ESCO REPORT

Prepared by the EFSA Scientific Cooperation Working Group on Analysis of Risks and Benefits of Fortification of Food with Folic Acid[1]

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ESCO WORKING GROUP MEMBERS[2]

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SUMMARY

In 2008, the European Food Safety Authority (EFSA) established an EFSA Scientific Cooperation Working Group (ESCO WG) on "Analysis of risks and benefits of fortification of food with folic acid". The Working Group was asked to: i) review current practice in Member States regarding the level of voluntary fortification of foods with folic acid, and ii) consider new evidence regarding the risk of high intakes of folic acid and the need to review current guidance on safe upper levels of folic acid for all population groups.

This report provides the results and conclusions of the activities of the ESCO WG to help inform whether an opinion on risk assessment of folic acid and cancer is needed.

Data were provided by Working Group members for each of the respective countries. The main issues considered were: national congenital anomalies registries, prevalence of neural tube defects (NTDs), recommended dietary intakes of folate, practices of voluntary and mandatory folic acid food fortification within European countries. The report also explores the relationship between folic acid and cancer risk.

National representative data on the prevalence of NTDs are not available for all European countries. Data from EUROCAT shows that the prevalence of NTDs varies widely between countries. Although all countries recommend women who might become pregnant to supplement their diet with 400µg/day of folic acid, the efficacy of this advice is not evaluated in all countries.

A range of foods are voluntarily fortified with folic acid at variable levels in most European countries. Mandatory fortification has not been introduced in any European country.

The health benefits of folic acid in relation to the reduction in risk of NTDs are well established through human intervention studies. Epidemiological studies have often reported diets high in folate are associated with a number of other health benefits such as reducing the risk of cardiovascular disease (CVD). Intervention studies using folic acid have produced a range of different results including adverse effects; overall they do not support the hypothesis that folic acid supplementation of human populations reduces the chronic disease risk.

EFSA convened a meeting of experts to consider the evidence on the possible relationship regarding folic acid and risk of cancer. The meeting identified key knowledge gaps and made a number of research recommendations. It considered the results of randomised control trials (RCTs) designed to test the effect of folic acid on recurrence of colorectal adenomas. These studies produced different results: four studies with 3 year interventions reported no adverse effects, where as one longer term study reported adverse effects on adenomas in the intervention group. The meeting also considered evidence on cancer occurrence collected from participants in a consortium of RCTs designed to test the hypothesis that folic acid and other B vitamins would reduce CVD risk. The totality of

evidence from the meta-analysis of these CVD trials does not suggest that folic acid is associated with increased cancer risk. However, the meta-analysis probably did not have sufficient power to detect over all or site specific cancer risk.

The research recommendations at the meeting included the need for further animal and human studies and for continued long term follow up of cancer risk in participants in supplementation trials. It was recommended that such studies should take better account of folate and folic acid exposure.

The working group concluded that there was currently insufficient data to allow a full quantitative risk assessment of folic acid and cancer.

KEY WORDS: folic acid, folate, neural tube defects, cancer risk, food fortification, ESCO.

DISCLAIMER

The conclusions and recommendations of this report reflect those of the national experts involved in the ESCO Working Group on Analysis of Risks and Benefits of Fortification of Food with Folic Acid and not necessarily represent the views of EFSA.

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BACKGROUND

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 naturally found in a variety of foods including green leafy vegetables, fruit, liver, and yeast. Folic acid is the synthetic form of folate which is widely used in supplements and food fortification. It is more stable in foods and is better absorbed than natural folates. Evidence from scientific studies has conclusively shown that folic acid supplementation can prevent the occurrence of neural tube defects (NTDs). This has lead many countries to recommend women who might become pregnant to supplement their diet with folic acid. Public health campaigns by countries to promote the awareness of this message and promote supplement intake have been unsuccessful in most countries,; many women are still not taking folic acid supplements prior to and for the first weeks after conception (EUROCAT, 2005). Some countries such as the US and Canada have introduced mandatory folic acid fortification of a staple food such as flour, as a strategy to reduce the prevalence of NTDs. This has 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.

The prevalence of NTDs varies across the European Union (EU). The rate is currently reported to range from 0.4 to 2.0 per 1,000 births. This difference in reported rates is probably at least in part due to differences in reporting and collating data on NTDs in the different European countries.

Voluntary fortification of food with folic acid is permitted in most European countries, with considerable variation on the levels of folic acid added to food. Currently no European country has implemented mandatory fortification, although some have considered it as a strategy to reduce the incidence of NTDs. Mandatory fortification is currently under review in the United Kingdom and has been rejected in Sweden. These reviews are taking place at least in part because of recent data suggesting an association between high intakes of folic acid and cancer risk. Mandatory fortification was recommended in Ireland in 2006 but is now on hold as there is currently sufficient folic acid in the diet from voluntary fortification sources.

In 2008, the European Food Safety Authority (EFSA) established the EFSA Scientific Cooperation Working Group (ESCO WG) on Analysis of Risks and Benefits of Fortification of Food with Folic Acid to share the experience and knowledge of folic acid fortification of foods across the EU and to evaluate possible risks in terms of the need to review the current guidance on tolerable upper level (UL) of folic acid for all population groups.

TERMS OF REFERENCE

The ESCO Working Group on Analysis of Risks and Benefits of Fortification of Food with Folic Acid was established in 2008 with the following terms of reference:

1. 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;
2. 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. To provide regular feedback to the Steering Group on Cooperation, the Advisory Forum and the Scientific Committee on progress made during the execution of the project.

Timeline:

It was anticipated that the timeframe for the completion of the terms of reference would be 1-2 years.

Milestones and deliverables:

The following milestones and deliverables were agreed:

1. Report to the Executive Director of EFSA.
2. Scientific meeting of experts to review data on potential cancer risks associated with folic acid.
3. Possible recommendation to the Executive Director of EFSA for an opinion on risk assessment on folic acid and cancer.

ACKNOWLEDGEMENTS

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WORKING METHODOLOGY

The ESCO Working Group undertook the following tasks:

To collect, compile and analyse information on the current situation and practices in selected European countries[3] on rates of NTD-affected pregnancies, dietary intakes of folate/folic acid and national dietary recommendations of folic acid. Information and data were compiled using scientific literature, country statistics on NTDs rates, if available, and national survey data on nutrient intakes. Details and results of this analysis are given in Appendix 1.

To assess the possible risks 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. A scientific meeting on *"Folic Acid: An Update on Scientific Developments"*, was organised on 21-22 January 2009, between international scientists to assess the latest scientific evidence regarding folic acid and risk of cancer. The meeting report can be found in Appendix 2.

[3] The collection of data was carried out by Working Group members, for the following countries: Belgium, Denmark, France, Germany, Hungary, Ireland, Italy, the Netherlands, Norway, Romania, Sweden, Switzerland, the United Kingdom. The term 'European countries' throughout this report refers to those countries listed above.

BENEFITS AND RISKS OF FOLATE

There is a large body of published work in the scientific literature on the potential benefits and risks of dietary folate and folic acid.

Evidence from randomised controlled trials (RCTs) has conclusively demonstrated that folic acid supplementation can reduce the risk of pregnancies with NTDs. In addition, findings from epidemiological studies have suggested that high intakes of folate can be associated with a lower risk of cardiovascular disease (CVD), some cancers and age related cognitive decline; although generally the results of RCTs with folic acid have not been demonstrated such positive benefits.

Against these benefits, high intakes of folic acid have been associated with potential risks of adverse effects. High doses of folic acid may delay the diagnosis of vitamin B12 deficiency by treating ("masking") the anaemia of vitamin B12 deficiency which can lead to irreversible neurological damage. The Scientific Committee on Food (SCF) set a tolerable upper intake level (UL) of folic acid at 1mg per day, based on the risk of progression of the neurological symptoms caused by the masking of haematological signs of vitamin B12 deficiency (SCF, 2000). This UL level is applicable to the whole population (also relevant for patients suffering from pernicious anaemia), adjusted downwards for children (1-17 years of age) on the basis of body weight. However, current medical practice does not rely on the presence of anaemia for the detection of vitamin B12 deficiency, which frequently presents without anaemia. New scientific evidence has emerged over the past three years suggesting a possible link between high intake levels of folic acid (levels near or exceeding the UL) and risks of various types of cancer, particularly colorectal cancer. Evidence from animal studies suggests that high folic acid intakes may have a dual modulatory effect on cancer risk: inhibiting the formation of neoplastic lesions in normal tissues but accelerating the malignant transformation of existing neoplasms. Other postulated adverse effects of folic acid intake include acceleration of cognitive decline with age and reduction of the efficacy of antifolate drugs such as methotrexate used in chemotherapy for cancer treatment and drugs used to treat epilepsy, but these research questions have been insufficiently studied.

CURRENT PRACTICES IN THE EU

Recommended intakes

In the EU, the recommended intake for folate is set by SCF. The EU population reference intake is 200µg folate/day for adults, 400µg folate/day for pregnancy and 350µg folate per day for lactation (SCF, 1993). Recommendations for children are extrapolated from the recommended intakes for adults on the basis of body weight and range from 200µg folate/day for 1-3 years to 800µg for folate /day for 15-17 years.

Recommended national daily intakes for population groups vary across the thirteen countries that were part of the ESCO Working Group. For adults the recommended daily intake ranges from 200 to 400µg folate[4]. For pregnancy, it ranges from 300 to 600µg folate/day and for lactation from 260 to 600µg folate/day. The reason for this range depends on the biological outcome used as a basis of the recommendation. Lower values tend to be based on maintenance of folate concentration in the liver, whereas higher values for adults tend to be based on maintaining/reducing homocysteine levels in plasma (and the hypothesis that homocysteine is inversely associated with cardiovascular risk). The recommendations of folate intakes of European countries are provided in Appendix 1, Table 3.

Many EU countries have established food consumption databases from national surveys. These show mean/median dietary folate intake ranges from 151 to 345µg/day for adult men and 122 to 339µg/day for women. Folate intake data can be found in Appendix 1, Tables 6 and 7.

Compared to the national recommendations, mean/median folate intakes of women were reported to be below the national recommendations in all countries except Belgium and UK (for details see Appendix 1; Table 7). For adult men, the folate intakes were below the national recommendations in France (except men aged 45-60 years), Germany, Hungary, the Netherlands and Sweden.

When comparing intakes across European countries it should be noted that there may be differences in methods of collecting and analysing consumption data. Also, the age groups included, and the quality of nutrient databases vary. In addition, it is useful to bear in mind that recommended daily intakes do differ across the European countries. Therefore caution should be applied in the interpretation of these values and making comparisons across countries.

[4] In some countries folate dietary recommendations are expressed as dietary folate equivalents (DFE).

Folate status

Data on folate status for population groups in the different countries were obtained from scientific studies on biomarkers and where available, national surveys (see Appendix 1, Table 8 for details). Suitable biomarkers for folate status include concentrations of red blood cell folate, plasma or serum folate. Some countries also use plasma homocysteine as a marker of folate deficiency.

Mean red blood cell concentrations, an indication of tissue stores of folate were not available for all countries. Values above 340nmol/l indicate sufficient stores, however, due to the limited data it is difficult to assess or make comparisons of the folate status across the countries. Data available from selected countries indicate that folate status based on serum, plasma and/or red blood cells concentration is adequate.

In addition, a review by Dhonukshe-Rutten et al. (2009) found that plasma or serum folate status in Europe ranged from 6.3 to 20.1nmol/l (based on data from 1990 to 2005). No obvious differences existed in status indicators between men and women. Plasma folatelevels lower than 10nmol/l was defined as being 'low', between 10 and 15nmol/l as 'moderate' and above 15nmol/l as 'favourable'. Folate intake was only weakly correlated with plasma folate levels.

Recommendations for pregnant women

In all countries, women of childbearing age who might become pregnant are advised to supplement their diet with 400µg of folic acid daily. However the timing of the advice and the duration differ across European countries. The duration of the recommended supplement intake varies across countries and ranges from 'as soon as contraception is stopped' to 'four weeks before conception' and from eight weeks after conception' to 12th week of pregnancy, 'throughout pregnancy and lactation'. It is most commonly advised to start taking the supplement four weeks prior to conception until the twelfth week after conception. Data were not available on compliance with recommendations in all countries, but in those reporting compliance, it varied from about 10 to 80%. In general, evidence from studies which have evaluated the efficacy of policies of recommending women to use folic acid supplements with the aim of reducing NTD-affected pregnancies have shown no or very limited effects in most European countries (Dolk, 2005).

The systems for monitoring NTDs within and across European countries are not harmonised. National congenital anomaly registers are only available in some countries. The best estimates of the prevalence of NTDs are available from EUROCAT, a Europe-wide network of population-based registries for the epidemiological surveillance of congenital anomalies. As part of this network more than 1.5 million births are surveyed annually and data come from 43 registries in 20 countries covering about 28% of European births. The prevalence of NTDs varies widely among the European countries,

estimated to be between 4.13 and 19.70 per 10,000 births. This variation is mainly due to the different reporting systems for collating data on congenital anomalies and differences in policies on pregnancy termination and antenatal screening across European countries. It is likely that actual figures are much higher. The number of NTD-affected live births in Europe has decreased considerably over the last 25 years due to preterm diagnostics, and as a consequence, higher numbers of induced abortions after diagnosed NTDs (Dolk, 2005).

Voluntary food fortification

Voluntary fortification is widely practiced in the EU under the regulation 1925/2006/EC on the addition of vitamins and minerals and of certain other substances to foods. This regulation allows the fortification of all foods except unprocessed foods and alcoholic beverages. Foods voluntarily fortified with folic acid are widely available on the European market, except in Sweden where voluntary fortification is not practiced, and in Denmark and Norway where approval is required. A range of product categories that are fortified on a voluntary basis includes dairy products, breakfast cereals, cereal bars, fruit juices, fat spreads, bread and beverages. Maximum levels for the addition of folic acid to foods are not yet set in European regulations. Data gathered as part of this project showed that the levels vary widely, with the highest levels added to spreads (up to 1000µg/100g).

As part of the implementation of the regulation 1925/2006/EC, work is currently ongoing on setting 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). The contribution of voluntarily fortified foods to the reduction of NTDs is difficult to assess as variable levels of folic acid are added to a range of folic acid fortified foods. However, recent data from Ireland suggests that as a result of voluntary fortification there has been an increase of about 30% in dietary folic acid intakes among women over the past 2-3 years, and the reduction in NTD prevalence in Ireland in recent years is likely due, in part, to the increased voluntary fortification of foods with folic. Evidence from the UK however indicates that some people exceed the UL for folic acid due to voluntary food fortification alone (others exceed the UL due to supplements alone and some due to a combination of fortificants and supplements).

Mandatory food fortification

Mandatory fortification with folic acid has been introduced in some 50 countries worldwide, as a strategy to help women increase their intake of folate. Reports from the US and Canada have shown an effective decline in NTDs (CDC et al., 2002; De Wals et al., 2007; Williams et al., 2005). Heseker et al. (2008) reported that countries with

mandatory folic acid fortification achieved a significant decrease in the prevalence of NTD. The prevalence of NTD at birth declined to approximately five cases per 10,000 births and 7-8 cases at birth or abortion per 10,000. This decline was independent of the amount of folic acid administered and reveals a 'floor effect' for folic acid-dependable NTDs. Thus, not all cases of NTDs are preventable by increasing folate intake (Heseker et al., 2008).

Mandatory fortification of folic acid is not implemented in any of the countries involved in this ESCO Working Group nor in any other EU country. Although it is under serious consideration in Ireland and United Kingdom, it has yet to be introduced as a public health policy.

The main issue for consideration when deciding whether to introduce mandatory food fortification with folic acid is the risk-benefit ratio concerning the possible increased risk with regards to an excessive intake of folic acid and the possible increased benefit with regards to reducing the prevalence of NTDs in a population.

FOLIC ACID AND CANCER RISK

High intakes of folic acid, the synthetic form of folate, have been associated with adverse effects. Some data suggest the possibility that high folic acid intakes may be associated with increased risks of cancer. 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.

Some 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 (thereby protecting against cancer) but increase the progression of established neoplasms (thereby increasing risk of cancer). The relationship between folic acid and colorectal cancer (CRC) seen in animal studies is broadly consistent with biochemical evidence on folate metabolism. 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 food fortification.

Human epidemiological data are inconclusive. A review by the World Cancer Research Fund (WCRF/AICR, 2007) concluded that there is limited evidence of a protective effect of folate (based on papers published before 2006). The report noted, however, uncertainty because of potential confounding, effect modification (particularly from B vitamins), and the quality of dietary exposure assessment (distinction between intake levels of natural folate *vs* synthetic folic acid etc.). Furthermore, it is important to note that findings from epidemiological studies come from observations that could be confounded by other dietary or non dietary factors associated with cancer risk. Therefore, it is not possible to reach conclusions about folic acid intakes and potential cancer risk from observation studies.

Time trend data from USA and Canada show an increase of colorectal cancer incidence around the same time as the introduction of mandatory folic acid fortification (Mason et al., 2007). However, there are a number of points of uncertainty that limit interpretation of the Mason study. These include: i) uncertainty regarding the precise timing of the increase in the population exposure to folic acid in relation to upturn in CRC incidence, ii) 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), iii) epidemiological associations between folic acid and cancer risk may differ due to pre-existing supplement use or voluntary fortification status in the studied populations, and iv) there were improvements in screening practice for colorectal cancer around the same time in question, which may have accounted for some changes in colorectal cancer incidence. Sudden increase 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.

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.

Four randomised controlled trials (RCTs) have assessed the effect of folic acid supplementation on the risk of recurrence of colorectal adenomas (Paspatis and Karmanolis, 1994; Cole et al., 2007; Jaszewski et al., 2008; Logan et al., 2008) and one unpublished US trial (E. Giovannucci, personal communication, 2009). The results from two small trials (n=60, Paspatis and Karamanolis, 1994 and n=93, Jaszewski et al., 2008, in which 1 and 5 mg folic acid were supplemented for 2 and 3 years, respectively) suggested that folic acid supplementation reduces adenoma risk; these trials were however likely to have been underpowered to assess the effect of folic acid on cancer risk. The trial by Logan et al. (2008) reported that folic acid supplementation (0.5mg/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. However, the available data comes from 3 years follow up.

The trial by Cole et al. (2007) reported that folic acid supplementation in a dose of 1mg per day 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, after 6 years (carried out 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). In addition to taking the assigned intervention dose of 1mg/d, subjects in this study were also exposed to mandatory folic acid food fortification in the US which is estimated to have provided at the time an additional 200µg folic acid per day. Therefore the findings of this trial suggested that folic acid at doses in excess of 1mg/day may increase the risk of developing multiple/advanced adenomas after a few years delay and consequently increase the risk of cancer.

A number of trials have investigated B-vitamins supplementation (including folic acid) for prevention of cardiovascular disease (CVD) in people with a prior history of CVD or renal disease. These trials also collected data on the effect of folic acid on cancer

outcomes. 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: 8 trials, n=37,485; R. Clarke, personal communication, 2009). Results from the meta-analysis found no significant beneficial or adverse effect of B vitamins on vascular events, all cause mortality, cancer, or cancer in any of the pre-specified sub groups or at any specific sites. The data presented were based on trials with folic acid doses ranging from 0.8–40mg/d for a median duration of 5 years. Interpretation of these results is limited by the short duration of follow up in comparison to the longer periods of time over which cancer usually develops and the limited power of the meta-analysis particularly for the site specific cancers (e.g. colorectal, breast and prostate). It is unlikely that adequately powered randomised controlled trials for site-specific cancer would be possible because of the very large number of people required.

The existing evidence base on breast cancer is inadequate to make a judgement on the possible association between folic acid and breast cancer risk. Breast cancer is a multifactorial and complex disease which makes assessment of any folic acid cancer association very difficult.

Scientific data published up until January 2009 and data presented at the scientific meeting on *"Folic Acid: An Update on Scientific Developments"* (Jan. 09) has been considered by the ESCO Working Group. The ESCO Working Group note that evidence base on folic acid and cancer continues to develop.

CONCLUSIONS AND RECOMMENDATIONS

- 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. Other population groups with low folate intakes (groups who have intakes below their requirement) would also benefit from folic acid. Although suggestions for additional benefits, including reductions in CVD, cancer occurrence, and cognitive decline, have been made, evidence for these benefits is not supported by randomised controlled trials.
- National recommendations/reference values for folate intake vary across the European countries ranging from 200µg to 400µg folate per day for adults. All countries also recommend women of childbearing age/planning a pregnancy to supplement their diet with an additional 400µg of folic acid per day.
- The available data on folate intake and status varies across European countries. National representative data is not available for all countries. Variation in the methods used for collecting and analysing data make comparisons between different population groups and countries difficult. Therefore, harmonisation of methodologies for data collection and reporting, e.g. on food consumption and assessment of folate status is important.
- There is wide variation in the prevalence of NTDs in European countries. It is likely that the number of NTDs is higher than reported as the data on NTDs compiled through EUROCAT does not provide national coverage for all European countries. Furthermore, there are differences in monitoring and reporting of NTDs. Few countries have national systems in place to collate data on all NTD-affected pregnancies, including those that are terminated. Efforts should be made to improve NTD registers so that reliable data is available on the incidence of all affected pregnancies.
- The effectiveness of advice to women planning a pregnancy on increasing folic acid intake has not been evaluated in all European countries. Where the policies have been evaluated, data shows that most women do not follow the advice in the critical very early stages of pregnancy (when the neural tube is developing) and there is limited/no impact on NTD incidence.
- Voluntary fortification of foods with folic acid is very common in Europe with wide variation in levels of folic acid added to various foodstuffs. Mandatory food

fortification is not practiced in the countries involved in this ESCO Working Group nor in any other EU country.

- Evidence from animal studies suggests a possible association between high intakes of folic acid and promotion of cancer development and progression. There is also a time trend study from the USA and Canada that suggests colorectal cancer incidence increased at around the same time mandatory fortification with folic acid was introduced. Interpretation of this evidence is limited for a number of reasons.
- Evidence on folic acid and cancer risk is also available from a number of randomised control trials. Some of these trials were specifically designed to test the effect of folic acid on recurrence of colorectal adenomas. These have produced different results: four studies with three year interventions reported no adverse effects where as one longer term study reported adverse effects on adenomas in the intervention group. Results on cancer risk have also been brought together in a meta-analysis from other randomised control trials designed to test the hypothesis that folic acid and other B vitamins would reduce CVD risk. The totality of evidence from these trials does not suggest that folic acid intakes is associated with increased cancer risk, however interpretation of these data is limited by a number of issues including duration of the trials and power of the meta-analysis.
- Other postulated adverse effects of folic acid, such as accelerating cognitive decline with age or reducing the efficacy of antifolate drugs, have been insufficiently studied.
- Present data do not allow determination of whether there is a dose-response relationship or a threshold level of folic acid intake associated with potential colorectal cancer risk. 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 may be useful; however, it should be noted that there is 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. The difficulty of assessing a threshold for a possible carcinogenic effect of folic acid, based on interpretation of the cancer studies in humans, is also recognised.
- Intakes of folic acid should not exceed the established upper intake level (UL) of 1mg/day (SCF, 2000). The UL is based on limited data and may need to be revised when further data become available.

- The uncertainties in relation to cancer risk highlight the importance of ensuring monitoring systems are set up for assessment of folic acid intake (from supplements and fortified foods), folate status and cancer incidence. In addition it is important to have reliable monitoring systems for NTDs. It is also important to distinguish between different sources of folic acid, i.e. from fortified foods and from supplements as the pharmokinetics may vary depending on the form and dosage (single, multiple) of application.
- Setting maximum safe levels for the amount of folic acid that can be added to foods voluntarily fortified 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 for any population subgroup, including young children.
- There is currently insufficient data to allow a full quantitative risk assessment of folic acid and cancer risk. Scientific developments within this area should be closely monitored.
- The 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.

REFERENCES

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>

EUROCAT (European Surveillance of Congenital Anomalies).

<http://www.eurocat.ulster.ac.uk>

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>

FSAI (Food Safety Authority of Ireland), 2008. Report of the Implementation Group on Folic Acid Food Fortification to the Department of Health and Children. <www.fsai.ie/publications/reports/folic_acid_implementation.pdf>

SCF (Scientific Committee for Food), 1993. Nutrient and energy intakes for the European Community. Reports of the Scientific Committee for Food, Thirty First Series. European Commission, Luxembourg.

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>

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

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.

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.

Dhonukshe-Rutten, R.A.M., de Vries, J.H.M., de Bree, A., van der, Put N., van Staveren, W.A. and de Groot, L.C.P.G.M., 2009. Dietary intake and status of folate and vitamin

B12 and their association with homocysteine and cardiovascular disease in European populations. *Eur J Clin Nutr*. 63:18-30.

Dolk H., 2005. EUROCAT: 25 years of European surveillance of congenital anomalies. *Arch Dis Child Fetal Neonatal Ed*. 90:F355–F358. doi: 10.1136/adc.2004.062810.

Heseker, H.B., Mason, J.B., Selhub, J., Rosenberg, I.H. and Jacques, P.F., 2008. Not all cases of neural-tube defect can be prevented by increasing the intake of folic acid. *Br J Nutr*.; 16:1-8.

Jaszewski R., Ullah N., Misra S. et al., 2008. High dose folic acid supplementation inhibits recurrence of colorectal adenomas. *Gastroenterology* , 128:631 (A-93).

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.

Mason, J.B., Dickstein, A., Jacques, P.F., Haggarty, P., Selhub, J., Dallal, G. and 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.

Paspatis G.A. and Karamanolis D.G., 1994. Folate supplementation and adenomatous polyps. *Dis Colon Rectum.*, 37:1340-1341.

Williams L.J. , Rasmussen S.A. , Flores A. , Kirby R.S. , Edmonds L.D. Decline in the prevalence of spina bifida and anencephaly by race/ethnicity. *Pediatrics*. 2005;116(3):580-6.

Appendix 1

DATA AND BACKGROUND INFORMATION ON FOLATE AND NEURAL TUBE DEFECTS IN THE EU

TABLE OF CONTENTS

INTRODUCTION

The purpose of this report is to provide an overview on the current practices of monitoring and registering neural tube defects (NTDs), recommended daily folate intakes, food fortification practices with folic acid, dietary intakes of folates and folate status of the European countries participating in the ESCO Working Group on Analysis of Risks and Benefits of Fortification of Food with Folic Acid.

The report comprises of the following areas:

- national congenital anomaly registries;
- data on prevalence of NTDs;
- recommendations on daily intakes of folate and folic acid;
- supplementation with folic acid;
- voluntary food fortification with folic acid;
- mandatory food fortification with folic acid;
- folate/folic acid intake data; and
- folate status.

Membership of the ESCO Working Group comprised of experts from the following countries: Belgium, Denmark, France, Germany, Ireland, Hungary, Italy, the Netherlands, Norway, Romania, Sweden, Switzerland and the United Kingdom. This report contains data only from those countries involved in the Working Group. Each member of the ESCO Working Group provided data in the form of a report for their respective country.

CONGENITAL ANOMALY REGISTRIES AND PREVALENCE OF NEURAL TUBE DEFECTS

Congenital anomaly registries

Data on NTDs are collated and registered through congenital anomaly registries. Many European countries collaborate in the European network of population-based registries for the epidemiologic surveillance of congenital anomalies (EUROCAT), which is a WHO Collaborating Centre for the Epidemiological Surveillance of Congenital Anomalies (http://www.who.int/genomics).

The surveillance of congenital anomalies serves the following main purposes: to facilitate the identification of teratogenic (malformation causing) exposures; to assess the impact of primary prevention, prenatal screening policy and practice at a population level. More than 1.5 million births are surveyed every year in Europe with 43 registries in 20 countries, covering 28% of the European birth population. The number and size of registries vary considerably across countries. In Hungary, Norway and Sweden all births are covered by the EUROCAT registry, whereas in Denmark, Germany, the Netherlands and Switzerland, 10% or less of all births are covered. In many cases, data is only available for certain regions of a country which participate in the EUROCAT system.

Table 1 provides an overview of the coverage of the European population by EUROCAT registries (from August 2006). All countries, except Romania, represented in the ESCO Working Group participate in EUROCAT. Sweden and Norway participate as associate members.

Table 1: EUROCAT registries of congenital anomalies in selected European countries

Country	Registry	Number of Annual births in registry	Number of Annual births in country	Country coverage (%)
Belgium	Antwerp	18,100		
	Hainaut	12,000		
	Total	30,100	115,500	26.1
Denmark	Odense	5,300	64,800	8.1
France	Auvergne	13,400		
	Paris	38,300		
	Central East	91,000		
	Strasbourg	13,400		
	Total	156,100	789,100	19.8
Germany	Mainz	3,200		
	Saxony-Anhalt	17,000		
	Total	20,200	742,500	2.7
Hungary		113,800	113,800	100.0
Ireland	Cork and Kerry	8,500		
	Dublin	23,400		
	SE Ireland	6,300		
	Total	38,200	60,500	63.2
Italy	Campania	59,900		
	Emilia Romagna	27,400		
	North East	60,200		
	Sicily	16,000		

Country	Registry	Number of Annual births in registry	Number of Annual births in country	Country coverage (%)
	Tuscany	27,700		
	Total	191,300	528,300	36.2
Netherlands	North	30,000	195,600	10.2
Norway		57,400	57,400	100.0
Sweden		99,500	99,500	100.0
Switzerland	Vaud	6,900	74,000	9.3
United Kingdom	Northern Region	30,300		
	North Thames	48,500		
	Oxford	6,700		
	Trent	64,300		
	Wales (CARIS)	31,300		
	Wessex	26,300		
	Total	207,400	721,200	28.8

Source: EUROCAT European Surveillance of Congenital Anomalies (from August 2006), http://www.eurocat.ulster.ac.uk

Prevalence of neural tube defects

NTDs are the most common major malformation of the central nervous system which can involve the brain, spinal cord, meninges (covering membranes), skull and spine. The terms used to describe these are based on clinical descriptions and the presumed embryological defect. These defects include spina bifida (also called myelomeningocele, accounting for approximately half of NTDs), anencephalus, encephalocele and iniencephalus.

The total prevalence of NTD-affected pregnancies includes all NTD affected live-births (including neonatal deaths), still-births and preterm abortions (including induced abortions). Data on the prevalence of NTDs from EUROCAT is provided in Table 2. Some countries have national surveillance systems for the registration of NTDs.

The available data show that the prevalence of NTD-affected pregnancies range from 4.13 to 19.70 per 10,000 births. However, reported data on NTDs are subject to under notification and diagnostic misclassification. Completeness of reporting varies according to the type of condition being notified; for example, data on cases of anencephaly and spina bifida, which are readily visible conditions, tend to be more complete compared to less visible conditions. Levels of reporting also vary according to the different regions within countries. Another issue is the underreporting of NTD terminations. In some countries terminations are illegal such as in Ireland, therefore the actual number of NTD-affected pregnancies is likely to be higher than reported.

Comparisons of the proportion of cases prenatally diagnosed, the average gestational age at diagnosis, diagnostic methods used, and the proportion of cases resulting in termination of pregnancy show enormous variation between and within countries. Such variation may result from cultural differences, underlying policy or individual uptake, differing interpretations of the scientific evidence in the design and implementation of screening, or differences in the health services provided.

REFERENCES

EUROCAT (European Surveillance of Congenital Anomalies).

<http://www.eurocat.ulster.ac.uk>

Table 2: Prevalence of NTD-affected pregnancies in selected European countries participating in EUROCAT (per 10,000 births)

Country	Year range	No. of live births (LB)	No. of fetal deaths/still births from 20 weeks gestation (FD)	No. of terminations of pregnancy for fetal anomaly following prenatal diagnosis (TOPFA)	No. of LB+FD+ TOPFA	LB rate	LB+FD rate	LB+FD+ TOPFA rate
Belgium, Antwerp	1990-2006	71	19	134	224	3.00	3.80	9.46
Belgium, Hainaut	1980-2006	101	28	185	314	3.37	4.29	10.43
Denmark	1980-2006	81	31	60	172	5.68	7.81	11.99
France, Auvergne	2002	2	0	9	11	1.51	1.49	8.21
France, Paris	1981-2006	155	89	932	1176	1.65	2.58	12.42
France, Strasbourg	1982-2004	58	23	266	347	1.95	2.73	11.70
France, Central East	1980-2004	459	49	797	1305	1.96	2.15	5.53

Country	Year range	No. of live births (LB)	No. of fetal deaths/still births from 20 weeks gestation (FD)	No. of terminations of pregnancy for fetal anomaly following prenatal diagnosis (TOPFA)	No. of LB+FD+ TOPFA	LB rate	LB+FD rate	LB+FD+ TOPFA rate
France, Rhone-Alps	2006	9	2	55	66	1.90	2.30	13.79
Germany, Mainz	1990-2006	70	7	40	117	11.78	12.96	19.70
Germany, Saxony-Anhalt	1987-2006	91	17	180	288	*3.44*	4.06	10.82
Hungary	1998-2002	135	11	260	406	*2.80*	2.60	7.23
Ireland, Cork and Kerry	1996-2003	54	26	0	80	8.62	12.70	12.70
Ireland, Dublin	1980-2006	741	308	0	1049	12.88	18.12	18.12
Ireland, Galway	1981-1999	66	18	0	84	12.19	15.41	15.41
Ireland, SE	1997-2006	52	12	0	64	8.41	10.35	10.35

Country	Year range	No. of live births (LB)	No. of fetal deaths/still births from 20 weeks gestation (FD)	No. of terminations of pregnancy for fetal anomaly following prenatal diagnosis (TOPFA)	No. of LB+FD+ TOPFA	LB rate	LB+FD rate	LB+FD+ TOPFA rate
Italy, Campania	1996-2004	94	8	223	325	1.97	2.13	6.78
Italy, Emilia Romagna	1981-2006	195	21	151	367	2.95	3.26	5.53
Italy, North East	1981-2003	327	45	333	705	2.90	3.30	6.25
Italy, Sicily	1991-2004	100	6	-	106	3.89	4.13	4.13
Italy, Tuscany	1980-2006	105	18	213	336	2.10	2.45	6.69
Netherlands	1981-2006	239	82	103	424	5065	7.54	9.96
Norway	1999-2005	139	13	267	419	3.45	3.74	10.30
Sweden (100,000 birth/year)	1999-2005	155		458				8.76

Country	Year range	No. of live births (LB)	No. of fetal deaths/still births from 20 weeks gestation (FD)	No. of terminations of pregnancy for fetal anomaly following prenatal diagnosis (TOPFA)	No. of LB+FD+ TOPFA	LB rate	LB+FD rate	LB+FD+ TOPFA rate
Switzerland	1989-2006	31	3	109	143	2.31	2.52	10.61
UK, England and Wales[5]	1995-2004		104[6]	298	402		1.7	6.4
UK, Scotland[7]	1992-2003		33	33	66		6.0	12

[5] Source of birth data: National Congenital Anomaly System. Data as at 1st September 2005.
Source of termination data: Series AB ONS Abortion statistics 1995-2001; DH Statistical Bulletin - Abortion statistics 2002-2004
[6] Figure include NTD miscarriages
[7] Source of birth data: Scottish Perinatal and Infant Mortality & Morbidity Report (2003), Information & Statistics Division, National Health Service Scotland Termination data source: General Register Office for Scotland and Notifications (to the Chief Medical Officer for Scotland) of abortions performed under the Abortion Act 1967.

Country	Year range	No. of live births (LB)	No. of fetal deaths/still births from 20 weeks gestation (FD)	No. of terminations of pregnancy for fetal anomaly following prenatal diagnosis (TOPFA)	No. of LB+FD+ TOPFA	LB rate	LB+FD rate	LB+FD+ TOPFA rate
UK, Northern Ireland[8]	1992-1993		16				6.9	

Source: EUROCAT Website Database: http://www.bio-medical.co.uk/eurocatlive (data uploaded 08/12/2008). NTDs resulting from chromosomal or monogenetic disorders are included. UK data: national surveillance data.

[8] Source of birth data: Child Health Systems (Northern Ireland and Social Services Boards)

RECOMMENDATIONS ON FOLATE INTAKES IN EUROPE

Function and metabolism

The term folate(s) 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.

Folic acid is the synthetic form of folate which is commonly used in supplements and food fortification. It is more stable in comparison to other forms of the vitamin and is not present naturally in foods.

Folate acts as a coenzyme in several single carbon transfer reactions to synthesize components of DNA, RNA and proteins.

The bioavailability of naturally occurring folate is lower than that of folic acid. This difference in bioavailability is partly due to the fact that folic acid can be absorbed directly, whereas food folates (mainly polyglutamates) need to be enzymatically hydrolysed by a (brush border associated) deconjugase enzyme to the monoglutamyl form in the gut before absorption. Other factors that influence bioavailability are matrix effects and the presence of inhibitors of the deconjugase enzyme in some foods.

Folic acid enters the folate cycle after reduction by (dihydro-)folate reductase. At higher intakes of folic acid (for example a single oral dose of 260µg) it may appear unchanged in the circulation (Smith et al., 2008). Under normal conditions 5-MTHF (as monoglutamate) is the only form present in plasma.

Folates enter cells as monoglutamates, but are rapidly modified by the addition of four to eight glutamate residues to form long side chains. The folates that are used as coenzymes and regulatory molecules in the body are all in the reduced form as tetrahydrofolate derivatives. About 50% of the body stores of folate, estimated to be 13-28mg, are considered to be present in the liver (IOM, 1998).

Recommended folate intakes

Recommended daily intakes are usually based on the requirement to maintain the level of folate in serum or plasma red blood and liver cells within normal physiological ranges and in some countries, to prevent high levels of plasma homocysteine concentrations. Often recommendations are expressed as dietary folate equivalents (DFE, 2000), thereby correcting for the lower bioavailability of food folate of 1µg of DFE is equal to 1µg food folate and is equal to 0.5µg folic acid taken on an empty stomach. In some European countries, the conversion proposed by the Institute of Medicine (IOM, 1998) in the USA is used, which accounts for the fact that synthetic folic acid, which is consumed together with foods, has a reduced bioavailability of 85%. Thus, 1µg folic acid from a fortified diet equals 1.7µg folate and 1 µg from supplements equals 2µg folate. Approaches using depletion of folate and considerations of folate catabolism suggest daily requirements of 50-100µg. The European population reference intake for folate is 200µg/day for adults and 400µg/day for pregnant

women (SCF, 1993). A higher requirement is set for pregnancy as red cell folate concentrations decrease during pregnancy. This decrease is due to haemodilution and the extra demand for folate by the foetus. This decrease in red blood cell folate can be prevented by an additional dietary folate intake of 200 µg or a supplement intake of 100µg folic acid daily. A daily increment of approximately 150µg is suggested for normal lactation (SCF, 1993). Across the European countries, recommended daily intakes for folate for adults range from 200 to 400µg/day depending on the biological outcome used as a basis of the recommendation. Lower values tend to be based on maintenance of folate concentration in liver samples, whereas higher values tend to be based on maintaining homocysteine levels in plasma which have been associated with cardiovascular disease (CVD) risk. For pregnant women, recommendations range from 300 to 600µg/day. For lactating women, recommended daily intakes range from 260 to 600µg/day. Recommendations for children in most European countries are extrapolated from the recommendations for adults on the basis of body weight. The recommended daily intakes for children are much higher in Germany and Switzerland than in other European countries. An overview of the recommended intake levels for folate is provided in Table 3.

Tolerable upper intake level

In 2000, the SCF set a tolerable upper intake level (UL) for folic acid of l mg/day for adults. The lowest-observed-adverse-effect level (LOAEL) was set at 5mg/day. It was concluded that doses up to 1 mg/day of folic acid were unlikely to cause masking of the haematological signs of vitamin B12 deficiency. No data were available to suggest that other life-stage groups had increased susceptibility to adverse effects of high folic acid intake. Therefore, the UL was also deemed applicable for pregnant or lactating women. The UL for children and adolescents were set on the basis of body weight, as follows:

Age (years)	**Tolerable upper intake level (UL) for folic acid (µg/day)**
1-3	200
4-6	300
7-10	400
11-14	600
15-17	800

REFERENCES

Data sources by countries:

Belgium:

Recommandations nutritionnelles pour la Belgique, March 2008. Conseil Supérieur d'Hygiène.

Denmark:

Nordic Council of Ministers. Nordic Nutrition Recommendations 2004. 4 ed. Åhus: *Scanprint*.

France:

Potier de Courcy, G., Christidès, J.P., Hercberg, S. in Martin, A., 2001. Apports nutritionnels conseillés pour la population française. *3ème Ed. Tec & Doc*.

Germany:

DGE (Deutsche Gesellschaft für Ernährung), Österreichische Gesellschaft für Ernährung, Schweizerische Gesellschaft für Ernährung, Schweizerische Gesellschaft für Ernährungsforschung, Schweizerische Vereinigung für Ernährung (Hrsg.), 2000. Referenzwerte für die Nährstoffzufuhr. Frankfurt am Main: *Umschau Braus Verlag*.

BfR (Bundesinstitut für Risikobewertung), 11-12 January 2007. Folate status of the population in the European Community and strategies for change. *BfR*.

Hungary:

OETI (National Institute of Food Safety and Nutrition). <http://efrira1.antsz.hu/oeti/taplal/varando2.pdf>

Ireland:

FSAI (Food Safety Authority of Ireland), 1999. Recommended dietary allowances for Ireland. <www.fsai.ie/publications/reports/recommended_dietary_allowances_ireland_1999.pdf>

The Netherlands:

Gezondheidsraad (Health Council), 2003. Voedingsnormen: vitamine B6, foliumzuur en vitamine B12. *Den Haag*.

Norway:

Nordic Council of Ministers, 2004. Nordic Nutrition Recommendations 2004. 4 ed. *Åhus*.

Switzerland:

D-A-CH (Deutsche Gesellschaft für Ernährung, Österreichische Gesellschaft für Ernährung, Schweizerische Gesellschaft für Ernährungsforschung, Schweizerische Vereinigung für Ernährung), 2000. Reference values for the nutrient intake. *Umschau/Braus-Verlag*, Frankfurt.

United Kingdom:

Department of Health. Dietary Reference Values for Food, Energy and Nutrients for the UK, (Report on Health and Social Subjects, No. 41). 1991.. London: HMSO

Food and Nutrition Board, Institute of Medicine, 1998. Report of the Standing Committee on the scientific evaluation of dietary reference intakes (DRIs) and its panel on folate and other B-vitamins and choline.

IOM (Institute of Medicine) Dietary Reference Intakes for Thiamin, Riboflavin, Niacin, Vitamin B6, Folate, Vitamin B12, Pantothenic Acid, Biotin, and Choline, 1998 (ISBN 0309065542)

SACN (Scientific Advisory Committee on Nutrition), Folate and Disease Prevention, 2006. London: TSO.

SCF (European Scientific Committee on Food), 2000. Opinion of the Scientific Committee on Food on the Tolerable Upper Intake Level of Folate (expressed on 19 October 2000). *Brussels: EC.*

SCF (European Scientific Committee on Food), 1993. Nutrient and energy intakes for the European Community (opinion expressed on 11 December 1992). Reports of the Scientific Committee for Food, thirty-first series. *Brussels: Directorate-General Industry*.

Smith, A.D., Kim, Y.-I. and Refsum, H.. 2008. Is folic acid good for everyone? *Am J Clin Nutr*. 87:517-533.

Table 3: Recommendations of daily intakes of folate in selected European countries (µg/day)[9]

Age (years) / sex	0-1	1-3	4-6	4-8	4-9	7-9	7-10	9-13	10-14	10-15	11-14	11-18	14-18	15-17	15-18	16-18	19+ men	19+ women	Pregnant women	Lactating women
Belgium	50	100	130				150				180				200		200	200	400	350
Denmark	50	60											300				300	300-400[a]	500	500
France	70	100	150			200				250						300/330	330	300	400	400
Germany	60-80	200	300			300			400						400		400	400	600	600
Hungary																	200	200		
Ireland	50	100	200				200				300			300			300	300	500[b]	400[c]
Italy	50	100	130				150				180				200		200	200	400	350
Netherlands	40-60	85		150				225					300				300	300	400	400
Norway													300				300	300-400[a]	500	500

[9] In some countries folate dietary recommendations are expressed as dietary folate equivalents (DFE).

Age (years) / sex	0-1	1-3	4-6	4-8	4-9	7-9	7-10	9-13	10-14	10-15	11-14	11-18	14-18	15-17	15-18	16-18	19+ men	19+ women	Pregnant women	Lactating women
Romania																				
Sweden													300				300	300-400[a]	500	500
Switzerland	60-80	200			300				400						400		400	400	600	600
UK	50	70	100				150					200					200	200[10]	600[11]	260
SCF for Europe	50	100	130				150				180			200			200	200	400	350

a In Denmark, Sweden and Norway the Nordic Nutrition Recommendations 2004 are used.
b Second half of pregnancy.
c First six months of lactation.

[10] 200µg is recommended for all women plus an extra 400µg of folic acid to women who might become pregnant
[11] 600µg is recommended until 12th week of pregnancy

RECOMMENDED PERICONCEPTIONAL FOLIC ACID SUPPLEMENTATION

In Table 4 an overview is presented on recommendations for periconceptional folic acid supplementation in European countries, including information about compliance and how the recommendations are implemented.

All countries advise women of childbearing age who are planning a pregnancy to supplement their diet with 400µg of folic acid per day to decrease the risk of having a baby with an NTD. In some countries (including Italy, Germany, Switzerland, Italy, UK, the Netherlands and Belgium) women who had a previous pregnancy affected by a NTD are advised to take a supplement of 4-5mg/day. The recommended period of folic acid supplementation differs across countries. In some countries it is recommended that supplementation starts as soon as contraception is stopped whilst in others it is recommended that all women who are planning a pregnancy or who could become pregnant take folic acid supplements. Most countries recommend continuing supplementation until the end of the 1st trimester or 12th week of pregnancy.

Compliance with the recommendations is not evaluated in all countries.

The Directive 2002/46/EC[12] of the European Parliament and Council 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. The aim is to harmonise the legislation and to ensure that these products are safe and appropriately labelled so that consumers can make informed choices.

[12] 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

REFERENCES

Data sources by countries:

Denmark:

Knudsen, V.K., Orozova-Bekkevold I., Rasmussen L.B., Mikkelsen T.B., Michaelsen K.F., Olsen S.F., 2004. Low compliance with recommendations on folic acid use in relation to pregnancy: is there a need for fortification? *Public Health Nutr*. 7:843-850.

Germany:

Koletzko B, von Kries R Prevention of neural tube defects by folic acid administration in early pregnancy, 1995. Joint recommendations of the German Society of Nutrition, Gynecology and Obstetrics, Human Genetics, Pediatrics, Society of Neuropediatrics. Gynäkol. *Geburtshilfl. Rdsch.* 35: 2-5.

Hungary:

(OETI) National Institute of Food Safety and Nutrition. <http://efrira1.antsz.hu/oeti/taplal/varando2.pdf>

The Netherlands:

Voedingsraad/Gezondheidsraad, 1993. Vervolgadvies inzake foliumzuurvoorziening in relatie tot neuralebuisdefecten. Den Haag: Voorlichtingsbureau voor de Voeding.

Switzerland:

Jans-Ruggli S, Baerlocher K., 2005. Kenntnisse über Folsäure und Folsäurestatus bei Müttern. Häufige Einnahme von Folsäuresupplementen, aber ungenügende Prävention von Neuralrohrdefekten. In: Eichholzer M, Camenzind-Frey E, Matzke A, et al. (eds), Fifth Swiss Nutrition Report. Berne: *Swiss Federal Office of Public Health*, pp.71-86.

Baerlocher K, Eichholzer M, Lüthy J, et al., 2002. Massnahmen zur Prophylaxe von Neuralrohr-Defekten und zur Verbesserung der Folsäure-Versorgung in der Schweiz. Report of the Working Group on „Folic acid prevention“ of the Swiss Federal Commission of Nutrition. *Swiss Federal Office of Public Health*, Bern.

United Kingdom:

Department of Health 2000. Department of Health 1992. The Health Survey of England 2002, Blake et al., 2003

Table 4: Recommendations for (periconceptional) folic acid supplementation in selected European countries

Country	Recommendation	Compliance	Method used to communicate recommendation
Belgium	After stopping birth control pills, or at least one month (but preferably three months) before conception it is recommended to take a supplement of 400µg/day until the third month of pregnancy. For high risk groups (previous child with NTD, epileptics, diabetics), a supplement of 4 mg/day is recommended. Supplementation throughout pregnancy and lactation is recommended.	One study group: more than 80% of the women complied with the recommendation.	Brochure (in French and Dutch) distributed by GP's, gynaecologists, pharmacists, targeted media to women of childbearing age and pregnant women.
Denmark	Women planning to become pregnant are recommended to take a supplement of 400µg folic acid daily until the twelfth week of gestation.	2000-2002: 13% complied with recommendation; about 60% took a daily folic acid supplement with a lower content or only during part of the recommended period.	Leaflets distributed through GP´s, gynaecologists, midwifes, libraries, pharmacist and articles in women's magazines
France	400 µg/day for women planning to become pregnant immediately after stopping contraception. Unplanned pregnancies: supplement to be taken for at least 4 weeks.		Information of women *via* nutrition guide.

Country	Recommendation	Compliance	Method used to communicate recommendation
Germany	Women who wish or could become pregnant are advised to take a supplement of 400 µg of folic acid per day from at least 4 weeks before conception until the end of the first trimester. If a woman already has a child with NTD, an additional intake of 4 mg folic acid per day is recommended.	Some regional data suggest that less than 10% of the women of childbearing age follow the recommendations with respect to time and dosage.	Information leaflets.
Ireland	Women of childbearing age who are sexually active are advised to take 400µg of folic acid per day; a supplement providing 400µg is recommended. Ideally folic acid supplements should be taken at least two months before pregnancy, continuing until the 12th week of pregnancy.		Information leaflets since early 1990s. In 2005, a "Folic Acid – Today and Everyday" campaign was undertaken which included print and television advertising, an information leaflet and a dedicated website: www.folicacid.ie.

Country	Recommendation	Compliance	Method used to communicate recommendation
Italy	Women planning or not actively excluding a pregnancy are advised to take a supplement of 400 µg/day of folic acid at least one month before conception and during the first trimester of pregnancy. High risk women should take 4-5 mg of folic acid daily.		
Netherlands	Women wishing to conceive are advised to take a supplement of 400µg folic acid daily from at least 4 weeks before conception until the 8th week of pregnancy.	90% of pregnancies are planned (1996); 77% of women know about folic acid (1996); 53% use PGA during part of recommended period (1998); 60% uses folic acid (2000) and 36% uses correctly (2000).	National and local media campaigns with follow-ups since 1995.
Norway	Women planning to become pregnant: 400 µg/day folic acid from 1 month before conception and during the first trimester.	2000-2003: folic acid supplement use increased from 12% at 2 months before pregnancy to 47% at gestational month; 10% of women used supplement as recommended.	

Country	Recommendation	Compliance	Method used to communicate recommendation
Sweden	Women of fertile age are recommended to take folic acid supplement before getting pregnant until the 12th week of pregnancy.		
Switzerland	All women of childbearing age without safe contraception are recommended, in addition to a folate rich diet, to take a supplement of at least 400µg folic acid daily (preferably as a multivitamin) until 12 weeks after conception. Women who had a previous pregnancy affected by NTD are advised to take a supplement of 4-5 mg daily.	In 2002/2003, 37% of women in hospitals used supplements during the recommended period of time. Overall 80% of pregnancies were planned; for the group younger than 25 years, this was 53%. 46% of women with planned pregnancies took folic acid correctly.	
UK	Women who could become pregnant or are planning a pregnancy are advised to take an additional 400 µg/day of folic acid as a supplement until the 12th week of pregnancy. They are also advised to eat more foods naturally rich in folate, and foods fortified with folic acid, especially breakfast cereals.	England 2002: 55% of women who planned their pregnancy increased their folate intake by taking supplements or modifying their diet, while 79% of women increased their folate intake during pregnancy.	Advice is given across the UK through, for example, Primary Care Trusts, family planning clinics and NHS Direct.

FOOD FORTIFICATION WITH FOLIC ACID

Voluntary food fortification

Voluntary fortification of foods with folic acid is currently practiced in many countries around the world, including the EU. It is considered to help increase the folate intakes of population groups. Food can be fortified in the EU under Regulation 1925/2006/EC[13] on the addition of vitamins and minerals and of certain other substances to foods. This regulation allows for the fortification of all foods, except unprocessed food sand alcoholic beverages, with folic acid. However, prior to this regulation, many foods have been voluntarily fortified with folic acid. According to the Regulation, the Commission has been given the competence to set maximum amounts of vitamins and minerals in food, and is currently holding discussions with stakeholders and Member States. Maximum amounts will be set concurrently for vitamins and minerals in fortified foods and in food supplements (European Commission, 2006), thus, in the future there will be harmonised maximum levels for addition of folic acid to supplements and fortified foods.

An overview of the voluntary folic acid food fortification practices in European countries is provided in Table 5. All countries except Sweden voluntarily fortify a wide range of foods with folic acid including flour/bread, breakfast cereals, dairy products, fruit juices and fat spreads. The levels vary widely, with the highest levels added to spreads (up to 1000µg/100g). Some countries (such as Belgium, Denmark, the Netherlands and UK) have restrictions in place on the level of folic acid which can be added to food. In Norway and Denmark approval is required before a folic acid-fortified food can be marketed.

Mandatory food fortification

Mandatory fortification of food with folic acid has been implemented in several non-European countries, including USA, Canada, and Chile, and is due to be enforced in Australia from September 2009. From 2004 to 2007, the number of countries who introduced national regulations for mandatory wheat flour fortification increased from 33 to 54 (CDC, 2008). Some of these countries fortify folic acid in combination with iron and other B vitamins. To date, no European country has implemented mandatory fortification of foods with folic acid. In Switzerland it was decided not to implement mandatory fortification following the recommendation by the Swiss Federal Commission of Nutrition. Ireland and the UK are currently considering mandatory fortification following recommendations by their national expert committees (see below for further details).

Ireland

In 2006, Ireland's National Committee on Folic Acid Fortification recommended that most white, brown and wholemeal breads sold in the country be fortified with 120µg of folic acid per 100g of bread as consumed (Food Safety Authority of Ireland & Department of Health and Children 2006). Ireland has one of the highest rates of NTDs in Europe. Although

[13] OJ L 404/26, 30/12/2006: 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.

supplements containing 400µg per day are promoted, the committee only recommended 120µg to allow a good safety margin for those who consume large amounts of bread. Excluding some minor bread products and retail flour from the requirements has also been suggested to improve consumer choice so that those wishing to avoid fortified products are able to do so. The level of 120µg folic acid per 100 g bread was estimated to reduce the prevalence of NTD-affected pregnancies by about 24%. However, in 2008, Ireland's Implementation Group on Folic Acid Fortification recommended that the proposal for mandatory fortification of bread be put on hold for three reasons: (1) there has been a reduction in NTD prevalence in Ireland in recent years, likely due to the voluntary fortification of foods with folic acid; (2) as a result of voluntary fortification there has been an increase of about 30% in folic acid in the diet in the past 2-3 years; and (3) new scientific studies have suggested a relationship between high folic acid intakes and cancer risk.

United Kingdom

In the UK, the Scientific Advisory Committee on Nutrition (SACN) reviewed the evidence regarding the health benefits of dietary folate and recommended mandatory fortification of flour with folic acid (SACN, 2005). In June 2007, the Food Standards Agency (FSA) recommended mandatory fortification with folic acid of bread or flour to UK Health Ministers. In October 2007, the Chief Medical Officer wrote to the Chair of the FSA to request a further expert view of the evidence on folic acid and colorectal cancer risk. On 21 January 2008, a group of experts, including Members of the Scientific Advisory Committee on Nutrition (SACN), and the Committee on Carcinogenicity, met to consider this evidence on colorectal cancer risk. The SACN committee on 7 February 2008 agreed to defer a final decision on folic acid and cancer risk until the results from a number of ongoing cardiovascular disease trials are available in early 2009.

Switzerland

In 1997 a request was submitted to the Swiss Parliament to consider mandatory fortification of food with folic acid for the prevention of NTDs. In 2002 the Swiss Federal Commission of Nutrition published a report and recommended the following measures to increase the folic acid intake of the Swiss population and for the prevention of NTD: fortification of baking flour with 3mg folic acid and 10µg vitamin B_{12} per kg flour on a mandatory basis. This would have resulted in an estimated additional daily intake supply of 275µg folic acid and 1µg vitamin B_{12} per person (Baerlocher et al., 2002). Following a request from the Swiss Federal Office of Public Health, it was concluded in 2006 that there is no legal basis for mandatory fortification in Swiss law.

Modelling exercises

Several countries including France, Germany, the Netherlands, Norway, Switzerland and the UK have carried out model calculations or studies to estimate the effect of fortifying flour with different doses of folic acid on folate intake and reduction of NTDs.

France

In France, fortification of 350µg folic acid per 100g bread would result in a reduction of 170-210 NTD cases annually (total number of NTDs per year: 800-1000). In addition, fortification with 10µg vitamin B12 per 100g bread was proposed for consideration by AFSSA.

Germany

In Germany it was concluded that a nationwide flour fortification with 150µg folic acid per 100g would make a significant contribution to folic acid status without a high proportion of the population consuming folic acid intakes above the UL. It was proposed that such a measure should only be introduced if the number of voluntarily fortified foods and the level of added folic acid were reduced (to a maximum of 100µg per portion). It was also proposed that beverages/soft drinks or foods whose consumption is not limited through satiation, should no longer be fortified with folic acid; and salt fortified with folic acid should not be sold in parallel with fortified flour. It was concluded that a level of 150µg/ 100g would not be effective in increasing folic acid intake to such an extent that women of childbearing age would reach the additional intake of 400µg per day recommended for NTD prevention. In order to achieve this dose, the regular taking of food supplements would be the most effective method.

The Netherlands

In the Netherlands, the Health Council advised to improve the use of folic acid supplements around the time of conception through education and preconception care. In addition, the Government was advised to consider fortifying staple foods, such as bread and bread substitutes with folic acid. Fortifying bread and bread substitutes with 150µg folic acid per 100g flour after preparation would provide an additional 100µg of folic acid daily. This level of fortification was found to be acceptable, provided voluntary fortification of specific food products is discontinued to avoid children consuming high intakes of folic acid.

Norway

In Norway a scenario study carried out to estimate the folate intake of different population groups showed that mandatory fortification would not provide the majority of women in the target group with 400µg/day without other population groups exceeding the safe upper level for folic acid. If bread was fortified with 100µg folic acid per 100g the least number of persons would exceed the UL (19% of children aged 4 years and 15% aged 9 years) whereas more than 50% of the fertile and pregnant women would not reach the recommended intake.

Switzerland

In Switzerland, data from a surveillance programme showed that individuals who consumed fortified food products (estimated maximum intakes of 919µg/day for women and 1454µg/day for men) had intakes above the recommended levels (Beer-Borst et al., 2005).

Italy

In Italy a preliminary evaluation carried out by the Italian Folic Acid Network, coordinated by the Italian National Health Institute (ISS), indicated that a substantial fraction of the adult and adolescent population may exceed the tolerable upper level of folic acid. The evaluation was based on flour fortification levels used in the United States and took into account the high consumption levels of flour based foods in Italy, as well as recent evidence on potential tumour promoting effects of high folic acid intake. The Network recommended that periconceptional supplementation and the promotion of healthy dietary habits are the most effective and safe strategies to prevent NTDs.

United Kingdom

The UK has modelled the effect of flour fortification on the UK population (SACN, 2006). The modelling showed that removing folic acid consumed through voluntary fortification (largely from breakfast cereals and fat spreads) and introducing mandatory fortification of white and brown flour with folic acid at a level of 300μg/100g (equivalent to 225μg/100g of food after processing), would reduce the number of NTD pregnancies each year by 11-18%, reduce the proportion of the population with intakes below the reference nutrient intake (RNI) (the RNI represents the amount of a nutrient likely to meet the needs of 97.5% of the population) (Department of Health, 1991) from 23% to 5%, while the number with intakes above the tolerable upper intake level (UL) (SCF, 2000) remain at the current level (0.2%).

Subsequently the Food Standards Agency (FSA) remodelled updated UK folic acid consumption data, exploring a range of different scenarios for voluntary fortification and supplement consumption.

REFERENCES

Data sources by countries:

Belgium:

Rasmussen, S.E., Andersen, N.L., Dragsted, L.O., Larsen, J.C., 2006. A safe strategy for addition of vitamins and minerals to foods. *Eur J Nutr*. 45:123-135.

France:

AFSSA (Agence française de sécurité sanitaire des aliments), 2003. Enrichissement de la farine en vitamines B en France proposition d'un programme-pilote. Paris. <http://www.afssa.fr>

Germany:

Weissenborn A, Burger M, Mensink GBM, Klemm C, Sichert-Hellert W, Kersting M, Przyrembel H, 2005. Folic acid intake of the German population. *BfR-Wissenschaft*. Berlin.

Hungary:

Czeizel, A.E. and Kökény, M., 2002. Bread is fortified with folic acid in Hungary. *BMJ*. 325:3 91.

Ireland:

FSAI (Food Safety Authority of Ireland), 2006. Report of the National Committee on Folic Acid Food Fortification. <http://www.fsai.ie/publications/reports/folic_acid.pdf>

FSAI (Food Safety Authority of Ireland), 2008. Report of the Implementation Group on Folic Acid Food Fortification to the Department of Health and Children. <www.fsai.ie/publications/reports/folic_acid_implementation.pdf>

Italy:

Pierini, A., Bianchi, F., Salerno P. and Taruscio D., 2006. National register of rare diseases: congenital malformation and folic acid. *Rapporti ISTISAN 06/34*, 114 p.

Salerno, P., Bianchi, F., Pierini, A., Baldi, F., Carbone, P., Mantovani, A. and Taruscio, D. 2008. Folic acid and congenital malformation: scientific evidence and public health strategies. *Ann Ig.* 20(6):519-30.

The Netherlands:

Gezondheidsraad, 2008. Naar een optimaal gebruik van foliumzuur (Towards an optimal use of folic acid). *Publicatienr. 2008/02*. Den Haag.

Norway:

Grimstad, A.L., 2007. Folic acid fortification of foods. Estimated intake of folate from different fortification scenarios in Norwegian population groups. University of Oslo, Department of Nutrition, Faculty of Medicine. MSc thesis.

Switzerland:

Verordnung des Eidgenössische Departement des Innern (EDI) über den Zusatz essenzieller oder physiologisch nützlicher Stoffe zu Lebensmitteln vom 23. November 2005; Artikel 18 Absatz 2 der Lebensmittel- und Gebrauchsgegenständeverordnung vom 23. November 2005 (LGV), SR 817.022.32. <http://www.admin.ch/ch/d/as/2005/6345.pdf>

Beer-Borst, S., Costanza, M.C. and Morabia, A., 2005. In: Eichholzer M et al. (eds). Fifth Swiss Nutrition Report. *Swiss Federal Office of Public Health*, Bern. pp.751-766.

Foundation Folic acid offensive, Switzerland. <www.folsaeure.ch>

Baerlocher, K., Eichholzer, M., Lüthy, J., Moser, U. and Tönz, O., 2002. Massnahmen zur Prophylaxe von Neuralrohr-Defekten und zur Verbesserung der Folsäure-Versorgung in der Schweiz. Report of the Working Group on „Folic acid prevention" of the Swiss Federal Commission of Nutrition. *Swiss Federal Office of Public Health*, Bern.

Rechtsgutachten über die Verfassungsmässigkeit einer "obligaten" Anreicherung von Getreidemehl mit Folsäure (Nov 2006).

United Kingdom:

Department of Health. *Dietary Reference Values for Food, Energy and Nutrients in the United Kingdom. (Report on Health and Social Subjects, No. 41).* London: HSMO, 1991.

SACN (Scientific Advisory Committee on Nutrition), Folate and Disease Prevention. London: TSO, 2006.

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>

SCF (European Scientific Committee on Food). Opinion of the Scientific Committee on Food on the Tolerable Upper Intake Level of Folate 2000. Online at <http://ec.europa.eu/comm/food/fs/sc/scf/out80e_en.pdf>

Other:

CDC (Centers for Disease Control), Trends in Wheat-Flour Fortification with Folic Acid and Iron - Worldwide, 2004 and 2007. MMWR Morbidity and Mortality Weekly Report, January 11, 2008 / 57(01);8-10. <www.cdc.gov/mmwr/preview/mmwrhtml/mm5701a4.htm>

Table 5: Voluntary folic acid food fortification in selected European countries.

Country	Voluntary fortification	Restriction	Major foods	Levels of folic acid
Belgium	Yes	A daily portion of foods labelled 'fortified with folic acid' must contain 15–200% of 200µg		
Denmark	Yes (Approval required)	Max. 23µg folic acid per 100 kcal	Only 2 products (with very low market share)are fortified: crackers and orange juice	2.8µg/100 g 9µg/100 ml
France	Yes	None		
Germany	Yes		Dairy products; beverages; breakfast cereals Sweets, fruit teas, convenience foods, fat spreads, etc Iodised and fluoridated salt	7 µg/100 ml (soft drinks) – 1.300 µg/100 g (sweets); 100µg/g salt
Hungary	Yes		Bread	330µg/100 g

Country	Voluntary fortification	Restriction	Major foods	Levels of folic acid
Ireland	Yes		FSAI survey identified 104 breakfast cereals, 38 breads. 27 cereal bars, 15 fat spreads, 11 fruit juices four milks, four dried soups and two yoghurts on the Irish market containing folic acid	0.07 (fruit juice) – 1000 (fat spreads) µg/100 g
Italy	Yes		Breakfast cereals, fruit juices, rusk, biscuits	
The Netherlands	Yes (since 2006)	Fortification with 100µg folic acid per 100 kcal allowed	Breakfast cereals; no products until 2003/2004	
Norway	Yes (Approval required)		Energy bar Milk variant	78µg 13µg/100 ml milk
Sweden	No			
Switzerland	Yes	The content of folic acid must be declared on the package	Large range of food products are fortified with folic acid	

Country	Voluntary fortification	Restriction	Major foods	Levels of folic acid
United Kingdom	Yes	Subject to the provisions of the Food Safety Act 1990	Breakfast cereals (about 3/4 on market are fortified) Cereal bars (brand specific)	150-400µg/100 g100-170µg/100 g
			Fat spreads (about 1/3 on the market are fortified)	500µg/100 g
			Soya milk (brand specific)	14-34µg/100 ml
			Some malt drinks (brand specific)	125-250µg/100 g dry weight
			Some savoury spreads (brand specific)	up to 500µg/100 g

FOLATE INTAKE

Dietary folate intake can be estimated from food consumption data. Tables 6 and 7 provide an overview of the available data on mean or median daily intakes of folate for children and adults respectively.

Children:

Data for children were provided by Germany, Denmark, the Netherlands, Norway and UK. For children the mean/median intake ranged from 113 to 314µg/day.

Adults:

The mean/median dietary folate intakes range from 151 to 345µg/day for men and from 122 to 339µg/day for women. Average dietary and supplement intakes for women range from 220 to 478µg/day and 338 to 385µg/day for men. Intakes below the European population reference intake of 200µg/day were reported in Hungary, the Netherlands, Switzerland and Norway.

In general, for most countries, the average daily intake of folate was higher in adult men than adult women. It is important to note that different dietary survey methods were used in the different countries to assess nutrient intakes, e.g. food records, 24hr recall, FFQ etc. Furthermore some countries assessed folate intake (including supplement use) whilst others assessed only dietary folate intake. There is also variation between the different analytical methods used for compilation of the nutrient databases.

Additional information on folate intake by country:

Ireland

Since 1999 many foods have been fortified in Ireland. The FSAI used a probabilistic approach to re-model the 1999 adult consumption data to provide updated estimates of folic acid intake based on folic acid food fortification patterns observed on the Irish market in 2007. It was estimated that the average folic acid intake for the target group of women aged 18 to 50 was now 90 µg per person per day. The updated estimate of folic acid intake is thought, by national experts, to still represent an underestimation of actual intake. It can be demonstrated that spreads and bread fortified with folic acid, whilst impacting on intake in the target group of women, have made an even greater contribution to folic acid intake in the older population groups. In contrast, the contribution of breakfast cereals is smaller in 2007 than it was in 1999 largely due to lower average fortification concentrations.

Germany

In Germany, recommended intakes (400µg/d) were only achieved by 50-70% of the adult population (Mensink et al., 2002). Also recent data from the German National Food Consumption Survey, in which fortified breakfast cereals and beverages were taken into consideration for the first time, show that adults only achieve median intakes below 300µg folate equivalents per day (MRI, 2008). However, consumption of fortified foods and resulting intake of folate equivalents has probably still been underestimated as there are also other food categories besides breakfast cereals and beverages fortified with folic acid.

The Netherlands

The Health Council in the Netherlands concluded in 2008 that the suboptimal folate intake was no reason for a change in policy. The Council saw no reason to improve folate intake in the general population through food fortification or through supplementation, as it remains unclear whether the suboptimal folate status amongst Dutch adults actually causes health problems.

United Kingdom

Modelling work carried out by the Food Standards Agency (FSA) in the UK revealed that while a large proportion (22%) of the population currently have folate intakes below the reference nutrient intake (RNI) (Department of Health, 1991), there are also a number of people (106,000; 0.2%) exceeding the tolerable upper intake level (UL) for folic acid (SCF, 2000) through consumption of voluntarily fortified foods and supplements (Figures are based on modelling carried out in September 2006). The Scientific Advisory Committee on Nutrition (SACN) advised that mandatory fortification of flour with folic acid be implemented alongside controls on voluntary fortification (SACN, 2006).

In the UK mandatory fortification is currently on hold. In the meantime discussions have taken place with industry to control the levels of voluntary fortification.

REFERENCES

Data sources by countries:

Belgium:

Food Consumption Survey, 2004.

Denmark:

Lyhne, N., Christensen, T., Groth, M.V., Fagt, S., Biltoft-Jensen, A., Hartkopp, H., Hinsch, H.-J., Mathiessen, J., Møller, A., Saxholt, E., Trolle, 2005. Danskernes kostvaner 2000-2002. Hovedresultater. Danmarks Fødevareforskning, *DFVF publication nr. 11, Søborg*.

Spagner, C., 2007. The contribution of micronutrients from dietary supplements to the total intake of vitamins and minerals in the Danish population. University of Copenhagen and Technical University of Copenhagen, Copenhagen.

France:

AFSSA (Agence française de sécurité sanitaire des aliments), 2002. Observatoire des Consommations Alimentaires. Simulations d'enrichissement de la farine en folates selon plusieurs scénarios et pour différentes catégories de consommateurs. Note technique: OCA/AD/MT/2002-411.

Mennen, L.I., de Courcy, G.P., Guilland, J.C., Ducros, V., Bertrais, S. and Nicolas, J.P., 2002. Homocysteine, cardiovascular disease risk factors, and habitual diet in the French Supplementation with Antioxidant Vitamins and Minerals Study. *Am J Clin Nutr*. 76(6):1279-1289.

Germany:

Mensink, G., Burger, M., Beitz, R., Henschel, Y., Hintzpeter, B., 2002. Was essen wir heute? Ernährungsverhalten in Deutschland. Beiträge zur Gesundheitsberichterstattung. *RKI*, Berlin.

Mensink GBM, Heseker H, Richter A, Stahl A, Vohmann C., 2007. Ernährungsstudie als KiGGS-Modul (ESKIMO). Foschungsbericht. Robert-Koch-Institut und Universität Paderborn.

Max Rubner-Institut, 2008. Nationale Verzehrstudie II. Ergebnisbericht Teil 2. *BFEL*.

Hungary:

Zajkás, G., Bíró, L., Greiner, E., Szórád, I., Agoston, H., Balázs, A., Vitrai, J., Hermann, D., Boros, J., Németh, R., Kéki, Z., Martos, E., 2007. Dietary survey in Hungary, 2003-2004. Micronutrients: vitamins. *Orv Hetil*. 148(34):1593-600.

Ireland:

FSAI (Food Safety Authority of Ireland), 2008. Report of the Implementation Group on Folic Acid Fortification to the Department of Health and Children. <http://www.fsai.ie/publications/reports/folic_acid_implementation.pdf>

Italy:

Sofi, F., Vecchio, S., Giuliani, G., Martinelli, F., Marcucci, R. and Gori, A.M., 2005. Dietary habits, lifestyle and cardiovascular risk factors in a clinically healthy Italian population: the 'Florence' diet is not Mediterranean. *Eur J Clin Nutr.* 584-591.

The Netherlands:

Gezondheidsraad, 2008. Naar een optimaal gebruik van foliumzuur (Towards an optimal use of folic acid). *Publicatienr. 2008/02*. Den Haag.

Norway:

Nordic Council of Ministers. Nordic Nutrition Recommendations 2004. 4th ed. Åhus: *Scanprint*.

Johansson, L. and Solvoll, K., 1997. Norkost II. Statens helsodirektorat [cited 2006 Oct 5], <http://www.shdir.no>

Brevik, A., Vollset, S.E., Tell, G.S., Refsum, H., Ueland, P.M., Loeken, E.B., Drevon, C.A. and Andersen, L.F., 2005. Plasma concentration of folate as a biomarker for the intake of fruit and vegetables: the Hordaland Homocysteine Study. *Am J Clin Nutr*. 81:434-839.

Øverby, N. and Andersen, L.F., 2002. Ungkost-2000. *Sosial- og helsedepartementet*. Oslo.

Grimstad, A.L., 2007. Folic acid fortification of foods. Estimated intake of folate from different fortification scenarios in Norwegian population groups. University of Oslo, Department of Nutrition, Faculty of Medicine. Master thesis.

Sweden:

Guelpen, B. van, Hultdin J., Johansson, I., Stegmayr, B., Hallmans, G. and Nilsson, T.K., 2005. Folate, vitamin B12, and risk of ischemic and hemorrhagic stroke: a prospective, nested case-referent study of plasma concentrations and dietary intake. *Stroke*. 36:1426-1431.

Switzerland:

Camenzind-Frey, E., Sutter-Leuzinger, A., Schmid, A. and Sieber, R, 2005. Beurteilung des Verbrauchs und angenäherten Verzehrs an Nahrungsenergie und Nährstoffen. In: Eichholzer M, Camenzind-Frey E, Matzke A, et al (eds): Fifth Swiss Nutrition Report. *Swiss Federal Office of Public Health*, Bern. 25–36.

Jacob, S., 1995. Ernährungsgewohnheiten und Nährstoffstatus von jungen Frauen in Zürich. Dissertation Nr. 11228, *ETH* Zürich.

Jacob, S., 2001. Wie ernähren sich die potenziell Schwangere und die Schwangere, *Monatsschr Kinderheilk.* (Supp, 1) 149: S7-S16.

United Kingdom:

Department of Health. *Dietary Reference Values for Food, Energy and Nutrients in the United Kingdom. (Report on Health and Social Subjects, No. 41).* London: HSMO, 1991.

Gregory J., Collins D.L., Davies P.S.W., Hughes J.M. & Clarke P.C., 1995. National Diet and Nutrition Survey: children aged 1½ to 4½ years. Volume 1: Report of the diet and nutrition survey. London: *HMSO*.

Gregory J., Lowe S., Bates C.J., Prentice A., Jackson L.V., Smithers G., Wenlock R. & Farron M., 2000. National Diet and Nutrition Survey: young people aged 4 to 18 years. Volume 1: Report of the diet and nutrition survey. London: *TSO*.

Henderson L., Irving K., Gregory J., Bates C.J., Prentice A., Perks J., Swan G. & Farron M., 2003. National Diet and Nutrition Survey: adults aged 19 to 64 years. Volume 3: Vitamin and mineral intake and urinary analytes. London: *TSO*.

Finch S., Doyle W., Lowe C., Bates C.J., Prentice A., Smithers G., Clarke P.C., 1998. National Diet and Nutrition Survey: people aged 65 years and over. Volume 1: Report of the diet and nutrition survey. London: *TSO*.

SACN (Scientific Advisory Committee on Nutrition), Folate and Disease Prevention. London: TSO, 2006.

SCF (European Scientific Committee on Food). Opinion of the Scientific Committee on Food on the Tolerable Upper Intake Level of Folate 2000. <http://ec.europa.eu/comm/food/fs/sc/scf/out80e_en.pdf>

Table 6: Mean or median folate intake of children in selected European countries (µg/day)

Country	Study population	Mean intake, (range or SD) or Median (P5; P95) [µg/day]	Dietary and supplement intake [µg/day]	Details
Denmark*	Children, 4-10 y (n=783)	251 (146, 418)	292 (181, 464)	2000-2004, 7-day food record (consecutive days): precoded, semi-closed questionnaire, nationwide
Germany	6-11 years:			
	Male	Median: 203 (109,0; 495,5)	Median: 203 (109,0; 495,5)	The dietary behaviour of 6–17-year-olds was assessed from January to December 2006 as part of the German Health Interview and Examination Survey for Children and Adolescents (KiGGS)
	Female	Median: 190,1 (100,8; 365,8)	Median: 190,1 (100,8; 365,8)	
	12-17 years:			
	Male	Median: 317 (142,9; 791,0)	Median: 320,2 (148,4; 929,2)	
	Female	Median: 274,8 (124,3; 693,3)	Median: 285,9 (128,5; 730,7)	

Country	Study population	Mean intake, (range or SD) or Median (P5; P95) [µg/day]	Dietary and supplement intake [µg/day]	Details
Netherlands	Baby, 9 months	123 (26)		Excludes folic acid from supplements, 1997/1998 Dutch National Food Consumption Survey: 1+y, 2002: babies and toddlers. Additional folic acid intake from formula milk is 59µg folate equivalents for 9 month old babies, 36µg for 12 month old babies and 4µg for 18 month old toddlers, Health Council 2008.
	Baby, 12 months	126 (23)		
	Toddler, 18 months	114 (26)		
	Child, 1-3 y	113 (33)		
	Child, 4-8 y	129 (35)		
	Child, 9-13 y	158 (39)		
	Child, 14-18 y	182 (46)		
	Children, 2-3 y (n=640)	126		2006 Dutch National Food Consumption Survey
	Children, 4-6 y (n=639)	134		
Norway	Children, 9 y (n=810)	196		2000-2001, Ungkost, n=1005 (Øverby and Anderson,

Country	Study population	Mean intake, (range or SD) or Median (P5; P95) [µg/day]	Dietary and supplement intake [µg/day]	Details
	Children, 13 y (n=1005)	205		2002)
United Kingdom	Boys, 1½ - 4½ y	132	133	NDNS 1½ to 4½ years. Fieldwork carried out 1992/3. (Gregory J et al., 1995)
	Girls, 1½ - 4½ y	129	130	
	Boys, 4-6 y	191	192	NDNS 4-18 years (split by age groups). Fieldwork carried out 1997 (Gregory J.,2000)
	Girls, 4-6 y	169	171	
	Boys, 11-14 y	245	247	
	Girls, 11-14 y	205	210	
	Boys, 15-18 y	305	309	
	Girls, 15-18 y	210	215	

Table 7: Mean or median intake of adults in selected European countries (µg/day)

Country	Study population	Meanintake , µg/day (range or SD)	Dietary and supplement intake	Details
Belgium	Non-pregnant women, 16-70 y (n=150)	217 (59-493)		2004, 7-day food record, German food composition database used
Denmark*	Men, 11-75 y (n=2383)	307 (163, 529)	385 (181, 702)	2000-2004, 7-day food record (consecutive days): precoded, semi-closed questionnaire, nationwide
	Women, 11-75 y (n=2684)	279 (148, 507)	390 (167, 707)	
	Women, 18-49 y (n=825)	295 (163, 495)	478 (256, 735)	
	Women, 18-49 y (n=671)	267 (139, 506)		
France	All (n=1474)	270.8 (99.9)		INCA1 data (OCA 2002)
	Men (n=672)	296.6 (102.6)		
	Men, 15-24 y (n=114)	277.3 (108.1)		
	Men, 25-44 y (n=263)	282.1 (86.4)		
	Men, 45-64 y (n=183)	323.2 (104.6)		

Country	Study population	Meanintake , µg/day (range or SD)	Dietary and supplement intake	Details
	Men, ≥ 65 y (n=112)	306.9 (117.3)		
	Women (n=802)	249.3 (92.1)		
	Women, 15-24 y (n=140)	218.7 (77.6)		
	Women, 25-44 y (n=323)	245.5 (90.1)		
	Women, 45-64 y (n=206)	268.8 (96.4)		
	Women, 65 y (n=133)	260.3 (94.8)		
	Women consuming folic acid supplement (< 4 mg) (n=42)	267.7 (100.4)		
	Men, 45-60 y (n=306)	360 (88)		SUVIMAX study data
	Women, 35-60 y (n=310)	268 (91)		
Germany	Men	271 (221, 330)		German Nutrition Survey, 1998
	Women	226 (188, 273)		
	Men regularly taking supplements		338 (267, 492)	Folate equivalents (folate and folic acid), Mensink et al., 2001
	Women regularly taking supplements		290 (220, 431)	

Country	Study population	Meanintake , µg/day (range or SD)	Dietary and supplement intake	Details
Hungary	Men, total (n=473)	151.8 (52.9)		Third Hungarian National Nutritional Survey 2003 (Zajkás et al., 2007)
	Men, 18-34 y (n=136)	162.2 (62.1)		
	Men, 35-59 y (n=199)	153.3 (46.7)		
	Men, ≥ 60 y (n=138)	139.2 (49.1)		
	Women, total (n=706)	131.0 (46.9)		
	Women, 18-34 y (n=176)	132.3 (47.5)		
	Women, 35-59 y (n=295)	135.4 (49.2)		
	Women, ≥ 60 y (n=235)	124.6 (42.9)		

Country	Study population	Meanintake , µg/day (range or SD)	Dietary and supplement intake	Details
Ireland	Women, 18-50 y	292		Mean total folate (natural folate + folic acid). Folic acid in fortified foods and food supplements, 7-day weighed diary recording, 1999. Modelling intakes with fortified foods: women 18-50 y: 90µg/day. Data updated to account for increased voluntary folic acid food fortification of food brands in 2007 (supplement usage not updated so it is likely this is an underestimation) Source: FSAI 2008. Report of the Implementation Group on Folic Acid Food Fortification to the Department of Health and Children
	Men, 50-64 y	371		
	Women, 50-64 y	271		
	Men, 18-35 y (n=253)	339 (135)		The North/South Ireland Food Consumption Survey, 7-days food dairy (O'Brien et al. 2001)
	Women, 18-35 y (n=269)	247 (120)		
	Men, 36-50 y (n=236)	339 (128)		
	Women, 36-50 y (n=286)	267 (141)		
	Men, 51-64 y (n=173)	314 (115)		
	Women, 51-64 y (n=162)	268 (182)		

Country	Study population	Meanintake , µg/day (range or SD)	Dietary and supplement intake	Details
Italy	Adults	213		1994-1996, survey by National Research Institute for Food and Nutrition
	Men, 20-60 y (n=211)	345 (140)		Food frequency questionnaire (Sofi et al., 2005)
	Women, 20-60 y (n=309)	339 (121)		
Netherlands	Men, 19-50 y	216 (61)		Excludes folic acid from supplements, 1997/1998 Dutch National Food Consumption Survey
	Women, 19-50 y	173 (50)		
	Men, 51-65 y	221 (59)		
	Women, 19-65 y	182 (49)		
	Men, > 65 y	202 (61)		
	Women, > 65 y	178 (53)		
Norway	Average Nordic countries	240-340µg/10MJ		Nordic Council of Ministers, 2004
	Adult men	309		Norkost II (Johansson and Solvoll, 1997), nationwide
	Adult women	250		representative sample (n=2672, 1997).

Country	Study population	Meanintake , µg/day (range or SD)	Dietary and supplement intake	Details
	Middle-aged men, 47-49 y (n=1216)	240		n=5533, Western Norway, food frequency questionnaire (Brevik et al., 2005)
	Middle-aged women, 47-49 y (n=1622)	209		
	Old men, 71-74 y	213		
	Old women, 71-74 y	187		
	Pregnant women (n=19711)	272		Women in the Norwegian Mother and Child Cohort Study, 2000-2001 (Grimstad, 2007)
Sweden	Men and women, 30-60 y	242 (201, 299)		Northern Sweden Health and Disease Cohort, 55% men, food frequency questionnaire (Van Guelpen et al., 2005)
Switzerland	Total population	284		2001/2002, approximate intake based on the amount of food available on the market (Camenzind-Frey et al., 2005)
	Single women, 25-35 y	122		1992, city of Zürich (Tönz, 2005)
United Kingdom	Men, 19-64 y	344	359	NDNS 19-64 years. Fieldwork carried out 2000/1 (Henderson L et al., 2003).
	Women, 19-64 y	251	292	

Country	Study population	Meanintake , µg/day (range or SD)	Dietary and supplement intake	Details
	Men, ≥ 65	270	279	NDNS 65 years and over. Fieldwork carried out 1994/5. Free living subjects only (i.e. not those in institutions) (Finch S et al., 1998)
	Women, ≥ 65 y	207	220	

* Data for Denmark and Germany are given as median intakes (P5, P95).

BIOMARKERS OF FOLATE STATUS

There are a number of biomarkers that are used to determine and monitor folate status. These include red blood cell folate and serum or plasma folate. Related biomarkers that are published in the literature also include plasma or serum concentrations of 5,10-methylenetetrahydrofolate reductase 677C→T genotype (MTHFR 677C-T), homocysteine, vitamin B12, and methylmalonic acid (MMA). Plasma homocysteine is a sensitive marker of low vitamin B12 and folate status. It is also recognised as a risk factor for cardiovascular disease. Studies on folate status are detailed in Table 8.

The concentration of folate in red blood cells gives an indication of tissue stores of folate and can thus be used to determine the intake status over a longer period (2-3 months retrospectively). Red cell folate values above 150µg/l (340nmol/l) are an indication of sufficiency. Serum folate levels below 7nmol/l (3µg/l) are considered to be indicative of inadequate folate status (IOM, 1998). Plasma homocysteine concentrations below 16µmol/l are often used to define normal levels (IOM, 1998)

Red cell folate concentrations were available for Germany, Denmark, France, the Netherlands, Switzerland and UK. Mean serum or plasma folate concentrations range from 7.2 to 25.8nmol/l, (16.5 to 19.3nmol/l for women and 8.1 to 20.8nmol/l for men). The available data show that mean serum or plasma concentrations were not below 7nmol/lfor the population groups studied.

Mean values for vitamin B12 concentrations in blood were in the normal range (150-800 pmol/l) and varied between 226 and 422 pmol/l. Data were only available for Germany, Ireland, Italy, the Netherlands and Sweden.

Plasma homocysteine concentrations ranged from 8.1 to 14.6µmol/l.

Folate status is dependent on the type of assay used and the different cut-off and ranges for defining normal or deficient levels. Therefore, direct comparisons of population mean levels between different European countries are difficult. Furthermore, not all biomarkes have been surveyed in all countries.

REFERENCES

By countries:

Denmark:

Rasmussen, L.B, Ovesen, L., Bülow, I., Knudsen, N., Laurberg, P. and Perrild, H., 2000. Folate intake, lifestyle factors, and homocysteine concentrations in younger and older women. *Am J Clin Nutr.* 72:1156-1163.

France:

Hercberg, S., Preziosi, P., Galan, P., Devanlay, M., Keller, H., Bourgeois, C., de Courcy, G.P. and Cherouvrier, F., 1994. Vitamin status of a healthy French population: dietary intakes and biochemical markers. *Int J Vitam Nutr Res*. 64(3):220-232.

Mennen, L.I., de Courcy, G.P., Guilland, J.C., Ducros, V., Bertrais, S., Nicolas, J.P., Maurel, M., Zarebska, M., Favier, A., Franchisseur, C., Hercberg, S. and Galan, P., 2002. Homocysteine, cardiovascular disease risk factors, and habitual diet in the French Supplement with Antioxidant Vitamins and Minerals Study. *Am J Clin Nutr*. 76:1279-1289.

Germany:

Rauh, M., Verwied, S., Knerr, I., Dörr, H.G., Sönnichsen, A. and Koletzko, B., 2001. Homocysteine concentrations in a German cohort of 500 individuals: reference ranges and determinants of plasma levels in healthy children and their parents. *Amino Acids.* 20:409-418.

Thamm, M., 2001. Folsäureversorgung von Frauen im gebärfähigen Alter. *Berliner Ärzte*. 8:21-24.

Ireland:

Brown, K.S., Kluijtmans L.A., Young, I.S., Woodside, J., Yarnell, J.W., McMaster, D., Murray, L., Evans, A.E., Boreham, C.A., McNulty, H., Strain, J.J., Mitchell, L.E. and Whitehead, A.S., 2003. Genetic evidence that nitric oxide modulates homocysteine: the NOS53 894 TT genotype is a risk factor for hyperhomocystenemia. *Arterioscler Thromb Vasc Biol.* 23:1014-1020.

FSAI (Food Safety Authority of Ireland), 2008. Report of the Implementation Group on Folic Acid Fortification to the Department of Health and Children. <http://www.fsai.ie/publications/reports/folic_acid_implementation.pdf>

Italy:

Cafolla, A., Dragoni, F., Girelli, G., Tosti, M.E., Costyante, A., Pastorell,i D., Bedogni, G. and Scott, S., 2000. Folate status in Italian blood donors: relation to gender and smoking. *Haematologica.* 85:694-698.

Sofi, F., Vecchio, S., Giuliani, G., Martinelli, F., Marcucci, R., Gori, A.M., Fedi, S., Casini, A., Surrenti, C., Abbate, R. and Gensini, G.F., 2005. Dietary habits, lifestyle and cardiovascular risk factors in a clinically healthy Italian population: the 'Florence' diet is not Mediterranean. *Eur J Clin Nutr*. 584-591.

Girelli, D., Martinelli, N., Pizzolo, F., Friso, S., Olivieri, O., Stranieri, C., Trabetti, E., Faccini, G., Tinazzi, E., Pignatti, P.F. and Corrocher, R., 2003. The interaction between MTHFR 677 C -> T genotype and folate status is a determinant of coronary atherosclerosis risk. *J Nutr.* 133:1281-1285.

The Netherlands:

Gezondheidsraad, 2008. Naar een optimaal gebruik van foliumzuur (Towards an optimal use of folic acid). Den Haag: *Gezondheidsraad. Publicatie nr. 2008/02*.

Bree, A. de, Verschuren, W.M., Bjorke-Monsen, A.L., van der Put, N.M., Heil, S.G., Trijbels, F.J. and Blom, H.J., 2003. Effect of the methylenetetrahydrofolate reductase 677C→T mutation on the relations among folate intake and plasma folate and homocysteine concentrations in a general population sample. *Am J Clin Nutr*. 77:687-693.

Bree, A. de, Verschuren, W.M., Blom, H.J. and Kromhout, D., 2001. Lifestyle factors and plasma homocysteine concentrations in a general population sample. *Am J Epidemiol*. 154:150-154.

Dhonukshe-Rutten, R.A.M., de Vries, J.H.M., de Bree, A., van der, Put N., van Staveren, W.A. and de Groot, L.C.P.G.M., 2009. Dietary intake and status of folate and vitamin B12 and their association with homocysteine and cardiovascular disease in European populations. *Eur J Clin Nutr*. 63:18-30.

Durga, J., Bots, M.L., Schouten, E.G., Kok, F.J. and Verhoef, P., 2005. Low concentrations of folate, not hyperhomocysteinemia, are associated with carotid intima-media thickness. Atherosclerosis. 1797:285-292.

Eussen, S.J., de Groot, L.C., Joosten, L.W., Bloo, R.J., Clarke, R., Ueland, P.M., Schneede, J, Blom, H.J, Hoefnagels, W.H. and van Staveren, W.A., 2006. Effect of oral vitamin B-12 with or without folic acid on cognitive function in older people with mild vitamin B-12 deficiency: a randomized, placebo-controlled trial. *Am J Clin Nutr*. 84:361-370.

Jansen, E.H.J.M., Ujcic-Voortman, J.K., Uitenbroek, D.G. and Ocké, M.C., 2007. Foliumzuur-, vitamine B12- en homocysteine status van de bevolking van Amsterdam. Bilthoven: *RIVM*, 2007.

Jong, N. de, Chin, A. Paw M., de Groot, L.C., Rutten, R.A., Swinkels, D.W., Kok, F.J and van Staveren W.A., 2001. Nutrient-dense foods and exercise in frail elderly: effects on B vitamins, homocysteine, methylmalonic acid, and neurophysiological functioning. *Am J Clin Nutr.* 73:338-346.

Melse-Boonstra, A., de Bree, A., Verhoef, P., Bjorke-Monsen, A.L. and Verschuren, W.M., 2002. Dietary monoglutamate and polyglutamate folates are associated with plasma folate concentrations in Dutch men and women aged 20-65 years. *J Nutr*. 132:1307-1312.

Verhoef, P., Kok, F.J., Kruyssen, D.A., Schouten, E.G., Witteman, J.C., Grobbee, D., Ueland, P.M. and Refsum, H. 1997. Plasma total homocysteine, B vitamins, and risk of coronary atherosclerosis. *Arterioscler Thromb Vasc Biol*. 17:989-995.

Norway:

Brevik, A., Vollset, S.E., Tell G.S., Refsum, H., Ueland, P.M., Loeken, E.B., Drevon, C.A. and Andersen, L.F., 2005. Plasma concentration of folate as a biomarker for the intake of fruit and vegetables: the Hordaland Homocysteine Study. *Am J Clin Nutr.* 81:434-439.

Christensen, B., Landaas, S., Stensvold, I., Djurovic, S., Retterstol, L., Ringstad, J., Berg, K. and Thelle, D.S., 1999. Whole blood folate, homocysteine in serum, and risk of first acute myocardial infarction. *Atherosclerosis*. 147:317-326.

Sweden:

Hultdin, J., Van Guelpen, B., Bergh, A., Hallmans, G. and Stattin, P., 2005. Plasma folate, vitamin B12, and homocysteine and prostate cancer risk: a prospective study. *Int J Cancer.* 113:819-824.

Wahlin, A., Backman, L., Hiltdin, J., Adolfsson, R. and Nilsson, L.G., 2002. Reference values for serum levels of vitamin B12 and folic acid in a population-based sample of adults between 35 and 80 years of age. Public Health Nutr. 91:1013-1019.Guelpen B. van, Hultdin J., Johansson I., Stegmayr B., Hallmans G., Nilsson T.K., Weinehall, L., Witthöft, C., Palmqvist, R. and Winkvist, A, 2005. Folate, vitamin B12, and risk of ischemic and hemorrhagic stroke: a prospective, nested case-referent study of plasma concentrations and dietary intake. *Stroke.* 36:1426-1431.

Switzerland:

Hess, S.Y., Zimmermann, M.B., Brogli, S. and Hurrell, R.F., 2001. A national survey of iron and folate status in pregnant women in Switzerland. *Int J Vitam Nutr Res.* 71:268-273.

Jans-Ruggli, S. and Baerlocher, K., 2005. Kenntnisse über Folsäure und Folsäurestatus bei Müttern. Häufige Einnahme von Folsäuresupplementen, aber ungenügende Prävention von Neuralrohrdefekten. In: Fifth Swiss Nutrition Report. Eichholzer M., Camenzind-Frey E., Matzke A., et al. (eds). *Swiss Federal Office of Public Health*, Berne, pp. 71-86.

United Kingdom:

Gregory J., Collins D.L., Davies P.S.W., Hughes J.M. and Clarke P.C., 1995. National Diet and Nutrition Survey: children aged 1½ to 4½ years. Volume 1: Report of the diet and nutrition survey. London: *HMSO.*

Gregory J., Lowe S., Bates C.J., Prentice A., Jackson L.V., Smithers G., Wenlock R. & Farron M., 2000. National Diet and Nutrition Survey: young people aged 4 to 18 years. Volume 1: Report of the diet and nutrition survey. London: *TSO.*

Henderson L., Irving K., Gregory J., Bates C.J., Prentice A., Perks J., Swan G. and Farron M. (2003). National Diet and Nutrition Survey: adults aged 19 to 64 years. Volume 3: Vitamin and mineral intake and urinary analytes. London: *TSO.*

Ruston D., Hoare J., Henderson L., Gregory J., Bates C.J., Prentice A., Birch M., Swan G. and Farron M., 2004. National Diet and Nutrition Survey: adults aged 19-64 years. Volume 4: Nutritional Status (anthropometry and blood analytes), blood pressure and physical activity. London: *TSO.*

Finch S., Doyle W., Lowe C., Bates C.J., Prentice A., Smithers G., Clarke P.C., 1998. National Diet and Nutrition Survey: people aged 65 years and over. Volume 1: Report of the diet and nutrition survey. London: *TSO*.

Bates C.J., Mansoor M.A., van der Pols J., Prentice A., Cole T.J., Finch S., 1997. Plasma total homocysteine in a representative sample of 972 British men and women aged 65 and over. European J Clin Nutr 51: 691-697

Bates C.J., Mansoor M.A., Gregory J., Pentieva K., Prentice A., 2002. Correlates of plasma homocysteine, cysteine and cysteinyl-glycine in respondents in the British National Diet and Nutrition Survey of Young People Aged 4-18 Years, and a comparison with the Survey of People Aged 65 Years and Over. *British J Nutr* 87: 71-79

Other:

Konings, E.J.M., Roomans, H.H.S., Dorant, E., Goldbohm, R.A., Saris, W.H.M. and van den Brandt, P.A., 2001. Folate intake of the Dutch population according to newly established liquid chromatography data for foods. *Am J Clin Nutr.* 73:765-776.

Table 8: Folate status in selected European countries (mean values and SD or range unless otherwise indicated)

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
Denmark	Women, 25-30 y (n=290)	Median (P25, P75): 648 (471, 840)		Median (P25, P75): 7.6 (6.5, 8.9)			1997-1998	Rasmussen et al., 2000
	Women, 60-65 y (n=288)	936 (703, 1151)		9.4 (7.7, 11.1)				
France	Men and women, 6-97 y (n=1039)	358-467	11.3-14.3				Val de Marne transversal study 1988	Hercberg et al., 1994

[14] Folate concentrations given in ng/ml was converted into nmol/l by multiplying with 2.266.
[15] Vitamin B12 concentrations given in pg/ml were converted into pmol/l by multiplying with 0.738.

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Women, 35-40 y (n=232)			8.1 (7.7, 8.4)			French Supplement with Antioxidant Vitamins and Minerals Study.	Mennen et al., 2002
	Women, 40-45 y (n=302)			8.6 (8.3, 8.9)				
	Men, 45-50 y (n=343)			10.4 (10.0, 10.7)				
	Women, 45-50 y (n=290)			8.7 (8.4, 9.1)				
	Men, 50-55 y (n=290)			11.1 (10.7, 11.5)				
	Women, 50-55 y (n=213)			9.3 (9.0, 9.7)				

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Men, 55-60 y (n=281)			11.1 (10.7, 11.5)				
	Women, 55-60 y (n=146)			9.2 (8.7, 9.6)				
Germany	Women of childbearing age (n=1266)	Median (P5, P95): 602.8 (367.1, 1,128.5)	Median (P5, P95): 17.2 (9.5, 29.2)				Subgroup of the sample of the German Nutrition Survey 1998	Thamm, 2001
	Women, 26-50 y (n=172)		16.5 ± 7.7	9.2 ± 3.3		346.6 ± 105.9		Rauh et al., 2001
Ireland	Children, ≤2 y (n=46)		Median 38.82 (4.31-89.51)	Median 6.69 (4.0-15.7)		Median 498 (117-1342)	December 2005-November 2007	

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Children, 3-7 y (n=69)		32.83 (7.25-99.02)	6.64 (4.3-14.8)		461 (150-1064)	Children (0-15y) Plasma folate: 0.9 % deficient; 1.4% possible-deficiency; 34.1% low-adequate; 39.1% adequate-high; 24.5% high	FSAI, 2008
	Children, 8-15 y (n=105)		21.03 (3.17-73.87)	7.86 (3.9-20.3)		336 (104-1128)		
	Women, 16-25 y (n=39)		21.12 (5.89-187.62)	8.64 (6.0-14.2)		233 (91-399)		
	Women, 26-40 y (n=220)		17.47 (3.17-79.31)	9.6 (4.0-37.3)		227 (66-676)	Women (16-65y) Plasma folate: 1.4 % deficient; 5.6% possible-deficiency;	

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Women, 41-65 y (n=237)		19.37 (2.49-86.56)	9.71 (5.0-46.0)		241 (80-596)	51.6% low-adequate; 32.7% adequate-high; 8.7% high	Women (16-65 years) Plasma folate: 1.4 % deficient; 5.6% possible-deficiency; 51.6% low-adequate; 32.7% adequate-high; 8.7% high
	Women, >65 y (n=236)		35.15 (3.85-653.97)	11.56 (5.8-38.2)		252 (72-878)		
	Men, 16-25 y (n=7)		13.6 (7.48-21.07)	10.63 (8.4-14.3)		244 (207-388)		
	Men, 26-40 y (n=93)		12.96 (2.72-71.83)	11.78 (6.8-18.7)		226 (127-493)	Men (16-65 years) Plasma folate: 1.7 % deficient;	

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Men, 41-65 y (n=192)		15.11 (4.31-75.91)	12.5 (7.0-21.0)		231 (93-714)	11.6% possible-deficiency; 63.4% low-adequate; 20.9% adequate-high; 2.4% high	
	Men, >65 y (n=187)		28.33 (4.08-161.11)	12.92 (6.5-48.5)		226 (39-631)		
Italy	Men, 20-60 y (n=99		10.7 (3.6)			434 (174)	Blood donors	Cafolla et al., 2000
	Women, 20-60 y (n=102)		11.3 (3.0)			422 (162)		

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Men, 20-60 y (n=211)		11.8 (5.7)	11.2 (4.3, 70.6)		281 (131)		Sofia et al., 2005
	Women, 20-60 y (n=309)		12.7 (5.9)	9.5 (5.2, 27)		294 (141)		
	Men (n=142) and women (n=80), 57.5 y		13 (12.3, 13.9)	14.7 (14, 15.5)		304 (290, 319)		Girelli et al., 2003
Netherlands	Men and women, 20-65 y (n=2051)		7.4 (2.4, 22.4)	13.6 (7.8, 39.5)		284 (102, 638)	MORGEN study 1993-1996	De Bree et al., 2001; De Bree et al., 2003, Jansen et al., 2007

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Men, 20-65 y (n=1493)			14.6 (6.1) 31% had plasma total Hcys > 15 µmol/l				
	Women, 20-65 y (n=1532)			13.1 (4.6) 20% had plasma total Hcys > 15 µmol/l				
	Men, 20-65 y (n=1275)		8.4 (4.2)				MORGEN study	Melse-Boonstra et al., 2002
	Women, 20-65 y (n=1160)		8.1 (4.0)					

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Men and women, 70+ y (n=195)	6% had RBC folate <305 nmol/l						Eussen et al., 2006
	Frail elderly men and women, 70+ y (n=130)	21% had RBC folate <337 nmol/l						De Jong et al., 2001
	Men (n=74) and women (n=26), 20-65 y			12.5 (5.7)		248 (97)		Verhoef et al., 1997
	Men (n=577) and women (n=224), 50-70 y		12 (10, 15)	13.8 (12.8, 15.4)		289 (242, 264)		Durga et al., 2005

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
Norway	Men, 47-49 y (n=1216)		7.3 (3.9)	10.8 (3.5)			Hordaland Homocysteine Study	Brevik et al., 2005
	Women, 47-49 y (n=1622)		8.5 (6.1)	9.1 (3.4)				
	Men (n=73) and women (n=30, 45-75 y			10.9 (3.0)				Christensen et al., 1999
Sweden	Men, 40, 50, 60 y (n=514)		8.8 (5.3)	13.2 (7.3)		300 (104)	Northern Sweden Health and disease Cohort	Hultdin et al., 2005
	Men and women, 35-80 y (n=961)		17.9 (10.9)			313 (136)		Wahlin et al., 2002

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Men and women, 35, 40 y (n=197)					332 (112)		
	Men and women, 40, 45 y (n=194)					325 (110)		
	Men and women, 45, 60 y (n=194)					310 (104)		
	Men and women, 30, 40, 50, 60 y (n=671)		7.2 (5.0, 9.9)			316 (256, 388)	Northern Sweden health and Disease Cohort, 55% men	Van Guelpen et al., 2005

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
Switzerland	Pregnant women at time of giving birth (n=598)	0.2%: ≤40 µg/l 45.5%: 140-628 µg/l 54.3%: ≥628 µg/l	0.5%: ≤3.5 µg/l 90.5%: 3.5-16 µg/l 9%: ≥16 µg/l				Pregnant women in three hospitals, 97.5% of the women (n=471) took a supplement of folic acid during their pregnancy, most often in form of a multivitamin	Jans-Ruggli and Baerlocher, 2005
	Representative sample of pregnant women (n=300)		7.5 (1.8-63.5) 4% below 2.5					Hess et al., 2001

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
United Kingdom	Boys, 1½ - 4½ y	934 (344)	20.9 (9.7)				NDNS 1½ to 4½ years. Fieldwork carried out 1992/3	Gregory et al., 1995
	Girls, 1½ - 4½ y	894 (326)	21.3 (9.8)					Gregory et al., 1995
	Boys, 4-6 y	736 (200.7)	25.0 (7.25)	5.16 (2.7-9.5)			NDNS 4-18 years (split by age groups). Fieldwork carried out 1997	Gregory et al., 2000
	Girls, 4-6 y	677 (209.3)	25.8 (6.86)	4.79 (2.2-8.1)				Gregory et al., 2000
	Girls), 7–10 y	604 (222.7)	22.0 (8.50)	5.69 (2.7-10.6				Gregory et al., 2000

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Males, 11- 14 y	598 (208.6)	20.8 (7.11)	6.18 (2.9-11.5)				Gregory et al., 2000
	Females 11-14 y	544 (174.4)	19.3 (6.93)	6.40 (3.5-11.8)				Gregory et al., 2000
	Males, 15-18 y	540 (181.0)	17.6 (6.98)	8.54 (4.1-20.1)				Gregory et al., 2000
	15-18 y	500 (171.3)	16.9 (7.51)	7.80 (3.9-14.3)				Gregory et al., 2000
	Males, 19-64 y	694 (288.2)	20.8 (8.55)	11.7 (4.55)			NDNS 19-64 years. Fieldwork carried out 2000/1	Ruston et al., 2004
	Females 19-64 y	685 (293.0)	22.1 (9.46)	10.1 (4.76)			NDNS 19-64 years. Fieldwork carried out 2000/1	Ruston et al., 2004

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Males, ≥ 65 y	495 (275)	15.3 (9.5)	15.67 (7.43)			NDNS 65 years and over. Fieldwork carried out 1994/5. Free living subjects only (i.e. not those in institutions)	Finch et al., 1998; Bates et al., 1997, 2002

Country	Study population	RBC* folate (nmol/l)[14]	Plasma or serum folate (nmol/l)	Plasma homocysteine (µmol/l)	Plasma methyl-malonic acid (µmol/l)	Plasma vitamin B12 (pmol/l)[15]	Remarks	References
	Females, ≥ 65 y	501 (287)	16.5 (10.4)	14.68 (6.29)			NDNS 65 years and over. Fieldwork carried out 1994/5. Free living subjects only (i.e. not those in institutions)	Finch et al., 1998; Bates et al 1997,2002

* RBC folate, red blood cell folate.

CONCLUSIONS

This report from the ESCO Working Group on Analysis of Risks and Benefits of Fortification of Food with Folic Acid presents data on the following: national congenital anomalies registries; estimates on the prevalence of NTDs; recommended dietary folate intakes; practices of voluntary and strategies on mandatory fortification with folic acid; folate intake and biomarkers of folate status.

EUROCAT collects data on NTD-affected pregnancies across Europe and covers about a quarter of all births in Europe.

The prevalence of NTD-affected pregnancies ranges from 4.1 to 19.70 per 10,000 births. Part of this variation can be explained by the different methods of data collection and reporting.

The recommended daily intake of folate for adults ranges from 200 to 400µg folate in European countries. For pregnant women it ranges from 300 to 600µg and for lactating women from 260 to 600µg per day.

In 2000, the SCF set a UL for folic acid of 1mg/day for adults. The UL is based on the risk of "masking" haematological symptoms of vitamin B12 deficiency.

All countries participating in this ESCO WG recommend that women of childbearing age should supplement their diet with 400µg of folic acid per day. However, the timing of the advice and the duration differ slightly across European countries with respect to the target group i.e. women of child bearing age, women planning a pregnancy and timing and duration of supplement intake.

Compliance with the recommendations has not been evaluated in all countries. However, studies which have evaluated the efficacy of policies of recommending women to use folic acid supplements with the aim of reducing NTD-affected pregnancies have shown no or very limited effects in most European countries.

Food can be fortified in the EU under Regulation 1925/2006EC on the addition of vitamins and minerals and of certain other substances to food. Voluntarily fortified foods with folic acid are widely available in all European countries who took part in this project except for Sweden. Foods fortified with folic acid include breakfast cereals, dairy products, juices, flour/bread and fat spreads.

At present no European country has introduced mandatory fortification although the UK is currently considering mandatory fortification.

Available data from studies and surveys indicate that across European countries, the average dietary folate intake ranges from 151 to 345µg/day for men, and from 122 to 339µg/day for women. Average intake of folate from diet and supplements range from 338 to 385µg/day for men and from 220 to 478µg/day for women. The average daily folate intakes were below the European population reference intake (200µg/day) in Hungary (all adult men and women), the Netherlands (women 19-50yrs), Switzerland

(women 25-35yrs) and Norway (women 71-74yrs). However, recommendations do differ across the European countries and different methods were used to assess folate intake. Therefore caution should be applied in interpretation of these values and making comparisons between countries.

Available data from studies and surveys suggest that folate status is adequate for adult men and women. Values for folate status and normal ranges are dependent on the assay method used, which makes it difficult to make direct comparisons between different studies and surveys. Caution should be applied in the interpretation of the results.

APPENDIX 2

EFSA MEETING SUMMARY REPORT

Folic acid: an update on scientific developments

21-22 January 2009, Uppsala, Sweden

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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[16] 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.

[16] 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.