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The Prevention of Neural Tube Defects with Folic Acid

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PRESENTATION

Neural Tube Defects (NTD's) include a large number of congenital malformations produced when the open neural tube present at very early stages in the development of the human embryo fails to close on or before the first month post conception. NTD's are considered to be one of the most common forms of congenital malformations, with varying degrees of incidence depending on genetic and environmental conditions.

Research shows folic acid plays an important role against NTD's. Numerous studies prove this B vitamin has a protective effect for the recurrence of NTD's and others indicate folate deficiency is linked to the occurrence of NTD's. In light of this, in 1992 the U.S. Public Health Services issued the recommendation that all women of child-bearing age should consume at least 0.4 mg (400 micrograms) of folic acid daily.

In order to increase the public awareness of the protective role of folic acid against neural tube defects, and working within the framework of health promotion, the Centers for Disease Control and Prevention (CDC) asked by the Pan American Health Organization (PAHO) elaborated this document for its dissemination in the Region of the Americas.

PAHO and CDC are pleased to publish this document.

I. Introduction

The purpose of this document is to review what is known about the prevention of neural tube defects (NTDs) with folic acid. The review includes an overview of the morphogensis and epidemiology of neural tube defects; the evidence for an association between folic acid and NTDs; the proposed mechanisms for the prevention of NTDs with folic acid; and the options available for increasing folic acid consumption in communities.

II. Neural Tube Defects

A. Clinical

From almost every perspective (phenotypic [1], epidemiological [2], etiological [3] and clinical [4,5]) neural tube defects (NTDs) include a broad scope of congenital malformations [4,5]. They are produced when the open neural tube present at very early stages in the development of the human embryo fails to close on or before the 28^{th} day of development [1]. Neural tube defects can range from a very uncomplicated and often subclinical small opening in the posterior vertebral canal to a lack of closure of the whole neural tube producing the most severe type of defect, craniorachischisis. The severity of NTDs varies making almost every case clinically unique [3].

Close to half of the cases of NTDs are anencephaly, which implies an absence or deficiency of a major portion of the cranial vault. Infants with anencephaly are stillborn or live a very short time. The other half of NTDs include defects occurring along the length of the neural tube. From the upper part of the neural tube (where the cranial bones develop giving origin to defects such as encephaloceles) to the spinal canal (where spina bifida occurs, ranging from mild defects affecting one vertebrae to extremely large open spines, with all or almost all of the vertebral arches open).

Although a lot of information exists on the classification of neural tube defects and their clinical consequences, little is known about why the neural tube does not close. Most of the genes driving the development of human embryonic structures are unknown. This is true for the development of the neural tube as well.

B. Diagnosis of NTDs

The occurrence of spina bifida will usually result in a high degree of impairment and disability for the affected individual. Because of the primary defect, there is a sequence of events that occur before birth that will result in severe impairment of the normal scope of functions that are dependent on the central nervous system. Even in the mildest cases and with very early surgical repair of the primary defect, severe disabilities can be expected [11].

Neural tube defects can be diagnosed prenatally using high-resolution ultrasound. Other techniques can be used to screen for NTDs including maternal serum alpha-

fetoprotein (MSAFP) testing, routine ultrasounds, and amniotic AFP measurements when amniocentesis is done for other purposes [12,15,16,17].

Higher than expected levels of alpha-fetoprotein in maternal serum and/or in amniotic fluid may indicate an open fetal defect. If amniocentesis is performed, acetylcholinesterase and AFP are measured and the alterations may indicate that the fetus is most likely affected by an NTD. When AFP levels are high or when birth defects are suspected based on a routine ultrasound, a high-resolution ultrasound is required to make a definitive diagnosis of an NTD.

C. Epidemiology

Neural tube defects are considered to have one of the highest incidence rates of all the congenital malformations. NTDs rates vary from one population to another and have been found to vary by geography, time, and selected maternal demographic characteristics. In the United States, reported birth prevalence rates vary from 4 to 10 per 10,000 live births [35]. Researchers in countries such as Ireland, The United Kingdom, China, Hungary, and Mexico have reported higher rates. Occasionally rates as high as 1% have been reported. [36]. Variation in reported rates may be due to true variation among different populations, differences in surveillance methodology, and the impact of prenatal diagnosis and elective terminations of NTD-affected pregnancies (many birth defect surveillance systems ascertain defects only among live-born infants).

NTDs are usually sporadic events, rarely resulting from chromosomal abnormalities or familial traits. However, the recurrence risks for a second NTD-affected pregnancy are higher than the population or occurrence risk [37]. Estimates for recurrence risk range from 3% to 5% depending on the population risk [38, 74].

Anencephaly seems to occur more often among females (female to male ratio of 2.3:1 for anencephaly in whites, Metropolitan Atlanta Congenital Defects Program, 1968-1996), whereas spina bifida rates have shown only a slight female predominance. Some studies have shown little variation by gender when taking account of race and the presence of associated defects [18,39.]. In the United States rates of NTDs have historically been lower among blacks compared to whites and higher among Hispanics compared to whites [40]. In other countries, rates for NTDs have been reported to be higher among some migrants groups (reflecting comparable rates with the country of origin) and for selected population groups such as Sikhs [41] and the Welsh and Irish [42].

D. NTDs in Latin America

Information on neural tube defects in Latin America is scant. There are no population-based birth defect registries but there is a collaborative hospital- based registry, the Latin America Collaborative Study of Congenital Malformations (ECLAMC). ECLAMC was started in 1967 and includes hospitals distributed over all South American countries. It is part of the International Clearinghouse for Birth Defects Monitoring Systems. The registry covers 215,000 births per year, which is less than 1% of all births in the region.

Also, the Mexican Registry and Epidemiological Surveillance of External Congenital Malformations (RYVEMCE) is a hospital-based registry that covers about 3.5% of all births in Mexico.

Below are the ECLAMC and the Mexican Registry and Epidemiological Surveillance of External Congenital Malformations (RYVEMCE) rates of NTDs published in the 1997 Clearinghouse report for 5-year time periods and the most recent year of complete data [32]. The rates are per 10,000 live births.

ECLAMC rates:

	1974-79	1980-84	1985-89	1990-94	1995
• Anencephaly	3.47	6.73	6.29	7.18	7.63
• Spina bifida	5.82	6.54	6.99	7.62	9.39
• Encephalocele	1.26	2.28	1.53	2.16	1.55

RYVEMCE rates:

	1974-79	1980-84	1985-89	1990-94	1995
• Anencephaly		18.23	19.99	16.51	16.42
• Spina bifida		14.78	18.57	17.00	8.94
• Encephalocele		3.29	3.18	2.28	3.12

In Colombia in 1993, a study based on 10,000 live births at a single hospital found a rate of NTDs of 13 per 10,000 [43].

E. Risk factors for NTDs

Existing research suggests that both genetic and environmental risk factors are associated with the occurrence of NTDs [17,18]. From the genetic point of view, some NTDs have been associated with chromosomal abnormalities (for example, trisomy 18) and some single gene defects (for example, Meckel-Gruber syndrome) [44].

It is believed, however, that the most common types of NTDs are multifactorial in origin. This occurs when there is a genetic predisposition to the malformation, which is triggered by an environmental risk factor. To date, a number of risk factors have been associated with NTDs including: 1) socioeconomic status [45]; 2) lead in drinking water [46]; 3) influenza [47]; 4) maternal heat exposure [48]; 5) parental occupation [49]; 6) maternal obesity [50]; and 7) maternal nutritional status [51]. One nutritional factor, folic acid, has been shown to play a very powerful role in the occurrence of neural tube defects.

III. Folic Acid and NTDs

In recent years there has been numerous studies supporting the protective effect of maternal use of folic acid on the occurrence of NTDs.

Dr. Lucy Wills first described folate in 1930 as a factor that cured nutritional deficiency anemia in pregnant women [52]. Since that time, much information on folates (includes naturally occurring folate compounds and synthetic folic acid in vitamins and fortified foods) has accumulated and has been related to congenital

malformations. About 30 years ago, folate levels among women of childbearing age became a new issue in the prevention of fetal and neonatal malformations [10]. Based on the research, CDC estimates that if all women capable of becoming pregnant consumed 400 mg of folic acid daily, 50-70% of all cases of spina bifida and anencephaly could be prevented [6].

A. Nutrition and physiology

Folate, a water-soluble B-complex vitamin, is considered an essential nutrient. This means that humans are not able to synthesize folate. There are folate-producing bacteria in the human intestine, however, the amounts that they are able to synthesize do not contribute in a significant way to the daily requirements for folate [29]. The only source for folates is the diet. Those natural forms share with the folic acid a pteridine ring, a PABA (para amniobenzoic acid) and a "tail" of one to six molecules of glutamic acid. The more molecules of the glutamic acid the folate molecule has, the less its bioavailability. Most folates have several molecules of glutamic acid that have to be converted to the monoglutamate form to be absorbable in the intestine. Since synthetic folic acid is already in the monoglutamate form, it is more bioavailable than natural forms. This for of folate is reduced by 20 to 30% by heat and cooking. Natural forms of folate are also susceptible to destruction by cooking or processing resulting in lower levels of food folate ingested [53].

Folates have two main physiological effects: it is a cofactor for the enzymes that synthesize DNA and RNA and is required for the conversion of homocysteine to methionine [29]. During early fetal development nucleic acid and protein synthesis are at their peak and maternal folate requirements increase rapidly during this time. When folate is insufficient, nucleic acid is inhibited and cells are unable to manufacture enough DNA for mitosis. In addition, inhibition of the methylation cycle results in an inability to methylate proteins, lipids and myelin [54].

B. Proposed mechanisms for NTD prevention

The underlying mechanism by which folic acid prevents the development of NTDs is unknown. One theory is that susceptibility to NTDs is not primarily from a dietary deficiency of folate but from an inborn error of folate metabolism. Under this scenario, the fetus may have a diminished supply of folate even when the maternal folate levels appear adequate. When folate metabolism is abnormal, homocycteine accumulates. When the metabolism of homcyteine is inadequate, any of three enzyme activities can be altered: 1) cystathione synthase, 2) methionine synthase, or 3) 5,10 methylene tetrahydrofolate reductase [54]. A number of studies have investigated the relationship between these enzymes and NTDs. [63-72]. Results from these studies are promising and support the idea that NTDs are multifactorial and are due to a genetic susceptibility that can be triggered by a number of different independent risk factors.

Another though less widely accepted theory is that folic acid may reduce the prevalence of NTDs by inducing selectively the abortion of affected fetuses. [33]. A biological explanation for this theory is unknown.

C. Scientific evidence for the prevention of NTDs with folic acid

Scientific interest in folic acid began in 1964 when Hibbard published a paper reporting an association between malformations (non NTDs) and folate deficiency [9]. Interest about folic acid among the scientific community increased but it wasn't until 1976 when Smithells was able to link folate (and some other vitamins) deficiency with the occurrence of NTDs [10]. In 1980 he published a non-randomized trial of multivitamin supplementation to women who had previously given birth to one or more NTD affected babies. He showed a 5% recurrence rate among the non-supplemented group against a 0.6% recurrence rate for the supplemented group [75].

Laurence, one of the most prolific investigators of NTDs, suggested in 1980 that women with adequate diets would have lower recurrence rates of NTDs. In 1981 he published a clinical trial that showed a 60% reduction risk (although with a large non-significant confidence interval) of recurrent NTDs among women who took folic acid [11,12].

Four more studies (observational) were published during the 1980's [6,19,20,21]; all reporting protective effects of the use of vitamin supplements containing folic acid during the periconceptional period (at least one-month before pregnancy through the first trimester). All the studies showed a protective effect for the recurrence of NTDs among the group that took folic acid compared with the control or unexposed group. All the results but one [20] were statistically significant. The reason for this null effect is unknown but small sample, underascertainment of cases and misclassification are some possible explanations.

In the early 1990's, two more observational studies, one non-randomized clinical study, and two randomized clinical trials were reported [22,23,24,25,26]. All of them, again, showed an NTD-protective effect of folate intake and/or folic acid supplementation among the exposed group compared to controls ~~for NTDs~~. All but one of the studies were statistically significant.

In 1991 CDC published a review of the evidence for the prevention of recurrent NTD affected pregnancies in families and recommended 4 mg of folic acid for women who previously had an infant or fetus with an NTD [8]. The following year, the U.S. Public Health Service issued the recommendation that all women capable of becoming pregnant should consume 0.4 mg (400 micrograms) of folic acid per day [7].

A proportion of NTD cases will not be preventable with the administration of folic acid [24, 27]. These finding show once more the heterogeneity of the condition. Of more concern is the suggestion that some ethnic groups may respond less to the protective effects of folic acid. One study published in 1995 found that the risk reduction for NTDs associated with folic acid was less marked is this the best way to describe result? for Hispanics than for non-Hispanic whites or blacks [24]. These findings have not yet been replicated elsewhere.

D. Dosage for occurrence and recurrence prevention.

Although the recommendation for the timing for the administration of folic acid to prevent NTDs appears to be clear in general (4 weeks before conception to the end of the first trimester), the optimal dose of supplementation is less certain [29]. The administration of folic acid has to be done before pregnancy and during the first weeks of gestation, until the stage of closure of the neural tube is completed (fourth week of development).

Evidence for the dosage of folic acid currently recommended in the United States comes from the Medical Research Council trial [25], Smithell's nonrandomized study [73] and studies by Werler [23] and Shaw [24].
The Medical Research Council study [25] found a 71% reduction rate in the recurrence of NTDs with daily 4mg of folic acid alone (i.e., without other vitamins). In Europe, a randomized clinical trial evaluated the effects of 0.8mg folic acid contained in a daily multivitamin and found no NTDs in the group taking the supplements [26]. Smithell's non-randomized trial found a protective effect with the administration of 0.36mg/day of periconceptional folic acid contained in a multivitamin[10].

An unpublished report of the results of an evaluation of a non-randomized community intervention in 2 areas of China provides further evidence to support the recommendation for women to get 400 mcg of folic acid daily in the periconceptional period. The intervention regimen was pills containing 400 mcg of synthetic folic acid, without other vitamins. In the Northern intervention area, the rate of NTDs in pregnancies of women who started taking folic acid supplements before their pregnancies began was 10 per 10,000 compared to 48 per 10,000 in the pregnancies of women who did not use the supplements (80% reduction). In the Southern area, the background rate of NTDs was much lower than in the Northern area. In the South, women who started taking 400 mcg folic acid supplements before their pregnancies began had a 40% lower risk of having an NTD-affected pregnancy, compared to women who did not use the supplements (6 per 10,000 compared to 10 per 10,000).

Since 1992, the U.S Public Health Service has recommended 0.4 mg of folic acid daily to prevent the occurrence of NTDs.
CDC in 1991 recommended 4 mg of folic acid daily to prevent the recurrence of NTDs. This year, the Institute of Medicine (IOM) reaffirmed the 0.4 mg recommendation in their evaluation of dietary reference intakes [29] .

E. Other Birth Defects Preventive Effects of Folic Acid

The evidence from research to-date suggests that folic acid supplementation should be given to prepregnant women to prevent the occurrence of NTDs. More recent research suggests that folic acid may prevent other birth defects as well such as cleft lip and palate [55,56,57], limb deficiency defects [58], conotruncal defects [59] and urinary tract anomalies[60]. More research is needed to confirm and elucidate the association between folic acid and these other congenital anomalies. Recent research in the area of cardiovascular disease has also shown that folic acid supplementation may also decrease the risk for coronary disease in elderly populations [29].

IV. Community interventions to increase folate intake

To increase dietary folic acid among prepregnant women, there are a number of issues to consider. The first problem is that consumption of naturally occurring folates is very difficult to be increase in a way that will produce acceptable levels in the women. As already discussed, the bioavailability of folate is low [28] and very large amounts of folate-rich food will be necessary to increase folate levels to the equivalent of 0.4. mg/day of folic acid.

The second issue is that the target population should be prepregnant women so that body levels of folate are adequate before and after conception. It would be difficult to target only women who are planning a pregnancy because many pregnancies are unplanned. In the U.S. the recommendation for folic acid is targeted at all women who are capable of becoming pregnant.

The main strategies for increasing folate levels among women are dietary modification, folic acid supplementation and food fortification. The efficacy of these approaches is dependent upon many factors (e.g., socioeconomic, health services infrastructure, diet, etc.) which will vary by population and culture.

A. Dietary modification

The estimated amount of folate in the diet of U.S. women is an average of 0.2mg/day. There are many folate-rich foods (e.g. fruits, leafy green vegetables, and grains), but to increase dietary folate to the equivalent of 0.4 mg of folic acid per day, women would have to significantly increase their consumption of these foods. This seems unlikely and it appears that women can only marginally increase their blood folate to adequate levels through dietary sources [28]. Although efforts to increase the consumption of folate-rich foods in several populations have not been very successful, women should be encouraged to eat good diets that include folate-rich foods for healthy pregnancies.

B. Folic acid supplementation

Another option is to supplement women's diet with folic acid tablets or multivitamins containing adequate levels of folic acid. Initially this appears to be an easier means of increasing blood folate levels than significantly changing dietary behaviors. However, studies of vitamin use among women show that vitamins are used consistently by only 30% of women aged 18 to 45 [34]. And some women, such as those who are younger, less educated and have lower incomes, are even less likely to use vitamins. The majority of pregnant women take prenatal vitamins on the advice of their health care professionals but the women don't start taking the vitamin until they discover that they are pregnant. For the prevention of NTDs, this is too late.

A number of campaigns to increase multivitamin consumption among women have been tried in the U.S. These campaigns have involved media events, print and TV and radio ads, health care provider education, distribution of folic acid tablets, school programs, and counseling. Some early results of these campaigns suggest that there is an increased knowledge about folic acid among the women targeted but

the effectiveness of these campaigns for reducing the occurrence of NTDs has not yet been proven.

C. Fortification of the food supply

Fortification of the food supply is another strategy for increasing folate levels among women. The major advantage of fortification is that it can reach a wide population without requiring a change in behavior. Studies have also shown that fortification may be a more cost-effective means of increasing folates than dietary changes or supplementation [61].

The appropriate food items to fortify with folic acid depend on the dietary habits of the population, the logistics of the fortification process, and the chemical relationship between the folic acid and the food product being fortified. In 1996, the U.S. Food and Drug Administration (FDA) selected flour, corn meal, pasta and rice as target foods to be fortified with folic acid beginning in January 1998. The level of fortification chosen was 140-micrograms/100 g of cereal grain product. The recent IOM report sets the *Tolerable Upper Intake Level* for adults (highest level of daily nutrient intake that is likely to pose no risks of adverse health effects to almost all individuals in the general population) for folic acid at 1,000 micrograms/day, exclusive of food folate [29]. At the fortification level of 140 mcg folic acid per 100 g grain, few, if any, persons are expected to consume more that the IOM *Tolerable Upper Intake Level*.

In the U.S., with the current level of fortification, it is estimated that the average woman will consume about 100 micrograms of folic acid daily from fortified cereal grain products. This level of fortification is likely to prevent some but not all NTDs that could be prevented by sufficient maternal folic acid intake. For this reason, it is necessary to promote folic acid supplementation and in some cases dietary modification so that all women will have adequate folate levels. A combination of strategies may be the best way to reach sub groups of any population.

D. Measuring the impact of interventions

Determining whether or not an intervention strategy is effective may involve measuring both intermediate outcomes and the final health outcome. Intermediate outcomes that may provide some evidence for the effectiveness of a particular strategy are changes in women's knowledge and attitudes about folic acid, and changes in their consumption of folate rich food or vitamins with folic acid. Changes in knowledge and attitude can be evaluated through surveys. Surveys or food diaries can measure changes in food or vitamin consumption.

The best method of determining whether women are consuming more folic acid is to measure changes in their blood folate levels. Blood folates have been shown to correlate with folic acid consumption and appear to correlate with NTD rates as well [Daley]. Folate can also be measured in serum, plasma, and urine [29]. The measurement of red cell folates is best for evaluating the long-term folate status of a person. Folates measured in serum, plasma and urine are good for evaluating recent folate intake levels. As the folate that is available to the fetus is the serum folate, serum folate levels may be good indicators of fetal exposure to folates during the critical periods of intrauterine life (such as the time when the neural tube closes).

The obvious measure of the impact of increased folic acid consumption is the reduction in NTD rates. However, in order to measure changes in rates, a baseline measurement must be available and a surveillance system must be in place to count the occurrences of cases over time. To get a reliable estimation of NTD incidence in a population, it is also necessary to count the cases, which are prenatally diagnosed, and electively terminated [62]. The ability to count NTD occurrences reliably varies considerably and may not be possible in some areas where the infrastructure for this type of surveillance has yet to be established.

Regardless of which strategies are used to increase folate levels among women, the intervention should be monitored and evaluated for its effectiveness. Measurement of intermediate outcomes (e.g., knowledge of folic acid's importance or blood folate levels) can be used to monitor progress.

V. Summary

Evidence shows that the administration of synthetic folic acid decreases the rates of NTDs, and may decrease the rates for other congenital defects. There are a number of intervention strategies that can be used to increase blood folate levels among women and ultimately decrease NTD rates. The choice of strategy will depend on the population targeted and the resources available. Monitoring and measuring the impact of the intervention is an important component of any effective strategy.

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