EFSA MEETING SUMMARY REPORT

Folic acid: an update on scientific developments

21-22 January 2009, Uppsala, Sweden

Table of contents

Acknowledgements

The organisers of the meeting and the ESCO Working Group on the "Analysis of risks and benefits of fortification of food with folic acid" would like to thank all the participants for their valuable contributions at the meeting and their help in the preparation of this report. In particular, many thanks go to the overall rapporteur Åke Bruce, the Food Standards Agency (UK), and the EFSA Scientific Cooperation Unit for their help in drafting and editing of this report.

Disclaimer

The information contained in this publication does not necessarily reflect the opinion or the position of the European Food Safety Authority.

1.0 INTRODUCTION

1. In 2008, the European Food Safety Authority (EFSA) established an EFSA Scientific Cooperation Working Group (ESCO WG) on the "Analysis of risks and benefits of fortification of food with folic acid", with the aim of sharing experiences and concerns regarding folic acid food fortification amongst Member States.

2. The ESCO WG on folic acid was asked to consider the following issues as part of their terms of reference:
   - To review current practice in Member States regarding the level of voluntary fortification of foods and categories of foods to which the addition of folic acid is allowed.
   - To consider new evidence regarding the risk of high intakes of folic acid and the need for a review of current guidance on safe upper levels of folic acid for all population groups.

3. EFSA and the Swedish National Food Administration organised a scientific meeting on *"Folic Acid: An Update on Scientific Developments"*, in Uppsala, Sweden, on 21-22 January 2009. The aim of the meeting was to consider the evidence regarding folic acid and risk of cancer.

4. Over 60 scientific experts from the European Union (EU), Switzerland, the United States and Canada attended the meeting to assess the latest scientific evidence on the possible relationship between dietary intakes (including fortified foods and food supplements) of folate and folic acid, and cancer risks, including cancer of the colon, breast and prostate.

5. All the available scientific evidence concerning folate metabolism, animal and mechanistic studies, and human studies was reviewed and discussed. In group discussions, experts considered whether it was possible: to identify an association of folic acid intake with risk of cancer; the population groups concerned; dose-response relationships; the different dietary sources of folic acid; and whether the available data were sufficient to allow a quantitative risk assessment. Areas for further scientific research were also identified.

6. Since the completion of this report, further papers on folic acid and cancer risk have been published. Only papers and presentations presented at the meeting are considered in this report.

2.0 BACKGROUND SESSIONS

7. Folate is a generic term for a naturally occurring family of B-group vitamins comprising an aromatic pteridine ring linked to p-aminobenzoic acid and one or more glutamate residues. It is found naturally in a variety of foods including green leafy vegetables, fruit, liver, and yeast. Folic acid is a synthetic form of folate which is widely used in supplements and for food fortification. Folic acid is more stable in foods and is better absorbed than natural folates.

8. Dietary folates are converted in the intestinal mucosa to 5-methyl tetrahydrofolic acid (5-MTHF) which is the form of folate present in the systemic circulation. Folic acid has to be reduced and methylated in the gut mucosa before it can be converted to 5-MTHF, the form found in the circulation. The capacity of the body to convert folic acid to 5-MTHF is limited and unmetabolised folic acid has been detected in the systemic circulation following folic acid supplementation (from both supplements and fortified foods) at oral doses above 260 µg (Kelly et al., 1997).

2.1 Benefits and potential risks

9. Randomised controlled trials have conclusively demonstrated that folic acid supplementation can prevent up to two-thirds of neural tube defects (NTDs) (MRC Vitamin Study Research Group, 1991). It might also reduce the risk of other congenital malformations such as orofacial clefts. The effectiveness of mandatory folic acid fortification programmes in the USA and Canada have resulted in significant declines in the occurrence of NTD affected pregnancies (Williams et al., 2005; De Wals et al., 2007). The percent declines range from 28% to 46% in the USA and Canada respectively.

10. Findings from observational studies had also suggested that high intakes of folate (or high blood levels of folate) were associated with a lower risk of cardiovascular disease (CVD) and cancer, and less-age related cognitive decline. Randomised trials had not confirmed any such associations with CVD and cancer. Although limited data from randomised trials have generally not demonstrated any significant beneficial or adverse effects of folic acid on cognitive function, one randomised controlled trial (Durga et al., 2007) reported that folic acid supplementation had a beneficial effect on improving cognitive function in older adults with low folate status and without vitamin B12 deficiency.

11. High intakes of folic acid have also been associated with theoretical risks of adverse effects. Since high dosages of folic acid can correct the anaemia associated with vitamin B12 deficiency, there are concerns that high intakes of folic acid could delay the diagnosis of vitamin B12 deficiency by treating ("masking") the anaemia of vitamin B12 deficiency which could lead to irreversible neurological damage if treatment with vitamin B12 is not provided. However, current medical practice does

not rely on the presence of anaemia for the detection of vitamin B12 deficiency, which frequently presents without anaemia.

12. While generally, observational studies have suggested that folic acid supplementation slows down the rate of cognitive decline with age, some have suggested that it may accelerate it.

13. Other postulated adverse effects of folic acid supplementation include reducing the efficacy of antifolate drugs such as methotrexate used in chemotherapy for cancer treatment and drugs used to treat epilepsy but this research question has been insufficiently studied. Concerns have also been raised about the presence of unmetabolised folic acid in the blood following folic acid at oral doses of 260 µg or greater (see paragraph 7). However, the current available data are insufficient to adequately assess the long-term effects of exposure to unmetabolised folic acid.

14. There are also data suggesting the possibility that high folic acid intakes may be associated with increased risks of cancer; the evidence suggesting a potential link relates specifically to folic acid. There is no evidence to suggest that high intakes of natural folates found in foods are associated with increased cancer risk.

15. A possible role of folic acid in cancer development is supported by biologically plausible mechanisms. Folate is essential in biological methylation reactions and nucleotide synthesis and impairment of these processes are thought to be involved in cancer development. The evidence regarding folic acid and cancer risk is considered in section 2.5.

2.2 Current recommendations

16. Many countries in the EU recommend that women who might become pregnant should take folic acid supplements to reduce the risk of NTD occurrence, but public health campaigns promoting this advice have been unsuccessful in most Member States. Directive 2002/46/EC on the approximation of the laws of Member States relating to food supplements establishes harmonised rules for the labelling of food supplements and introduces specific rules on vitamins and minerals in food supplements in the EU.

17. Some countries in the EU have considered mandatory fortification of wheat flour or bread as a strategy to reduce the prevalence of NTDs. Mandatory wheat flour fortification is currently under review in the United Kingdom but has not been endorsed in Sweden or Italy. It has been recommended in Ireland but implementation has been deferred.

18. Voluntary fortification is permitted in most European countries. There is considerable variation across the EU in the levels of folic acid that have been added to foods on a voluntary basis, and variation in the categories of foods that are

fortified. Recently the EU introduced new rules to regulate voluntary food fortification. These are set out in Regulation (EC) No 1925/2006 on the addition of vitamins and minerals and of certain other substances to foods. As part of the implementation of this regulation, work is currently ongoing on the setting of maximum amounts for the addition of vitamins and minerals. Maximum amounts will be set concurrently for vitamins and minerals in fortified foods and in food supplements (European Commission, 2006).

2.3 Recommended upper intake levels for folic acid in Europe

19. In 2000, the Scientific Committee on Food (SCF) set a tolerable upper intake level (UL) for folic acid of 1 mg/day for adults (SCF, 2000). ULs for adolescents and children were adjusted downwards on the basis of body weight. The UL is an estimate of the highest level of usual intake of a nutrient which carries no appreciable risk of adverse health effects. The UL was based on the risk of progression of neurological symptoms caused by the "masking" of the haematological symptoms of vitamin B12 deficiency.

2.4 Relation of dietary sources of folic acid to blood folate concentrations

20. There are two sources of folic acid: foods fortified with folic acid and supplements containing folic acid. Data from the National Health and Nutrition Examination Survey (NHANES) 2001-2004 of adults in the USA showed that higher intakes of folic acid and higher blood folate concentrations were primarily associated with use of supplements containing folic acid and were moderately associated with intake of voluntarily fortified foods containing folic acid; lower folic acid intake from mandatory fortification was not associated with these higher values (Yeung et al., 2008).

2.5 Folic acid and cancer risk

Animal studies

21. Animal models have suggested the possibility of a dual role of folic acid in cancer development, depending on the timing and dose of the intervention: high intakes may suppress development of early lesions in normal tissue but increase the progression of established neoplasms.

22. Data from animal studies suggest that animals maintained on folate deficient diets are at increased risk of colorectal cancer and that modest folic acid supplementation reduces this risk (Kim, 2004). However, in folate replete animals, and animals with

established lesions, high doses of folic acid increase the risk of colorectal cancer (Kim, 2003).

Human studies

Ecological studies

23. In the USA, voluntary fortification of enriched grain products with folic acid was first authorised in March 1996 and compliance became mandatory from January 1998. In Canada, fortification of foods with folic acid was permitted in December 1996 and cereal grains, especially white flour, were mandated to be fortified with folic acid since November 1998.

24. Time trends in colorectal cancer incidence in the USA and Canada between 1986 and 2002 indicated an abrupt reversal in the downward trend in colorectal cancer incidence between 1996 and 1998 at around the time of the introduction of folic acid fortification. The downward trend later resumed with the incidence curve shifted upwards because of the temporary increase. Mason et al. (2007) hypothesised that folic acid fortification may have been responsible for the significant deviation from the pre 1996 trend resulting in an excess of about 4-6 additional cases of colorectal cancer cases per 100,000 individuals.

25. This type of ecological evidence cannot exclude the possibility that the observed fluctuations in colorectal cancer were due to improved screening programmes for colorectal cancer. While there was an increase in colorectal cancer incidence at around the time of the introduction of folic acid fortification, there was no corresponding increase in colorectal cancer mortality, which is consistent with the fluctuations being due to improved screening rather than increased incidence of cancer. However, cancer mortality may not be a useful endpoint in this context as an ecologic study can not take account of the effects on cancer mortality of new cancer treatments that became available in the 1990s.

Observational studies

26. Several epidemiological studies have explored associations of folate intake and blood folate concentrations with cancer, and in particular with colorectal or breast cancer.

27. Although the results are inconsistent, most studies of folate intake and colorectal cancer risk suggest a protective effect of high folate intakes on colorectal cancer risk. Studies of serum folate and colorectal cancer risk are inconclusive. Several studies using folate biomarkers are difficult to compare due to, for example, different analytical matrices (serum, plasma, or blood).

28. The available epidemiological studies of folate and breast cancer risk have reported that folate intake or folate status is unrelated with breast cancer risk, but some studies have suggested an increased risk of breast cancer associated with low folate intake combined with alcohol consumption. One observational study (Stolzenberg-Solomon et al., 2006) reported that folic acid supplements of 400µg or more per day may be associated with an increased risk of breast cancer in postmenopausal women (hazard ratio: 1.19; 95% CI, 1.01-1.41) compared with women consuming no folic acid supplements.

29. Since there is potential for differential effects of natural dietary folates obtained from food and folic acid from fortified foods and supplements it is important to clearly distinguish between the two. However, many epidemiological studies did not distinguish between intakes of natural folates from foods and folic acid from supplements and fortified foods. Some studies addressed this issue indirectly by examining the use of supplements; other studies considered intakes of natural food folates and total folate intake separately.

30. Findings from epidemiological studies come from observations that could be confounded by other dietary and non-dietary factors associated with cancer risk.

MTHFR gene variants and cancer risk

31. Genetic variability of a number of enzymes that are involved in folate metabolism can modify their activity and affect folate status. Several studies have investigated associations of polymorphic genes involved in folate metabolism with colorectal and breast cancer risk. Most studies have considered the MTHFR 677 C→T and 1298 A→C polymorphisms, which are associated with high homocysteine levels in the setting of low folate status. Most, but not all studies, have reported reduced colorectal cancer risk associated with the MTHFR 677TT variant. The MTHFR 1298 A→C polymorphism has been less extensively studied, and results have been inconsistent (Sharp & Little, 2004; Hubner & Houlston, 2006; Huang et al., 2007).

32. Genetic variability in folate metabolism is still inadequately characterised and the ability to jointly investigate multiple factors in a biological pathway is very limited.

Randomised controlled trials

33. Data from randomised controlled trials on the effects of folic acid intakes on breast, prostate and other cancers are limited. One study (Charles et al., 2004), which followed up approximately 3000 women that had participated in a folic acid supplementation trial during pregnancy reported an increased risk of all cancer and a trend for an increased breast cancer risk in women who had been supplemented with 5 g/d of folic acid. However this study was not designed to test the hypothesis that folic acid supplementation has an effect on cancer risk and the study design and statistical analysis may not have been appropriate. A trial that examined the efficacy

of folic acid (1 mg/day) for prevention of recurrent colorectal adenomas (n=1021) reported that folic acid supplementation was associated with a significantly increased risk of prostate cancer. However, the authors noted that this could be a spurious finding given the number of adverse events evaluated. This trial is described in further detail in paragraph 36.

34. Two categories of randomised controlled trials have provided evidence on effects of folic acid on risk of cancer and in particular on colorectal cancer: (i) those which have investigated the effects of folic acid supplementation for the prevention of new recurrent colorectal adenomas in individuals with a previous history of colorectal adenomas and (ii) those which have investigated the effect of B-vitamins (including folic acid) on CVD risk, which also collected data on cancer outcomes.

Colorectal adenoma prevention trials

35. Four small randomised controlled trials (Paspatis and Karmanolis, 1994; Cole et al., 2007; Jaszewski et al., 2008; Logan et al., 2008) and one unpublished UStrial (E.Giovannucci 2009 personal communication) have assessed the effect of folic acid supplementation on the risk of colorectal adenoma recurrence in individuals with a prior history of colorectal adenomas. Only the trial by Cole et al. (2007) had duration of more than 3-4 years.

36. Paspatis and Karamanolis (1994) reported that folic acid supplementation (1mg/day for 2 years; n=60) decreased colorectal adenoma risk compared with placebo, although the differences were not statistically significant; Jaszewski et al. (2008) reported that folic acid supplementation (5 mg/day for 3 years; n=93) significantly reduced adenoma recurrence compared with the placebo group. The results from these two small trials suggested that folic acid supplementation reduced the risk of colorectal adenoma. The results of these small trials need to be treated with caution as they are likely to be statistically underpowered.

37. Cole et al. (2007) investigated the effect of folic acid supplementation (1 mg/d; n=1021) with or without aspirin for up to 8 years. This trial reported that folic acid supplementation did not prevent the development of colorectal adenomas. There was no difference in the incidence of at least 1 colorectal adenoma between the placebo group and the folic acid groups after 3 years (RR, 1.04; CI, 0.90-1.20; p=0.58) or after 6 years (RR, 1.13; CI, 0.93-1.37; p=0.23). However, during subsequent treatment/follow-up in a sub-group analysis of this trial (n=607) there was a significantly greater incidence of advanced lesions in the folic acid group compared to the placebo group (RR, 1.67; CI, 1.00-2.80; p=0.05) and significantly more people in the folic acid group with 3 or more adenomas (RR, 2.23; CI, 1.23-4.35).

38. Results of the trial by Cole et al. (2007) suggested that folic acid at doses in excess of 1 mg/day may increase the risk of developing multiple/advanced adenomas after a few years' delay and consequently increase the risk of colorectal cancer.

39. The trial by Logan et al. (2008) reported that folic acid supplementation (0.5 mg/day for 3 years; n=853) did not have a significant effect on adenoma recurrence (RR, 1.07; 95% CI, 0.85-1.34). The unpublished US trial (E.Giovannucci, personal communication, 2009) also found no effect of folic acid supplementation (1mg/day for 3 years; n=692) on colorectal adenoma recurrence.

40. Of the three larger trials (n=700 to 1000) participants received 0.5mg/day of folic acid in one study (Logan et al., 2008) and 1 mg/day in the other two studies (Cole et al., 2007; unpublished trial). Only the trial by Cole et al. (2007) followed participants for more than 3 years and increased risks were observed in the longer follow-up (6-8 years). The trial by Logan et al. (2008) and the unpublished trial both had short follow-up periods (3-4 years); risk ratios from these trials are consistent with those reported by Cole et al. (2007) during their first follow-up (3-4 years). A meta-analysis (n=2652) of the results from these 3 trials (Cole et al., 2007; Logan et al., 2008; unpublished trial) found no evidence of any significant effects of folic acid supplementation on any cancer in this population (unpublished results). This meta-analysis was limited to the shorter follow-up time frame of 3-4 years.

CVD prevention trials

41. A number of intervention trials have investigated B-vitamin supplementation (including folic acid) for prevention of cardiovascular disease (CVD) in people with a prior history of CVD or renal disease. These trials also examined effects of folic acid supplementation on overall risk of cancer, cancer at specific sites, and mortality from cancer.

42. Few of the individual trials of B-vitamin supplements for prevention of vascular diseases had adequate statistical power to assess the effects of B-vitamins on CVD or on cancer. The B-Vitamin Treatment Trialists' Collaboration (BVTT) was set up as a prospective meta-analysis of results from all the B-vitamin trials in order to provide more reliable evidence for the effects of B vitamins on vascular and non-vascular outcomes (unpublished results).

43. The preliminary results of the BVTT meta-analysis of 8 of the trials, involving 37,485 participants, found no significant beneficial or adverse effects of B-vitamin supplementation (folic acid dose of 0.8-40mg/day for a median duration of 5 years) on vascular events, all-cause mortality, cancer, or cancer in any of the pre-specified sub-groups or at any specific sites (including colorectal, lung, prostate or breast cancer (unpublished results). The interpretation of these results is limited by the short duration of follow-up in comparison to the longer periods of time over which cancers usually develop.

44. Results from a sub-group of two of the B-vitamin trials from Norway (NORVIT & WENBIT) involving 6837 participants with an additional three years of follow-up after the end of the intervention period were due to be presented in June 2009 at the International Homocysteine Conference in Prague (http://www.homocysteine2009.org/).

3.0 REPORTS FROM DISCUSSION GROUPS

3.1 Discussion Group 1: Folic acid and colorectal cancer risk

45. The available evidence on the associations of folic acid with cancer was considered hierarchically.

Animal studies

46. Although animal studies are useful for exploring potential mechanisms, caution should be exercised in their interpretation and extrapolation to humans. For example, the doses of folic acid used in animal studies are 4 to 10 times higher than the expected intakes from folic acid food fortification.

Human studies

Ecological evidence

47. This type of evidence is useful for generating hypotheses but should be treated with caution because of a number of inherent limitations.

48. A number of points were raised in relation to the study by Mason et al. (2007), including:

- Uncertainty regarding the precise timing of the increase in the population exposure to folic acid in relation to the upturn in colorectal cancer incidence.
- The plausibility of an immediate cancer effect, although this finding is consistent with a possible very late and immediate progression of established adenomas to colorectal cancer.
- Improvements in screening for colorectal cancer in the USA occurred at around the same time as the introduction of folic acid fortification and this could have accounted for the increase in colorectal cancer incidence. Sudden increases in cancer incidence can be caused by a change in screening practice or data collection (case ascertainment, definition, or diagnostic practice). Although this is supported by the fact that there was no subsequent increase in colorectal cancer mortality, the introduction of new chemotherapeutic agents in this time period may have had positive effects on cancer mortality rates.

49. It was agreed that, as an ecological study, the paper by Mason et al. (2007) had a number of limitations. However, the paper had raised issues about the safety of folic acid and had also highlighted the importance of monitoring trends in colorectal incidence for countries that decide to introduce mandatory fortification with folic acid in the future.

Observational studies

50. Although the results are inconsistent, most observational studies have shown a protective effect of higher intakes of total folate on colorectal cancer risk compared to those with the lowest folate intakes. Most studies investigated total dietary folate and did not distinguish between natural folates and folic acid.

51. Epidemiological data on folate (natural folates and folic acid contained in supplements and fortified foods) and cancer risk were reviewed by the World Cancer Research Fund (WCRF/AICR, 2007). The WCRF concluded that there is limited evidence suggesting a protective effect of folate against colorectal cancer (based on papers published before 2006). The report noted, however, uncertainty because of potential confounding and effect modification (particularly with intake of dietary fibre). The WCRF report did not distinguish between folic acid from supplements/fortified foods and natural folates.

52. It is not possible to reach conclusions about folate and potential colorectal cancer risk from observational data because of problems with assessment of dietary folate intake, potential confounding with other factors that may affect cancer risk and effect modification by other factors that could interfere in 1-carbon metabolism (particularly B vitamins or other methyl donors). Associations between folic acid and potential cancer risk in epidemiological studies may also differ due to pre-existing supplement use or voluntary fortification status in the studied populations.

Randomised controlled trials

53. Of the five randomised controlled trials which assessed the effect of folic acid supplementation (0.5-1mg/day) on risk of recurrence of colorectal adenomas in people with a prior history of colorectal adenoma (see paragraphs 34-39), none reported adverse effects within 3 years of folic acid supplementation. Only one randomised controlled trial (Cole et al., 2007) reported data on follow-up of more than 3 years; this trial reported that during the later treatment/follow-up, folic acid supplementation (1mg/d) was associated with more multiple, advanced, and larger (unpublished information) adenomas compared with the placebo group. It was agreed that results from this study raise concerns about long-term exposure to folic acid.

54. The BVTT meta-analysis showed no evidence of any significant effect of folic acid supplementation on overall risk of cancer (Unpublished). There were extensive discussions on the power of this meta-analysis to detect an association between folic acid and cancer risk. It was agreed that an adequately powered meta-analysis for site-specific cancers such as colorectal cancer would not be possible because of the very large numbers of people that would be required and it was therefore unlikely that this question could be resolved in the near future. It was also agreed that the current data involved relatively short follow-up time periods in comparison to the time usually required for the development of cancers.

55. It was noted that cancer endpoints from 3 further B-vitamin trials would be included in the meta-analysis in 2009 and 2011 and that 2 Norwegian studies (NORVIT and WENBIT) were expected to report follow-up cancer outcomes in 2009. Since Norway does not allow foods to be fortified with folic acid, background exposure to folic acid would have been very low in these trials. Prolonged follow-up of participants in such trials after the cessation of folic acid supplementation may provide useful information on possible long-term effects of folic acid on cancer risk.

56. The general consensus was that the findings from the B-vitamin treatment trials did not support or refute the suggestion that high folic acid intakes increase colorectal cancer. The levels of folic acid intake associated with potential risk are considered in paragraphs 58-60.

Population groups and cancer risk

57. Population groups potentially at greater risk of developing colorectal cancer with folic acid supplementation may include individuals with cancer, undetected cancer, or premalignant colorectal adenomas. Older people, who are at increased risk of developing colorectal adenomas may also be at increased risk.

58. The effects of folic acid on treatment efficacy of commonly used chemotherapeutic drugs (such as methotrexate and 5-fluorouracil) have been insufficiently studied.

Intake levels and cancer risk

59. The difficulty of assessing a threshold for a possible carcinogenic effect of folic acid, based on interpretation of the cancer studies in humans, was recognised.

60. The possibility of using the amount of folic acid that would cause the appearance of free folic acid in the circulation as a threshold for intake was discussed. However, it was noted that there was insufficient evidence to assess possible risks associated with unmetabolised folic acid in the circulation. Since folate metabolism is under polygenetic control it would be difficult to factor genetic considerations into any reconsideration of the UL.

61. It was agreed that people should not consume more than the current UL of 1 mg/day of folic acid. Although the UL is based on limited supporting evidence, it could be used as a general guidance value in order to prevent potential adverse effects of excess intakes of folic acid. It was not possible to identify whether there was a dose response relationship or a threshold for the effects of folic acid on potential colorectal cancer risk.

62. It is also important to distinguish between different sources of folate, i.e. natural food folates and folic acid from fortified foods and from supplements. Data from the USA (NHANES) have shown that the population group of ≥ 60 years of age had the highest folic acid intakes with the largest amounts deriving from supplements.

In this population group, which is at highest risk for colorectal cancer, lower dosage mandatory fortification was not likely to have influenced serum folate levels.

3.2 Discussion Group 2: Folic acid and other cancers (breast, prostate, pancreatic, oesophageal)

Consideration of the evidence

63. Data from animal studies regarding the relationship between folic acid and breast cancer are limited.

64. Time trend data from the USA do not show temporal changes in the incidence of breast and prostate cancer following voluntary and mandatory fortification of enriched grain products with folic acid (1996-1998). In Canada, there was a significant increase in the incidence of prostate cancer after 1996 (voluntary fortification was introduced in December 1996).

65. A prospective cohort study has suggested a potential harmful effect of folic acid intake (≥ 400µg/d) on breast cancer risk (Stolzenberg-Solomon et al., 2006) (see paragraph 27). The WCRF report concluded that the epidemiological data for an association between folate and breast cancer was too inconsistent or limited to allow conclusions to be reached and that there was limited evidence that foods containing folate protect against pancreatic and oesophageal cancer.

66. It was noted that the existing evidence is inadequate to make a judgement on the possible association between folic acid and breast cancer risk and that breast cancer is a multifactorial and complex disease which makes assessment of any folic acid-cancer association very difficult. It was agreed that issues that required further consideration included:

- Interactions between folate and alcohol intake
- Age at menarche and menopause
- Form of folate (natural vs folic acid)
- Interaction of folate with other nutrients
- Dose
- Other risk factors
- Genetic background.

Folic acid food fortification

67. A range of foods are voluntarily fortified[1] with folic acid at variable levels. This makes it difficult for individuals and risk managers to assess the actual intakes of folic acid. Modelling work undertaken in the UK (SACN, 2006) suggests that mandatory folic acid fortification of flour together with restriction of folic acid from all voluntary sources would result in a more even distribution of folic acid intakes across the population.

Population groups and cancer risk

68. Population groups that might be vulnerable to folic acid supplementation were not discussed as food fortification would have an impact on the whole population.

Intake levels and cancer risk

69. It was agreed that it was not possible to determine whether there was a dose-response or threshold level associated with possible risk of breast, pancreatic or oesophageal cancer. However, the consensus was that intakes should not exceed the UL.

[1] Voluntary folic acid food fortification is regulated under the provisions of Regulation (EC) No. 1925/2006 on the addition of vitamins and minerals and certain other substances to foods.

4.0 PLENARY DISCUSSION AND CONCLUSIONS

4.1 Final comments and conclusions

70. Divergent views were expressed during the discussion and there was disagreement between experts regarding the interpretation of the trial evidence and the UL of 1mg/d. Some considered that the available evidence did not support an association of high intakes of folic acid with possible cancer risk or the UL of 1 mg/day which is based on limited data. The following general conclusions reflect the consensus of participants.

71. The beneficial effect of folic acid in reducing the risk of NTDs is well established. Women who might become pregnant are the target population for this benefit. Others with low folate intakes would also benefit from folic acid fortification. Suggestions for additional benefits, including reductions in CVD, cancer occurrence, and cognitive decline, have also been made; evidence for these benefits is not supported by randomised controlled trials.

72. Evidence from animal studies, trend data for colorectal cancer incidence, and a randomised controlled trial have raised concerns of a possible association between high intakes of folic acid and promotion of cancer development and progression. While the totality of the randomised trial evidence from the CVD trials does not suggest that folic acid intakes are associated with increased cancer risk, these trials probably did not have sufficient power to detect overall cancer risk or site-specific cancer risk and their duration of follow-up may have been too short to detect cancer risk.

73. There are currently insufficient data to allow a full quantitative risk assessment of folic acid and cancer or to determine whether there is a dose-response relationship or a threshold level of folic acid intake associated with potential colorectal cancer risk

74. The current evidence does not show an association between high folic acid intakes and cancer risk but neither do they confidently exclude a risk. The uncertainties in relation to cancer risk highlight the importance of ensuring monitoring systems are set up for assessment of folic acid intake and status and NTD and cancer incidence in countries that decide to introduce mandatory fortification.

75. Targeted generation of additional data and knowledge, both epidemiological and animal/mechanistic, might be important in informing the risk/benefit assessment of folic acid in the future.

76. Intakes of folic acid should not exceed the established UL of 1mg/day (SCF, 2000). However, the UL is based on limited data and may need to be revised when further data become available.

77. Setting maximum safe levels for the amount of folic acid that can be added to foods voluntarily fortified with folic acid and supplements will be important in ensuring that consumption of foods fortified with folic acid and folic acid supplements does not lead to intakes above the UL.

4.2 Further research

78. Further research in the following areas may be helpful in informing future risk assessments on the possible association between high intakes of folic acid and cancer risk:

79. Continued long-term follow-up (5-10+ years) for cancer risk in participants in folic acid supplementation trials after the cessation of the trials.

80. An update of the B-Vitamin Treatment Trialists' meta-analysis to assess the effects on risk of any cancer and on site-specific cancers after completion of the 3 ongoing B-vitamin trials that are due to report in the next 18-24 months.

81. Future studies need to take better account of total folate and total folic acid exposure (natural food folate and folic acid from voluntary and mandatory fortified foods and supplements) and folate status (measured by best/recommended assays, including measurement of different folate forms and unmetabolised folic acid).

82. Further experimental studies on the pharmacokinetics of folic acid in animals and humans (including folate metabolism in adenomas).

83. Modelling of population effects of food folates and folic acid intakes from voluntary and mandatory fortification and from supplements.

84. Animal studies on the effect of folic acid supplementation on precancerous-resected lesions.

85. In vitro and in vivo studies on proliferation effects.

86. Monitoring possible effects of unmetabolised folic acid on health outcomes.

5.0 REFERENCES

1. Charles D., Ness A.R., Campbell D., Davey-Smith G. & Hall M.H., 2004. Taking folate in pregnancy and risk of maternal breast cancer. *BMJ.*, 329:1375-1376.

2. Cole B.F., Baron J.A., Sandler R.S., Haile R.W., Ahmen D.J., Bresalier R.S., McKeown-Eyssen G., Summers R.W., Rothstein R.I., Burke C.A., Snover D.C., Church T.R., Allen J.I., Robertson D.J., Beck G.J., Bond J.H., Byers T., Mandel J.S., Mott L.A., Pearson L.H., Barry E.L., Rees J.R., Marcon N., Saibil F., Ueland P.M., Greenberg E.R., and Polyp Prevention Study Group, 2007. Folic acid for the prevention of colorectal adenomas: a randomised clinical trial. *JAMA*, 297(21):2351-2359.

3. Commission Regulation (EC) No 1925/2006 of the European Parliament and of the Council of 20 December 2006 on the addition of vitamins and minerals and of certain other substances to foods, OJ L 404 of 30 December 2006. <http://eurlex.europa.eu/smartapi/cgi/sga_doc?smartapi!celexplus!prod!DocNumber&lg=en&type_doc=Regulation&an_doc=2006&nu_doc=1925>

4. De Wals P, Tairou F, Van Allen MI, et al. Reduction in neural-tube defects after folic acid fortification in Canada. *N Engl J Med*. 2007;357(2):135-42.

5. Directive 2002/46/EC of the European Parliament and of the Council of 10 June 2002 on the approximation of the laws of the Member States relating to food supplements. OJ L 183, 12.7.2002, p. 51–57. <http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=CELEX:32002L0046:EN:NOT>

6. Durga J, van Boxtel MP, Schouten EG, Kok FJ, Jolles J, Katan, MB, Verhoef P. Efect of 3-year folic acid supplementation on cognitive function in older adults in the FACIT trial: a randomised, double-blind, controlled trial. *Lancet*. 2007;369:208-16.

7. EFSA (European Food Safety Authority), 2008. ESCO working group on the Analysis of Risks and Benefits of Fortification of Food with Folic Acid. <http://www.efsa.europa.eu/EFSA/National_Focal_Points/Scientific_Cooperation_projects/efsa_locale-1178620753812_folic_acid.htm>

8. EFSA (European Food Safety Authority), 2006. Tolerable upper intake levels for vitamins and minerals. European Publications Office. <http://www.efsa.europa.eu/cs/BlobServer/Scientific_Document/upper_level_opinions_full-part33.pdf?ssbinary=true>

9. European Commission, Health and Consumer Protection Directorate General, 2006. Discussion Paper on the setting of maximum and minimum amounts for vitamins and minerals in foodstuffs.

<http://ec.europa.eu/food/food/labellingnutrition/supplements/discus_paper_amount_vitamins.pdf>

10. Huang Y., Han S., Li Y., Mao Y. & Xie Y., 2007. Different roles of MTHFR C677T and A1298C polymorphisms in colorectal adenoma and colorectal cancer: a meta-analysis. *J Human Genet.*, 52, 73–85.
11. Hubner R.A. and Houlston R.S., 2006. MTHFR C677T and colorectal cancer risk: a meta-analysis of 25 populations. *Int J Cancer;* 120:1027–1035.
12. Jaszewski R., Ullah N., Misra S. et al., 2008. High dose folic acid supplementation inhibits recurrence of colorectal adenomas. *Gastroenterology* , 128:631 (A-93).
13. Kelly P, McPartlin J, Goggins M, Weir DG, and Scott JM., 1997. Unmetabolized folic acid in serum: acute studies in subjects consuming fortified food and supplements. *Am J Clin Nutr.*, 65:1790-1795.
14. Kim Y.I., 2003. Role of folate in colon cancer development and progression. *J Nutr*., 133 Suppl. 3731S-3739S.
15. Kim Y.I., 2004. Will mandatory folic acid fortification prevent or promote cancer? *Am J Clin Nutr*., 80:1123-1128.
16. Logan R.F., Grainge M.J., Shepherd V.C., Armitage N.C., Muir K.R., 2008. Aspirin and folic acid for the prevention of recurrent colorectal adenomas. *Gastroenterology*, 134(1):29-38.
17. Mason J.B., Dickstein A., Jacques P.F., Haggarty P., Selhub J., Dallal G. Rosenberg I.H., 2007. A temporal association between folic acid fortification and an increase in colorectal cancer rates may be illuminating important biological principles: a hypothesis. *Cancer Epidemiol Biomarkers Prev*., 16:1325-29.
18. MRC Vitamin Study Research Group, 1991. Prevention of neural tube defects: results of the Medical Research Council Vitamin Study. *Lancet.* 1991; 338: 131-137 137.
19. Paspatis G.A. and Karamanolis D.G., 1994. Folate supplementation and adenomatous polyps. *Dis Colon Rectum.*, 37:1340-1341.
20. SCF (Scientific Committee on Food), 2000. Opinion of the Scientific Committee on Food on the Tolerable Upper Intake Level of Folate. European Commission, SCF/CS/NUT/UPPLEV 18 final, 28 November 2000, Brussels. <http://ec.europa.eu/food/fs/sc/scf/out80e_en.pdf>
21. SCF (Scientific Committee on Food), 2000. Guidelines of the Scientific Committee on Food for the development of tolerable upper intake levels for vitamins and minerals (adopted on 19 October 2000). SCF/CS/NUT/UPPLEV/11 Final 28 November 2000. <http://ec.europa.eu/food/fs/sc/scf/out80a_en.pdf>

22. Scientific Advisory Committee on Nutrition. *Folate and Disease Prevention.* London: TSO, 2006.

23. Sharp L and Little J. Polymorphisms in genes involved in folate metabolism and colorectal neoplasia: a HuGE review. *Am J Epidemiol*. 2004; 159:423-443.

24. Stolzenberg-Solomon R.Z., Chang S.C., Leitzman M.F., Johnson K.A., Johnson C., Buys S.S., Hoover R.N., Ziegler R.G., 2006. Folate intake, alcohol use, and postmenopausal breast cancer risk in the Prostate, Lung, Colorectal, and Ovarian Cancer Screening Trial. *Am J Clin Nutr.*, 83:895-904.

25. WCRF/AICR (World Cancer Research Fund/American Institute for Cancer Research Publisher), 2007. Food, Nutrition, Physical Activity, and the Prevention of Cancer: a Global Perspective. Washington DC: AICR.

26. Williams LJ, Rasmussen SA, Flores A, Kirby RS, Edmonds LD. Decline in the prevalence of spina bifida and anencephaly by race/ethnicity. *Pediatrics*. 2005;116(3):580-6.

27. Yeung L., Yang Q., Berry R.J., 2008. Contributions of total daily intake of folic acid to serum folae concentrations. *JAMA*, 300 (21):2486-2487.