Our medical education is constantly being revised. Faculties around the world tirelessly critique and mould their curricula, with each syllabus revision promising greater empathy, empowerment, information literacy and clinical excellence in students. As these changes take place, we challenge faculties to recognize that our education extends beyond our classrooms and clinical teaching. The development of social networking, communication techniques, and student advocacy are a few of the many skills that are refined outside the standard curriculum. Student initiatives provide a valuable forum for the development of these characteristics in medical students.

NZMSJ is representative of the educational and professional benefits of student initiatives. As the executive of NZMSJ we have been privileged to be part of an exceptionally successful student initiative. From humble beginnings a mere three years ago we have become a recognized biannual journal. NZMSJ is becoming a valuable tool for students to gain experience publishing academic articles. This third edition represents our largest issue yet, and we are confident of continued growth in submission numbers. Alongside the growth of the journal, student authors have had the opportunity for detailed expert review of their work, and reviewers have been exposed to student articles as colleagues instead of markers. We believe that all of the students, staff and authors involved in the NZMSJ enjoy many of the learning outcomes curriculum designers strive for.

The diversity of articles in this edition is remarkable. We have published articles from authors as far away as Nepal and the USA, whilst ensuring that New Zealand students have also been well represented. Original research, interesting reviews and topical opinion pieces have ensured that this issue of the NZMSJ will make quite exceptional reading. Letters to the editor in response to any article are welcome.

We are also pleased to announce the winner and runner up for the NZMSJ writing prize advertised in our last edition. The academic editorial board and advisors were immensely impressed by the standard of submissions. We selected Shannon McCarthy's literature review of Folic Acid supplementation for first prize; an excellent example of clear, relevant, interesting writing for students. The Gale *et al.* group from Wellington received runner up for their research into the sick building syndrome at the Wellington School of Medicine. Brian Grainger's original research into ATP release in cochlear cells was commended as an impressive investigation of a complex topic .

We look forward to receiving further excellent submissions.

The NZMSJ Executive

Folic acid supplementation as a preventative for defects of neural tube closure

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INTRODUCTION

Neural tube defects such as spina bifida and anencephaly are rare birth defects that affect around 0.8 per 1000 total births in New Zealand[1] and are due to failure of the neural tube to close during development. For the past 60 years it has been known that folic acid supplementation is effective in preventing neural tube defects by an unknown mechanism[2]. Since 1993 the New Zealand Ministry of Health has recommended 0.8mg of folic acid per day for first time pregnancies and 5mg per day for high risk (previous NTD pregnancy) women to be taken four weeks prior to conception until the end of the twelfth week of pregnancy[3]. A woman needs to receive daily folic acid before she becomes pregnant otherwise it is too late. This review will examine various intervention studies which have provided strong evidence for the protective role of folic acid supplementation in preventing neural tube defects and the proposed scientific mechanisms of action.

BACKGROUND

Neural tube formation and closure occurs between days 22 and 28 of gestation in humans and involves paired neural folds being brought together at the dorsal midline and adhering to each other with a merging of cells.[4] In mammals, closure is initiated at several places along the anterior-posterior axis until the neural tube forms a closed cylinder which is separated from the surface ectoderm[4]. Neural tube defects (NTDs) occur when various parts of the neural tube fail to close. Spina bifida refers to failed closure of the posterior neural tube. It varies in severity, and is characterised by neural tissue covered by meninges that extrude through the vertebral column[5]. Anencephaly is a lethal malformation that occurs when the anterior neuropore fails to close and the brain remains in contact with the amniotic fluid and degenerates.[5] Epidemiological studies have suggested that environmental and genetic factors have a joint role in causing NTDs[6]. From such studies it has been determined that poor nutrition and folate deficiency in particular puts foetuses most at risk[4]. Folate, or Vitamin B_9, acts as a cofactor for enzymes essential in DNA and RNA synthesis and is also required in the transfer of methyl groups in the amino acid methylation cycle, an essential step in the recycling of homocysteine back to methionine[5].

STUDIES

Several intervention studies have shown that taking folic acid supplements can reduce the occurrence and recurrence of neural tube defects (NTDs). Smithells *et al.* [7] found a significant 85 per cent reduction in the recurrence rate of NTDs in mothers given a multivitamin supplement containing 0.36mg folic acid during the periconceptional period (before conception to early pregnancy), compared with unsupplemented mothers. This trial was controversial however, due to the absence of a placebo controlled double blind approach[8]. In 1988, Mulinare *et al.* [9] found that, as well as multivitamin use reducing the risk of NTDs, it also has a protective effect among women without prior NTD-effected pregnancies.

Milunsky *et al.* [10] found a substantially reduced risk of NTDs among women who took standard doses of folic acid containing multivitamins during the first six weeks of pregnancy, and they estimated that folic acid supplements taken during the first six weeks of pregnancy would prevent the occurrence of NTDs by more than 50 per cent.

The most convincing evidence that folic acid supplementation aids in primary prevention of NTDs and recurrence of NTDs has been provided by three studies. In 1992, a randomised control trial performed by Czeizel and Dudas[11] found that the incidence of a first occurrence of NTDs was reduced among women who took folic acid supplements during the periconceptional period. A placebo controlled study by the MRC Vitamin Research Group (1991) assigned women with previous NTD-affected pregnancies to one of four groups: daily supplementation with 4mg folic acid, 4mg folic acid and other vitamins, other vitamins without folic acid, neither folic acid or vitamins[12]. The trial found that high-dose folic acid supplementation (4.0mg) alone reduced NTD recurrences by 72 per cent and that the addition of other vitamins conferred no extra benefit in averting NTDs[12].

During 1993-1995, the Centre for Disease Control and Prevention in the USA and the People's Republic of China conducted a population based intervention study to the efficacy of periconceptional use of folic acid in preventing NTDs[13]. The study included almost 250,000 women

from northern (high NTD rate) and southern (low NTD rate) China[13]. After finding that periconceptional use of 400 µg folic acid reduced the risk of NTDs by 79 percent for northern China and 41 percent for the southern region they were able to demonstrate that ingestion of 400 µg of folic acid alone per day during the periconceptional period prevents NTDs in areas of both high and low frequency[13].

This strong evidence led the US Public Health Service to recommend 0.4mg per day of folic acid during the periconceptional period as this would reduce the incidence of NTDs by a possible 50 percent[2]. Three approaches to increase folic acid consumption in the US were proposed: improving dietary habits, fortifying foods with folic acid, and recommending the use of dietary supplements containing folic acid[14]. Some population groups were concerned that fortification would expose all members of society to folic acid in larger amounts than was usual[1]. Proposed adverse effects included toxic effects from folic acid ingestion; the masking of vitamin B_{12} deficiency; a relationship between increased folic acid intake and multiple births; and adverse effects with zinc, anticonvulsants, and oral contraceptives[1]. There is no conclusive evidence that folic acid intake produces any of these[1]. After mandatory fortification, a primary mechanism for improving the folate status of the majority of women in the population, of cereal grain products with 140 mcg folic acid per 100g flour the reported prevalence of annual NTD affected pregnancies decreased by 27 percent[14]. The US still recommends that women take folic acid supplements as it is an effective method of ensuring they get the full 400 mcg as diet alone does not provide sufficient amounts[14].

The New Zealand Ministry of Health responded to these various findings by recommending 0.8 mg per day four weeks prior to conception until the end of the twelfth week of pregnancy[3]. For high risk (previous NTD pregnancies) they recommend 5 mg per day[3]. There are many issues regarding supplementation including the fact that most women are still not aware that folic acid prevents NTDs, the use of folic acid remains low, and women who do take supplements are usually not taking them periconceptionally[1]. Many pregnancies are unplanned and NTDs can form before women are even aware they are pregnant.

Current research shows that there are no known toxic effects from folic acid ingestion either by diet, supplementation or fortification. In fact, folic acid has been shown to prevent other birth defects, cancers, and cardiovascular disease[1]. Since 1996 voluntary fortification of certain food products such as breads, breakfast cereals, and food drinks with folic acid has been permitted in New Zealand[3]. Although mandatory fortification would cost initially, the benefits would be far greater. In New Zealand, NTDs are a major component of fetal and infant death and morbidity[1]. They also contribute to the overall 'burden of diease' in terms of medical treatment and other associated costs with spina bifida having the fourth highest lifetime cost from birth defects[1].

PROPOSED MECHANISMS OF ACTION

Although the evidence for scientific mechanisms on how folic acid action prevents NTDs is much less convincing and the underlying causes of NTDs are still unknown, a few explanations have been proposed. A women's risk of having a child with NTD has been found to be associated with early pregnancy red cell folate levels in a continuous dose-response relationship[15]. Their finding that the supply of folate to the embryo may be diminished even with seemingly normal maternal folate levels led Daly *et al.*[15] to believe that susceptibility of NTDs may come from an inborn error of folate metabolism rather than a dietary deficiency[5].

A study in 1996 by Rosenquist *et al.*[16] found that homocysteine, a teratogenic agent at high concentrations, was found in increased levels in folate depletion. Treating avian embryos with folate supplementation prevented this rise in homocysteine to teratogenic levels therefore the primary effect of folic acid supplementation may be to protect against NTDs by reducing the levels of maternal serum homocysteine[16]. Hook and Czeizel[17] found a statistically significant association between folic acid supplementation and an increased prevalence of recognised spontaneous abortion causing embryonic and foetal death. They proposed folic acid as a strong candidate to be a terathanasic agent which may diminish the rate of NTDs by selectively inducing abortion of affected conceptuses.

Five to fifteen per cent of the general population is homozygous for a thermolabile variant of 5,10 methlenetetrahydrofolate reductase (MTHFR), a folate-related enzyme which is positively associated with the risk of NTDs[18]. Molloy *et al.*[18] found that plasma and red cell folate levels were significantly reduced in women homozygous for this variant causing an additional requirement for folate. This may be the first genetic risk factor for NTDs to be identified and is estimated to account for 13 percent of all cases[5].

Antony & Hansen[19] hypothesised that folate receptors were critical for neural tube development because when they were eliminated in mice a high percentage of folate-responsive NTDs were reported. They proposed that a reduction in folate receptor expression would reduce fetal folate delivery causing decreased proliferation of neural tube cells in early pregnancy leading to NTDs. Saitsu *et al.*[20] agreed with these findings that folate receptors have an important role in embryogenesis. One such receptor is folate-binding protein 1(FBP1), a membrane protein that binds folate with a high affinity and incorporates it into cells by endocytosis[20]. Saitsu *et al.* completed a study on FBP1 and discovered it is mainly localised to the most dorsal regions of the neural folds and is expressed prior to dorsal closure of the anterior tube. They also found that folate transport through the human placenta and mouse yolk sac was mediated by FBP1 at the stage of neural tube closure. Therefore a defect in FBP1 expression could explain how a deficiency in folate leads to NTDs.

Genetic variants of folate-pathway enzymes or folate receptors do not account for the 70 per cent reduction in NTDs associated with folic acid supplementation. However, Rothenburg *et al.*[21] identified autoantibodies against the folate receptor membrane protein in serum from women who had a previous pregnancy complicated by an NTD. Folate responsive NTDs may be due to the autoantibody binding to folate receptors on placental membranes and cells with a high affinity and inhibiting the binding and uptake of folate[21]. Folic acid may bypass this autoantibody-mediated blocking of cellular folate uptake as it is reduced and methylated in vivo and transported into cells by another membrane protein, reduced-folate carrier (RFC)[21]. Folic acid also has a high affinity for the folate receptor so may displace an autoantibody with a lower affinity and explain why folic acid supplements are more readily absorbed than folate naturally contained in food[21]. This would provide a mechanism for how folic acid supplementation can dramatically decrease the recurrence of NTDs.

In New Zealand, NTDs are a major component of fetal and infant death and morbidity.

CONCLUSION

There is strong evidence from epidemiological studies that folic acid supplementation can prevent defects of neural tube closure. This has led the NZ Ministry of Health to recommend a 0.8 mg daily intake for all women before conception and during early pregnancy, and higher doses to prevent the recurrence of NTDs in women with a previously NTD-effected pregnancy. Although it is still unclear as to how folic acid acts to prevent NTDs such as spina bifida and anencephaly, several explanations have been proposed which implicate various other factors in depriving the highly proliferative neural tube cells of folate during the period when they need it most, neural tube closure.

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