

**Periconception Care  
and  
Primary prevention of neural-tube defects  
and other congenital abnormalities  
by periconceptional folic acid/  
multivitamin supplementation**

Andrew E. Czeizel, M.D., Ph.D., Doct Sci.  
Budapest, Hungary

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# **Concept of Hungarian Periconception Service (HPS) established in 1984**

- Counselling, examinations, and interventions conducted by qualified nurses as part of primary health care.
- Selected couples at risk are referred to specialists in the secondary health care.

# Hungarian periconceptional service (HPS)

Included 33,000 couples between  
1984 and 2009.

# Three stages of HPS

- 1) Reproductive health check-up.  
Preconception screening for reproductive risk factors.
- 2) The 3-month preparation for conception period.  
The beginning of life is the conception.
- 3) Better protection of early pregnancy.  
In general pregnant women visit first prenatal clinics at 6th-10th gestational week

# 1) Reproductive health check-up

- a) Taking family history of prospective mothers and fathers, and obstetric history of females.
- b) Take the medical history and check available medical records of females, e.g., for epilepsy, diabetes.
- c) Vaginal and cervical smear screening for sexually transmitted infections/disorders.
- d) Sperm analysis to detect subfertility and pyosperm.
- e) Psychosexual assessment.
- f) Varicella vaccination in women not previously exposed to varicella.
- g) Blood screening (rubella seronegativity, HIV positivity, in addition carrier screening for cystic fibrosis).

## Rate of preterm birth (%) in pregnant women with vaginal infections/diseases according to treatment in the HPS

Study groups	Treated %	Untreated %
Sexually transmitted diseases	6.1	45.8
Sexually transmitted infections (subclinical)	4.8	30.0
Vaginal candidiasis	4.5	10.3
Subclinical vaginal candidiasis	4.0	9.3

Fetal  
Varicella  
Disease due  
to  
the lack of  
vaccination



## 2) The 3-month preparation for conception period

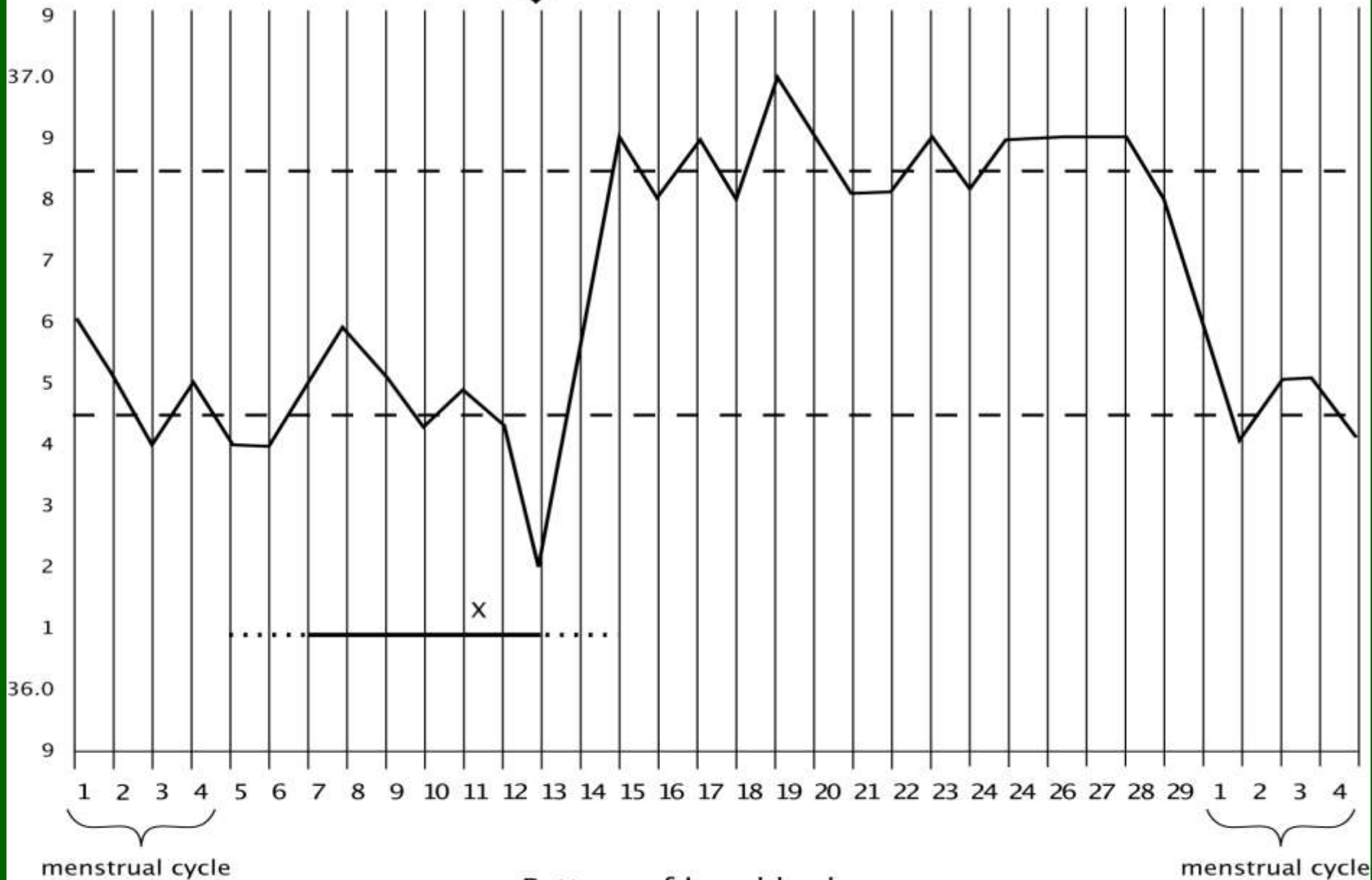
- a) Protection of germ cells: avoidance of tobacco, alcohol or narcotic consumption, intake of unnecessary drugs.
- b) Discontinuation of oral contraception, and removal of IUDs (condoms are suggested).
- c) Occupational history of females.
- d) Menstrual history and measurement of basal body temperature for detection of hormonal dysfunction.
- e) Start of pre-conceptual multivitamin (including folate) supplements.
- f) Recommendation that dental status is checked.
- h) Guidelines for physical exercise.
- i) Guidelines for healthy diet.



# Smoking cessation in HPS

Period	Females	Males
Previous smokers	34%	46%
At the first visit <b>Educational course</b>	17.9%	24.2%
At the conception	7.9%	18.0%

Day of ovulation



Pattern of basal body temperature measurement

# Characteristics of Congenital Abnormalities: (CAs)

Have a very early onset

Caused by unfavourable conditions

Optimal solution is prevention

# Neural-tube defects (NTD)

1. anencephalus (a, b)

2. encephalocele, occipital

3. spina bifida aperta (a)

spina bifida cystica (b)

closed spina bifida (c)

spinal dysraphism (d)

1/a



1/b





2







3/a



3/b





3/c

3/d



# Characteristics of NTD

1. Polygenic predisposition: recurrence is 10-fold higher than general birth rate.
2. Environmental factors: for example socioeconomic factors (diet ?).
3. Early critical period: between 15th and 28th postconceptional days, this explains the use of "periconceptional supplementation".

# Data and results of previous intervention studies for the reduction of recurrent NTD

Type	Method	Location	Supplement	Risk Reduction
Recurrence	Non-randomized	Yorkshire	Multivitamin (0.36 mg Folic Acid)	91%
		Northern Ireland		83%
	Randomized	Multicenter MRC	Folic Acid (4.0 mg)	71%



# Goals of the Hungarian randomized double-blind controlled trial (RCT)

- About 95% of women with NTD offspring have no previous NTD pregnancies.
- Thus the question is whether the periconceptional folic acid-containing multivitamin supplementation can reduce the first occurrence of NTD.
- The pharmacological dose ( $> 1$  mg, e.g., 4 mg) of folic acid cannot be recommended for the population at large or without medical supervision.
- Thus, the question is whether a physiological dose ( $< 1$  mg) is effective.
- Investigate possible other beneficial or adverse effects of periconceptional multivitamin supplementation.

# RCT

50% of participants in HPS were supplied by 'multivitamin' while other half were supplied by placebo-like trace elements.

# Composition of supplements

"Multivitamin in (Elevit Pronatal)"	"Placebo-like Trace Elements"	
<b>Vitamins</b>		
A	4000 IU	
B1	1.6 mg	
B2	1.8 mg	
Nicotinamid	19.0 mg	
B6	2.6 mg	
Calcium Panthothenate	10.0 mg	
Biotin	0.2 mg	
B12	4.0 mcg	
C	100.0 mg	7.5 mg
D	500.0 IU	
E	15.0 mg	
<i>Folic Acid</i>	0.8 mg	
<b>Minerals</b>		
Calcium	125.0 mg	
Phosphorus	125.0 mg	
Magnesium	100.0 mg	
Iron	60.0 mg	
<b>Trace Elements</b>		
Copper	1.0 mg	1.0 mg
Manganese	1.0 mg	1.0 mg
Zinc	7.5 mg	7.5 mg

# Result of the RCT: Reduction of the First Occurrence of NTD

Study groups	Number of informative offspring	Observed NTD		Expected NTD	
		No.	per 1000	No.	per 1000
Multivitamin	2,471	0	0.00	6.9	2.78
Placebo-like trace element	2,391	6*	2.51	6.6	2.78
Relative risk (with 95% confidence interval) = 0.06 (0.00, 0.63)					
Fisher test $P_2 = 0.01$					

\* anencephaly 2, spina bifida aperta 2, anencephaly + spina bifida 2



# Number and rate (per 1000) of different CA-groups in multivitamin and no multivitamin supplemented group

Categories of CAs Group of CAs	Multivitamin (N=2,471)		No multivitamin (N=2,391)		RR (with 95% CI)
	No.	Rate	No.	Rate	
Isolated CAs					
NTD	0	0.0	6	2.51	0.07 (0.04, 0.13)
Orofacial clefts	4	1.62	5	2.09	0.77 (0.22, 2.69)
Cardiovascular CAs	10	4.05	20	8.36	0.42 (0.19, 0.98)
CAs of urinary tract	2	0.81	9	3.76	0.21 (0.05, 0.95)
Limb deficiencies	1	0.40	5	2.09	0.19 (0.03, 1.18)
Cong. pyloric stenosis	2	0.81	8	3.34	0.24 (0.05, 1.14)
Others	22	8.90	32	13.38	0.68 (0.37, 1.10)
Multiple CAs	10	4.05	12	5.02	0.81 (0.36, 1,26)
<b>Total</b>	<b>51</b>	<b>20.64</b>	<b>97</b>	<b>40.57</b>	<b>0.53 (0.35, 0.70)</b>

# Conclusion

- Periconceptional supplementation with multivitamin and folic acid reduced the risk of occurrence of NTD and some other CA.

# The objectives of cohort controlled trial ( CCT )

1. To confirm or to exclude the preventive effect of periconceptional multivitamin supplementation for urinary tract and cardiovascular defects, limb deficiencies and pyloric stenosis.
2. To get a more accurate estimation for the source of NTD reduction.
3. To collect more data of orofacial clefts.

## Two Hungarian intervention trials based on HPS to estimate the efficacy of periconceptional folic acid-containing multivitamin supplementation for the prevention of first occurrence of neural tube defects (NTD)

<u>Intervention trials</u>	<u>Supplement</u>	<u>No supplement</u>
<u>Randomized controlled trial (RCT)</u>		
Number of offspring	2,471	2,391
Number of NTD	0	6
RR (with 95% CI)	<b>0.07 (0.04-0.13)</b>	
<u>Cohort controlled trial (CCT)</u>		
Number of offspring	3,056	3,056
Number of NTD	1	9
OR (with 95% CI)	<b>0.11 (0.01-0.91)</b>	
<u>Pooled data</u>		
Number of offspring	5,527	5,447
Expected/observed number of NTD	15.4/1	15.2/15
OR (with 95% CI)	<b>0.08 (0.01-0.47)</b>	

# The efficacy of periconceptional multivitamin supplementation (MS) in the primary prevention of some major groups of congenital abnormalities (CA)

CA groups	RCT No MS (n=2,391)	RCT MS (n=2,471))	CCT No MS (n=3,056)	CCT MS (n=3,056)	Pooled data No-MS (n=5,447)	Pooled data MS (n=5,447)
<b>Cardiovascular CA</b>	20	10	50	31	70	41
OR (95% CI)	<b>0.42</b>	<b>(0.19-0.98)</b>	<b>0.60</b>	<b>(0.38-0.96)</b>	<b>0.57</b>	<b>(0.39-0.85)</b>
Conotruncal CA	10	3	20	8	30	11
<b>Urinary tract CA</b>	9	2	19	14	28	16
OR (95% CI)	<b>0.21</b>	<b>(0.05-0.95)</b>	0.71	(0.33-1.50)	0.56	(0.30-1.04)
Obstructive CA	5	1	19	10	24	11
<b>Cong. limb deficiencies</b>	5	1	3	1	8	2
OR (95% CI)	0.19	(0.03-1.18)	0.33	(0.01-3.71)	0.25	(0.05-1.16)
<b>Orofacial clefts</b>	5	4	3	4	8	8
Cleft lip ± palate	3	4	2	3	5	7
Cleft palate	2	0	1	1	3	1
OR (95% CI)	0.77	(0.22-2.69)	1.63	(0.31-2.88)	0.99	(0.37-2.63)
<b>Multiple CA</b>	5	6	15	12	20	18
OR (95% CI)	1.16	(0.35-3.81)	0.79	(0.40-1.48)	0.89	(0.47-1.68)

# Other observational studies regarding periconceptional (folic acid containing) multivitamin supplementation

“Other” CAs	Association	
	confirmed	refused
Cardiovascular CAs	5	1
CAs of urinary tract	3	0
Congenital limb deficiencies	3	0
Congenital pyloric stenosis	0	1

# Question 1.:

Can folic acid-containing multivitamin prevent other defects beyond neural-tube defects?

# Reply – Probable

Congenital abnormality (CA)	Our cohort controlled trial OR 95% CI	US observational studies Yes / No
Cardiovascular CA	0.60, 0.38-0.96	-
Conotruncal CA	0.26, 0.09-0.72	3 / 1
CA of urinary tract	0.74, 0.34-1.55	2 / 0
Obstructive CA of pelvic - ureteric junction	0.15, 0.02-0.68	-
Limb deficiencies (/terminal transverse)	0.25, 0.05-1.16	3 / 0
	-	



# WHO Expert Committee (2004)

Folic acid-containing multivitamins  
can reduce the incidence of  
congenital defects by about one third

## Question 2.:

What is the mechanism of folic acid or folic acid-containing multivitamins in the prevention of NTD and other CA?

# Reply

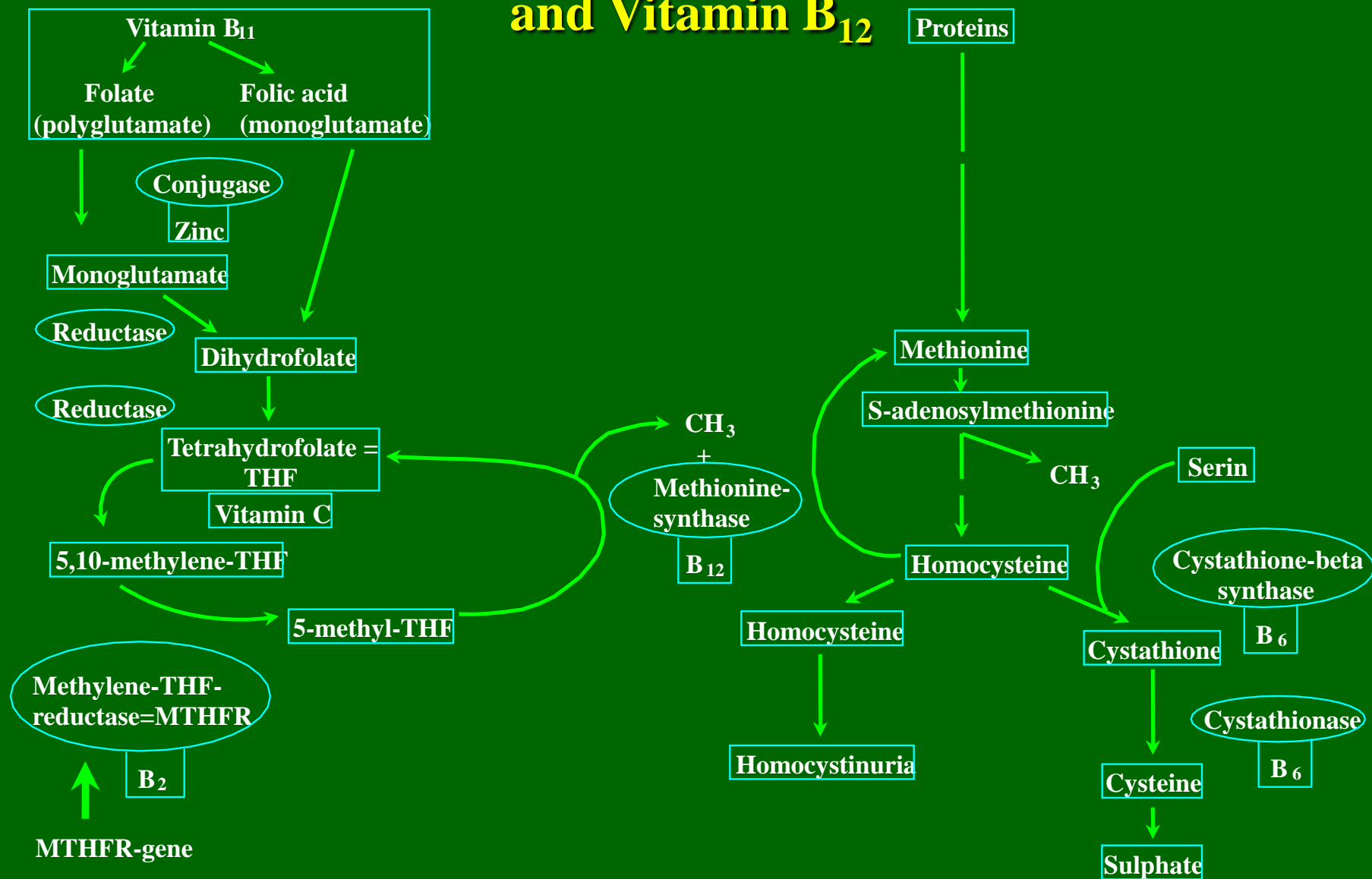
These CAs are caused by

polygenic — environmental interaction

different gene  
mutations, e.g.  
hyperhomocysteinemia

different factors,  
mainly dietary  
vitamin (folate)  
deficiency

# Metabolism of Homocysteine and the Effect of Folate-Folic Acid (Vitamin B<sub>11</sub>), Vitamin B<sub>2</sub>, Vitamin B<sub>6</sub> and Vitamin B<sub>12</sub>



# MTHFR gene

- Gene location: Chromosome I, short arm 36.3
- Mutation: 677 T → C
- Frequency of
  - mutant homozygosity: 5-15 % (11%)
  - heterozygosity: 25-65% (45%)

# Detoxication of homocysteine needs

CH<sub>3</sub> produced by MTHFR due to folate/folic  
and

MTHFR gene-pair + vitamin B2

Methioninesynthase + vitamin B12

Cystationsynthase + vitamin B6

# Reduction of hyperhomocysteinemia

by

Vitamin B11 (folate, folic acid)

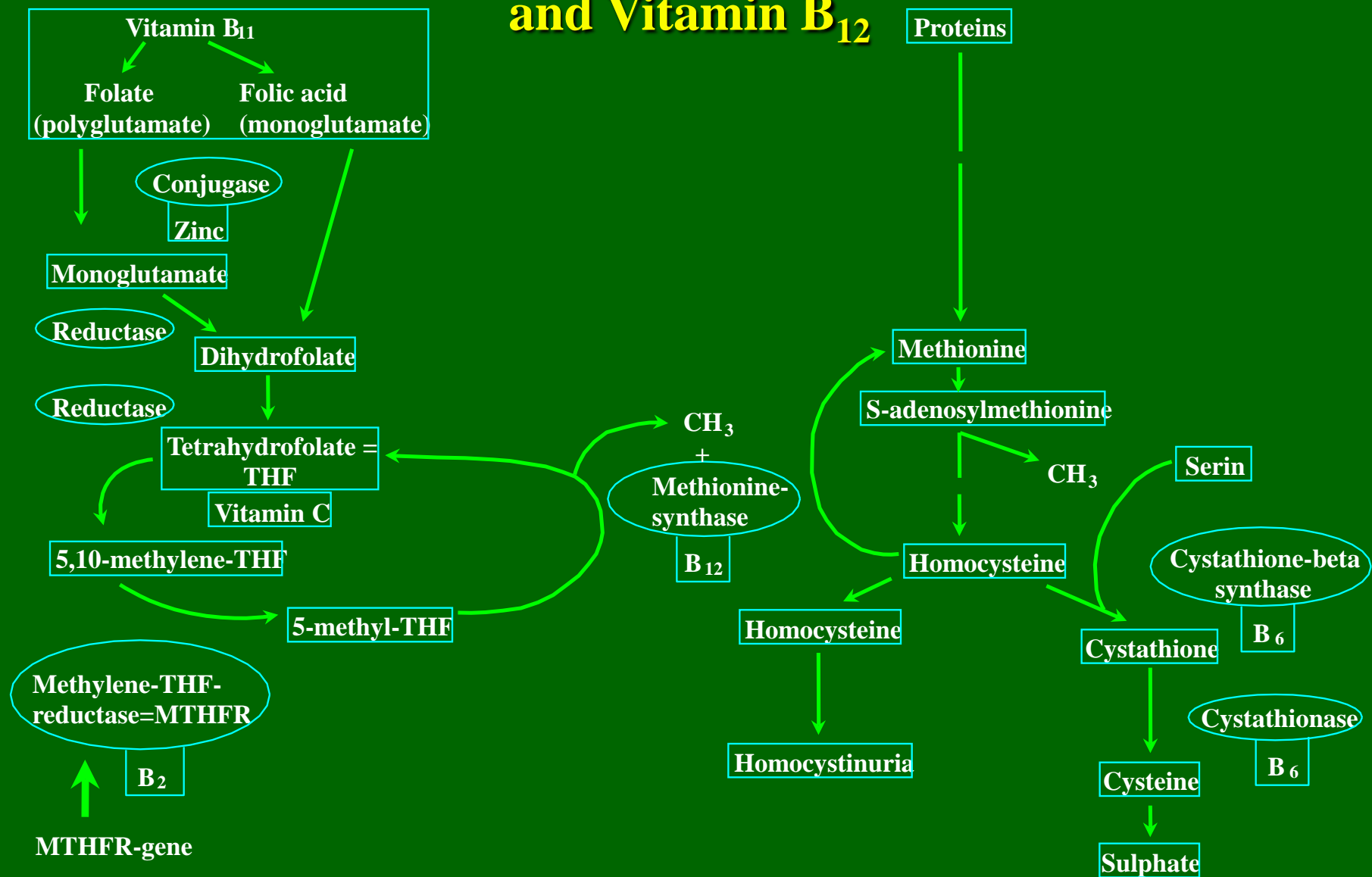
Vitamin B12 (cobalamine)

Vitamin B6 (pyridoxine)

Vitamin B2 (riboflavin)

because these “fetal protective vitamins” can stimulate MTHFR  
activity on the contrary of homo – or heterozygosity of MTHFR  
gene-pair

# Metabolism of Homocysteine and the Effect of Folate-Folic Acid (Vitamin B<sub>11</sub>), Vitamin B<sub>2</sub>, Vitamin B<sub>6</sub> and Vitamin B<sub>12</sub>





## Question 3.:

Is dietary strategy to increase folate intake can neutralise the genetic predisposition for these CAs?

**Reply:** unlikely

# Why?

Low mean folate intake	0.18 mg/day
Optimal dose for prevention of NTD (McPartlin et al., 1993)	0.66 mg/day
Difference (15 plates of spinach or broccoli!)	0.50 mg/day
Low bioavailability of folate in food (30-80%)	
There is a threshold in folate absorption from gastroenteral system	

# Question 4.:

What is optimal recommendation ?

Periconceptional folic acid or folic acid-containing supplementation seems to be appropriate .

# Question 5.:

Whether folic acid alone or folic acid-containing multivitamin is better?

# Folic acid alone or folic acid-containing multivitamin

**Folic acid alone**

**Multivitamin**

## Efficacy

70% of NTD

90% of NTD

## Other effects

?

Prevention of other major CAs

## Other arguments in hyperhomocysteinemia related NTD

Key factor  
are

Vitamin B12, B2 and B6  
independent factors

## Cost

Low

Moderate (reimbursement)

## Question 6.:

Which multivitamin is recommended?

There is only one product (Elevit®) that was tested in RCT.

# Question 7.:

**What is the optimal dose of folic acid?**

No scientific evidence.

There are two forms of Vitamin 11 (or 9)  
dietary polyglutamate folate  
synthetic monoglutamate folic acid.



US recommendation

0.4 mg (400 microgram) folic acid

The Institute of Medicine, US National  
Academy (1998) –

European Commission Scientific Committee on  
Food (1998)

**physiological dose** of folic acid (less than 1 mg)  
for preventive purpose in healthy people;  
**pharmacological dose** of folic acid (more than  
1 mg) for treatment of patients or under permanent  
medical control.

# Pros and cons

Wald et al (2001): dose/effect relation for folic acid in the reduction of hyperhomocysteinemia and related NTD.

Daily et al (1997): there is no obvious increase in the reduction of NTD due to higher doses of folic acid.

## Reasonable recommendation:

Folate	0.2 – 0.3 mg
<u>Folic acid</u>	<u>0.7 – 0.8 mg</u>
Together	1.0 mg

# Food Fortification

**USA:**

Folic Acid:

0.14 mg/100 g flour

Canada

Chile

Some other countries

**Hungary:**

Folic Acid: 0.20 mg

Vitamin B<sub>12</sub>: 1 mcg

Vitamin B<sub>6</sub>: 1.80 mg

to 200 g bread

and flour

## Conclusion

Inertia on the use of folic acid or folic acid containing multivitamins for the primary prevention of CAs is medical malpractice



### 3) Better protection of early pregnancy

- a) Undertaking of all additional investigations/treatments necessitated by conditions and disorders detected at the preconception check-up.
- b) Check the investigations and treatment of women shown to suffer from hormonal dysfunction.
- c) **Optimal timing of conception in relation to ovulation.**
- d) Early pregnancy confirmation using pregnancy tests and ultrasound scanning.
- e) Post-conceptual multivitamin supplementation.
- f) Avoidance of teratogenic and other risks.
- g) Referral of pregnant women to prenatal care clinics.

chance of  
conception (%)

35

30

25

20

15

10

5

0

-6

-5

-4

-3

-2

-1

0

+1

days

10

16

14

27

31

33

Perioovulation day with the chance of conception



Optimal day of conception is one day prior to ovulation due to reduction of overripened egg and high efficacy of conception.

# The rate of adverse pregnancy outcomes (%) in the participants of HPS and in the Hungarian population

Adverse pregnancy outcomes	HPS	Hungarian population
<b>Fetal death</b>		
Ectopic pregnancy	0.3	1.0
Miscarriage	10.6	12.5
Stillbirth	0.3	0.6
<b>Live births</b>		
Preterm birth	4.4	9.2
Congenital abnormality	2.1	4.0
Mental retardation	0.8	3.3



# Final conclusion

Proper preparation for conception is the most effective method for the prevention of adverse pregnancy outcomes.