nickel exposures from dust in infants and young children aged 6 months to 5 years.

Nickel concentration (mg/kg)	Exposure (µg/kg bw/day)					
	Age (months)					
	6 to <9	9 to <12	12 to <15	15 to <18	18 to <24	24 to <60
53.3 (Median)	0.18	0.17	0.30	0.29	0.27	0.20
97.1 (Maximum)	0.34	0.30	0.55	0.52	0.49	0.36

Values rounded to 2 SF

Soil

56. Potential exposures of UK infants aged 6 to 12 months and young children aged 1 to 5 years to nickel in soil were calculated assuming ingestion of 30 or 50 mg/day, respectively (US EPA, 2011a). Younger infants, who are less able to move around and come into contact with soil, are likely to consume less soil than children of these age groups. Median and 95th percentile soil nickel concentrations of 23 and 42 mg/kg respectively were used in these exposure estimations (paragraph 28) (Table 11).

Table 11. Possible nickel exposures from soil in infants and young children aged 6 months to 5 years.

Nickel concentration (mg/kg)	Exposure (µg/kg bw/day)					
	Age (months)					
	6 to <9	9 to <12	12 to <15	15 to <18	18 to <24	24 to <60
23 (Median)	0.079	0.072	0.11	0.10	0.096	0.071
42 (95th percentile)	0.14	0.13	0.20	0.19	0.18	0.13

Values rounded to 2 SF

Air

57. Potential exposures of UK infants aged 0 to 12 months and young children aged 1 to 5 years to nickel in air were estimated using the body weights shown in Table 2, and by assuming the mean ventilation rates presented in Table 12; these rates have been derived from the US EPA exposure factors handbook (US EPA, 2011b). The resulting exposures are presented in Table 13.

Table 12. Mean ventilation rates used in the estimation of nickel exposures from air (derived from US EPA, 2011b)

Age group (months)	Ventilation rate (m^3/day)
0 to <4	3.5
4 to <6	4.1
6 to <9	5.4
9 to <12	5.4
12 to <15	8.0
15 to <18	8.0
18 to <24	8.0
24 to <60	10.1

58. The nickel concentrations used in the exposure calculations were the lowest and highest median values and lowest and highest 99th percentile values of 0.27, 6.80, 2.23 and 56.23 ng/m^3, respectively, from monitoring sites in the UK (paragraph 31).

Table 13. Possible exposures to nickel in infants and young children from air

Nickel concentration (ng/m^3)	Exposure (µg/kg bw/day)							
	Ages (months)							
	0 to <4	4 to <6	6 to <9	9 to <12	12 to <15	15 to <18	18 to <24	24 to <60
0.27 (lowest median value)	0.00016	0.00014	0.00017	0.00015	0.00020	0.00019	0.00018	0.00017
6.80 (highest median value)	0.0040	0.0036	0.0042	0.0038	0.0051	0.0049	0.0045	0.0043
2.23 (lowest 99th percentile value)	0.0013	0.0012	0.0014	0.0013	0.0017	0.0016	0.0015	0.0014
56.23 (highest 99th percentile value)	0.033	0.030	0.035	0.032	0.042	0.040	0.037	0.035

Values rounded to 2 SF

Risk Characterisation

59. This section will be completed in the next draft of this document, once a TDI value has been agreed upon by Members.

Secretariat
January 2017

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