BENEFITS E-BOOK COLLECTION

Role of Active Folate in Human Health

Unlocking the Full Potential of Folate

Quatrefolic®

Quatrefolic®: Unlocking the Full Potential of Folate

The e-book is structured to provide an in-depth look at how Quatrefolic® can influence health across various stages of life—from pregnancy and fertility to brain health and aging. Its superior bioavailability makes it particularly beneficial during pregnancy for preventing neural tube defects, crucial for supporting cognitive functions in the elderly, and effective in maintaining cardiovascular health due to its role in homocysteine metabolism.

Each section of this e-book combines rigorous scientific data with practical health insights, enriched with interactive links to videos, detailed references, and notes that enhance understanding and engagement. Our objective is to create a dynamic learning experience that highlights the significance of advanced folate supplementation in modern health practices.

As we navigate through the contents, we will explore the mechanism of action of Quatrefolic® in greater detail. This includes its role in the methylation cycle, which is essential for cellular regulation and expression, and its impact on DNA synthesis and repair. By providing the biologically active form of folate, Quatrefolic® supports these processes more effectively than traditional supplements, enhancing cellular function and overall health.

A Comprehensive Tool for Enhanced Learning and Engagement

This e-book is designed not only as a resource but as an interactive educational tool. It features an interactive index and multimedia links that provide a user-friendly experience, allowing easy access to complex scientific content. This format is intended to cater to a wide audience, ensuring that both experts in the field and those new to the topic of folate supplementation can gain a thorough understanding of the material.

Quatrefolic® offers significant health benefits that span across life stages and health conditions, emphasizing its critical role in enhancing modern nutritional practices. I invite you to delve into the pages of this e-book to discover how Quatrefolic® can support your health goals and contribute to a healthier future.

Prof. Luca Tiano PhD
Coordinator of the MSc in Food and Nutrition
at the Polytechnic University of Marche

Quatrefolic®

Active Folate and...

- Pregnancy
- Fertility
- Menopause
- Mood Health
- Cognitive Function
- Cardiovascular Health
- Glucose Metabolism
- Anemia
- Sports
- Beauty
- The Elderly

Active Folate and Pregnancy

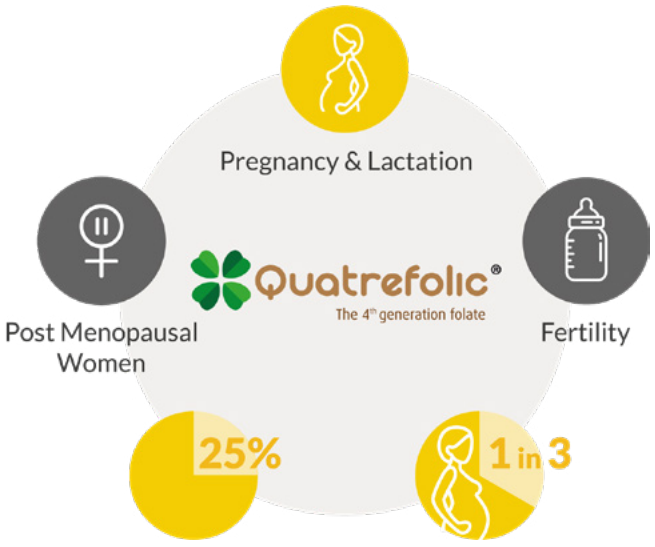
The Genetic Polymorphism Facts

The prenatal vitamins and supplements market is growing steadily due to increased awareness of their benefits and rising demand for essential nutrients like folate to support fetal growth.
The market is projected to continue growing as more women realize the vital role that prenatal supplements play in ensuring the healthy growth and development of their babies.

Today, we can assist to an inversion of the balance. The advance of scientific studies supports that not everyone can benefit of folic acid supplementation in the same way.

Therefore, the natural form is preferred and represents more than half of the shares. The trend of switching from folic acid into reduced folate is worldwide, from USA to EMEA, and is also starting in some emerging countries.

In this context, Quatrefolic® represents a real opportunity to offer innovative solutions to answer this market need.



Studies have shown that folate deficiency is relatively common in pregnant women worldwide, affecting up to 25% of pregnancies.

According to the Centers for Disease Control and Prevention (CDC), approximately 1 in 3 women in the United States do not consume enough folate.

Quatrefolic® and Pregnancy outcome vs. Folic Acid

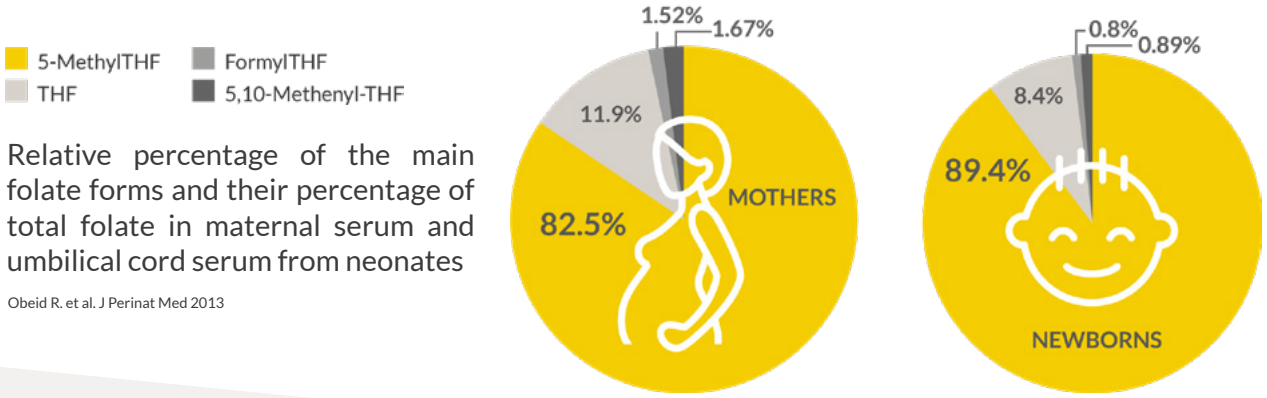
Folate is needed in cell growth, cell division, cell synthesis and repair of DNA.
During pregnancy folate requirements[G1] increase to support embryonic and fetal development and maternal tissue growth but also to reduce risk of low birth weight, preterm birth, elevated homocysteine[G2] levels and related adverse pregnancy outcomes.
For these reasons, and because humans cannot synthetize folate, its supplementation is required before and during pregnancy, with a daily dose established worldwide by relative Health Authorities (i.e. FDA, EFSA).

Quatrefolic® provides naturally active folate (5-MTHF) to protect all women and babies and maximize the benefit of folate intake.

Folate (vitamin B9) is a generic definition of a group of correlated compounds. It includes the folic acid, the synthetic form of folate that is not biologically active and the 5-methyltetrahydrofolate (5-MTHF), the naturally occurring active folate in human plasma that constitutes 95–98% of folate in serum or red blood cells (RBCs).
In vivo, the body converts dietary folic acid to 5-MTHF (also named L-methylfolate) through a multi-steps process where the enzyme methylenetetrahydrofolate reductase (MTHFR) owns a key role. Some individuals, due to their unique genetic patterns and expression, have polymorphic forms[G3] of this enzyme and do not produce adequate or effective MTHFR, with a reduction of active 5-MTHF availability.

Since the association between MTHFR polymorphism and low folate concentration has been assessed 5-MTHF the direct supplementation of the active form 5-MTHF through prenatal vitamins should be strongly considered as universally beneficial.

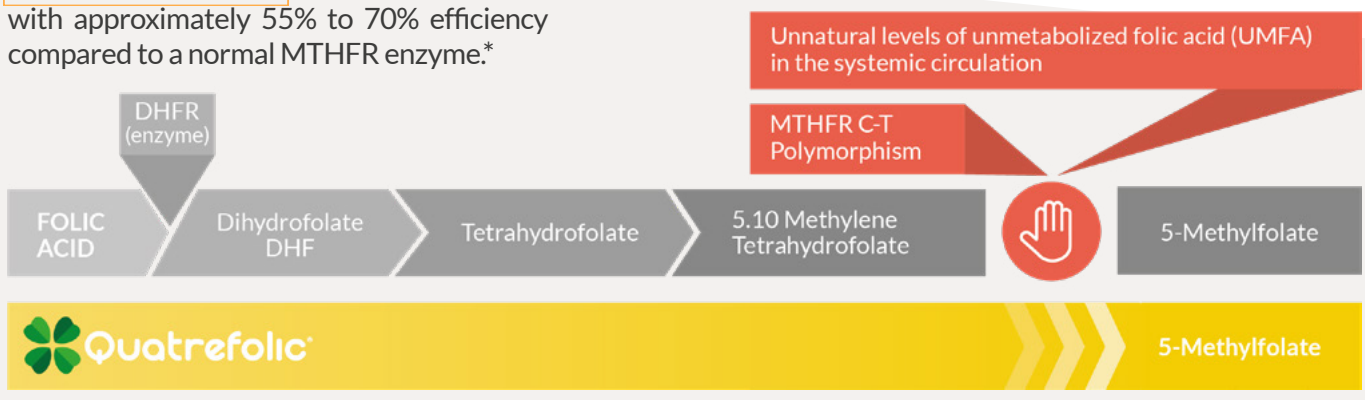
Quatrefolic® as a source of 5-MTHF might be particularly useful to provide the nutritionally active form of folate during preconception, pregnancy and lactation.



Relative percentage of the main folate forms and their percentage of total folate in maternal serum and umbilical cord serum from neonates

Obeid R. et al. J Perinat Med 2013

Polymorphic MTHFR[G4] enzyme may function with approximately 55% to 70% efficiency compared to a normal MTHFR enzyme.*



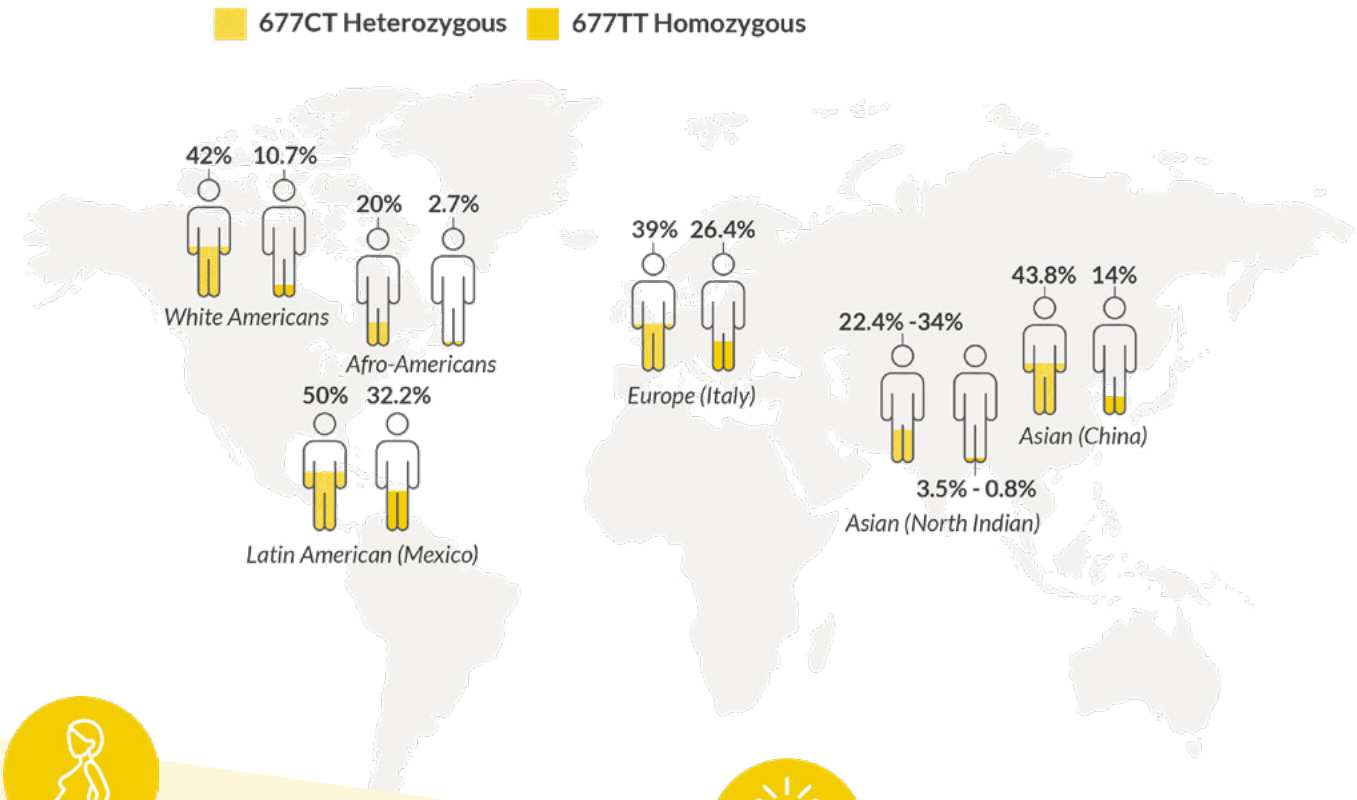
Safi J. et al. Pregnancy. 2012; Pitkin R.M. Am J Clin Nutr 2007; Castaño E. et al. PLOS ONE 2017; Nauman et al. Asia Pac J Clin Nutr. 2018; Kubo et al. Nutrients 2020; Van Mil N.H. e al. Reproduction. 2014; Sullivan M. et al. J Pharm Pharmacol 2015; Greenberg J.A. et al. Rev Obstet Gynecol 2011; Smith D.A. et al. Am J Clin Nutr 2008; Patanwala I. Am J Clin Nutr 2014; Van der Put N.M.J. et al. Am J Hum Genet 1998.
*Nowadays a total of 9 common variants (polymorphisms) has been reported. The two most common ones are C677T and A1298C. The numbers refer to their location on the gene.

Quatrefolic®, MTHFR Polymorphism and *Pregnancy* care

Testing every pregnant woman for the existence of a mutated MTHFR gene is not a medical standard protocol. The MTHFR mutation test is prescribed by doctors to people with elevated homocysteine levels and to women with cases of spina bifida or malformations in previous pregnancies.

Choosing the right prenatal vitamins, with the active form of folate like Quatrefolic®, could represent a great advantage for women that could trust in the effective bioavailability of the 5-MTHF, independently by the presence or not of MTHFR polymorphism.

Frequency of MTHFR Polymorphism and Populations screening:



Pregnancy and Preeclampsia

Preeclampsia is a pregnancy complication characterized by high blood pressure and signs of damage to another organ system, most often the liver and kidneys, that occur in women after the 20th week of pregnancy.
While rare, preeclampsia also may occur in a woman after delivering her baby, most often within 48 hours. This is called postpartum preeclampsia. It is a multifactorial and complex disorder where the involvement of MTHFR polymorphism is under evaluation: positive correlation has been found by Wang in 2013 in overall Caucasian, and East Asia populations.

Quatrefolic®, as it is the active folate, can better provide adequate levels of folate in pregnant women, independently of their genetic patterns.

Mood in Pregnancy and Postpartum

In a cohort study was showed that the prevalence of PPD (Post-Partum Depression) was significantly higher among pregnant women taking folic acid (FA) for a duration of 6 months or less than those who reported taking FA for a duration of more than 6 months during pregnancy ($P<0.05$).

Quatrefolic® might offer protection against mood impairment during pregnancy and postpartum through the normalization of the level of folate in all women, providing the direct bioavailable form of folate.

Ulrich C.M., Potter J.D. Cancer Epidemiol Biomarkers Prev 2006; Wilcken B. et al. J Med Genet. 2003; Seremak-Mrozikiewicz A et al. Neuro Endocrinol Lett 2010; Botto L.D., Yang Q. Am J Epidemiol 2000; Plumptre et al. Am J Clin Nutr 2015; Greenberg J.A. et al. Rev Obstet Gynecol 2011; Obeid R. et al. J Perinat Med 2013; Patanwala I. Am J Clin Nutr 2014; Wang W. et al. Meta Gene 1 2013.

Quatrefolic® and Hyperhomocysteinemia in Pregnancy

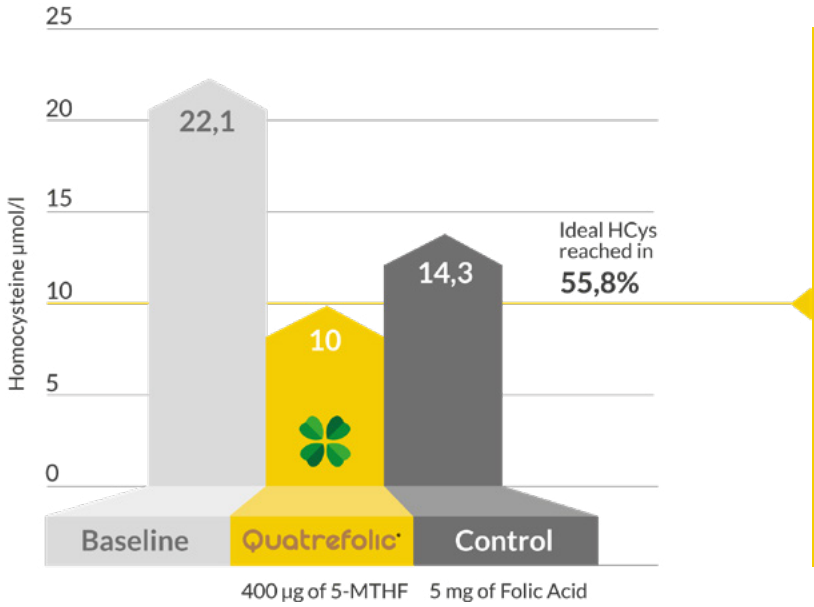
Homocysteine (Hcy) is a common amino acid found in the bloodstream and produced in the one carbon cycle. Elevated homocysteine level is considered an independent causal factor for pregnancy complications and adverse neonatal outcome.

Quatrefolic® is effective in lowering Homocysteine.

Folate is an important regulator of Hcy metabolism and helps to maintain its normal plasma concentrations. Polymorphism of the enzyme MTHFR is commonly associated with defects in folate-dependent homocysteine metabolism. Disturbance of maternal and fetal homocysteine metabolism (and low level of active folate) has been associated with fetal neural tube defects, with various conditions characterized by placental vasculopathy, such as pre-eclampsia and abruption, and with recurrent pregnancy loss.

Quatrefolic® clinical data on Homocysteine Reduction

The efficacy of Quatrefolic® (400 µg of Quatrefolic® plus B6, and B12) in lowering homocysteine serum levels (HCys) versus a conventional vitamin supplementation with highly dosed folic acid (5 mg/day) has been demonstrated in hypertensive subjects at low cardiovascular risk (104 patients with HCys 15 µmol/l).



Quatrefolic® offers a homocysteine -lowering and -normalizing effect clinically proven.

Mazza A. et al. Biol Regul Homeost Agents 2016

The result shows significant HCys reduction in comparison with baseline from 21.5 µmol/l to 10.0 µmol/l with the product containing Quatrefolic®.
The treatment was significantly effective, and the ideal HCys level was reached in 55.8% of cases in the Quatrefolic® group, and it was significantly higher than in controls.

Isotalo P.A. et al Am. J. Hum. Genet. 2000; Moustafa A.A. et al. Front Behav Neurosci 2014; Ambrosino P. et al Nutr Res 2015; Kedar R., Chandel D. Egypt J Med Hum Genet 2019; Gong M. et al. PLoS One. 2015; Laanpere M. et al. Nutr Rev 2010; Kelly C.B. et al. J Psychopharmacol 2004; Servy E.J. et al. J Assist Reprod Genet 2018; Clement A. et al. J Gynecol Obstet Hum Reprod. 2020.

Active Folate and Fertility

Impact of Active Folate and Homocysteine

According to the World Health Organization, almost **15% of reproductive-aged couples worldwide** are affected by *fertility problems*.
Available data suggests that between 48 million couples and 186 million individuals have infertility globally.

Infertility is considered as a disease of the male or female reproductive system, defined by the failure to achieve a pregnancy after 12 months or more of regular unprotected sexual intercourse.

While female infertility alone represents 35% of all infertility cases, male's represent 20–50% of them.

As it exists a wide range of causes, a low level of folate could play a crucial role. Indeed, folate is not only a nutrient needed in pregnancy to help to prevent neural tube defects (NTD) and pregnancy outcomes but it is also a vitamin essential for reproductive health. **According to the NBJ report, in 2020, folate supplements (vitamin B) represented 26% of global supplements sales in this application.**

Infertile individuals globally

Aliakbari, F. et al. Reprod Biol Endocrinol. 2020; "World Health Organization https://www.who.int/health-topics/infertility; Tamura T., Picciano MF. Am J Clin Nutr. 2006; Nutrition Business Journal, 2020 Condition Specific Report.

Fertility issues and Folate levels

WOMEN INFERTILITY

Female infertility comprises a wide range of causes affecting ovarian development, maturation of oocytes, and fertilization competence, as well as preimplantation development, implantation, and fetal growth.

MALE INFERTILITY

Male infertility is a disorder caused by numerous genetic and environmental factors that lead to defects in spermatogenesis.

LOW Folate!

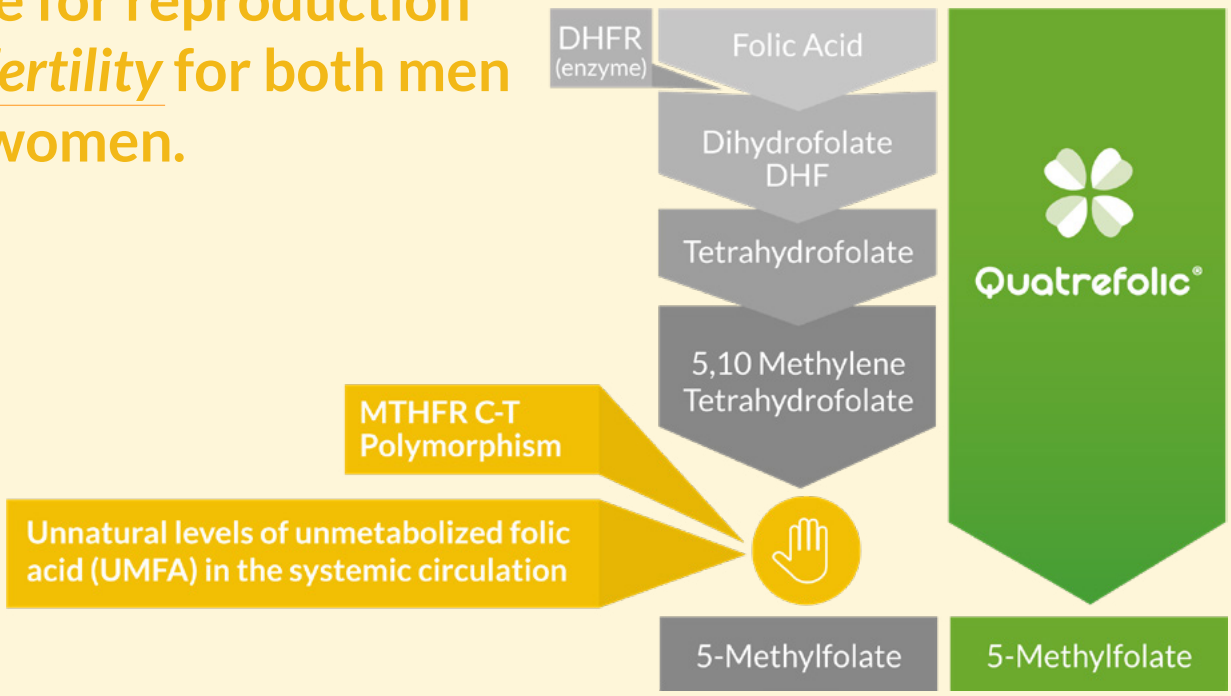
Level of folate and homocysteine status are critical factors in the early stages of human reproduction. Women and men experiencing fertility issues can present low folate availability, often linked to the presence of MTHFR enzyme polymorphism. Preconceptional folate supplementation has been linked to beneficial reproductive outcomes in both natural pregnancies and those after assisted reproductive technology (ART) treatment.

In women, folate is indispensable during the periods of rapid cell growth and proliferation, which occur during follicular and embryonic development. Insufficient folate status disrupts DNA methylation and integrity, increases blood homocysteine levels and oxidative stress one of the factors related to the pathogenesis of fertility disorders.

In men, studies support a positive correlation between serum folate concentrations, density, and normal morphology of sperm. Folate is essential in spermatogenesis and low levels in semen has been related to poor sperm DNA stability and damage. Studies have also pointed out that MTHFR gene can contribute to male infertility.

Quatrefolic® guarantees all the benefits of active folate for reproduction and *fertility* for both men and women.

Quatrefolic® demonstrates to be effective in fertility both in women and men (with MTHFR polymorphism), because it is the active folate form immediately



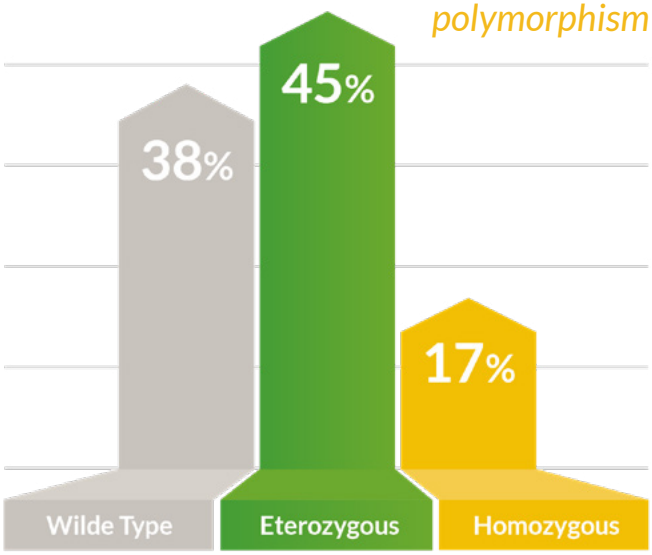
Yatsenko SA, Rajkovic A. Biol Reprod. 2019; Forges T. et al. Hum Reprod Update 2007; Bykowska-Derda A. et al. Appl Sci 2020; Rotondo JC, et al. Front Cell Dev Biol. 2021

A case series study (*Servy et al. 2018*) has evaluated the effect of Quatrefolic® in couples with recurrent miscarriages, lasting for at least 4 years; at least one of the partners was a carrier of one of the two main MTHFR isoforms.

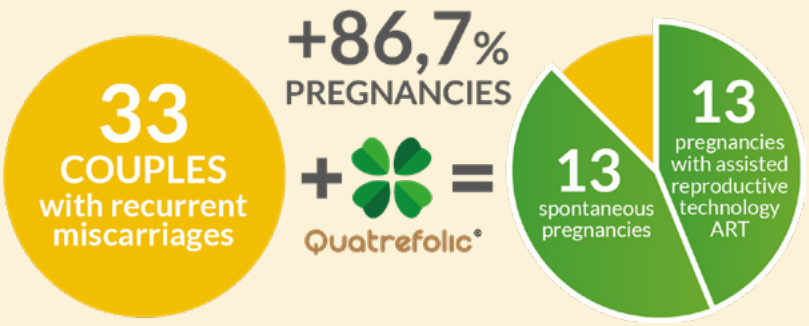
The selected population showed a strong link between an impaired folate cycle, due to the presence of MTHFR, and consequently the capacity to achieve conception and carry a pregnancy to term.

Quatrefolic® has been supplemented in men and women (with B vitamin complex and zinc) at the dosage of 800 µg/day, according to the 5-MTHF glucosamine salt requirements in healthy women. Most of the women had been previously treated unsuccessfully with high doses of folic acid (5 mg/day).

Women distribution of MTHFR polymorphism



The C677T polymorphism distribution of the women population in the clinical trial is very close to what is generally observed in Europe



The couples had long history of infertility with repeated fetal loss, premature ovarian failure or abnormal sperm parameters.

Of 33 couples, 13 spontaneous pregnancies were observed at the end of the treatment and other 13 pregnancies were obtained after assisted reproductive technology (ART), with the overall ongoing pregnancy rate of 86.7%.

The study highlights that the conventional use of large doses of folic acid (5 mg/day) has become obsolete.

A physiological dose of Quatrefolic® (800 µg) bypasses the MTHFR polymorphism and is suggested to be an effective treatment for couple fertility problems.

Laanpere M. et al. Nutrition reviews 2010; Scaglione F. et al. Xenobiotica 2014; Servy E.J. et al. J Assist Reprod Genet. 2018

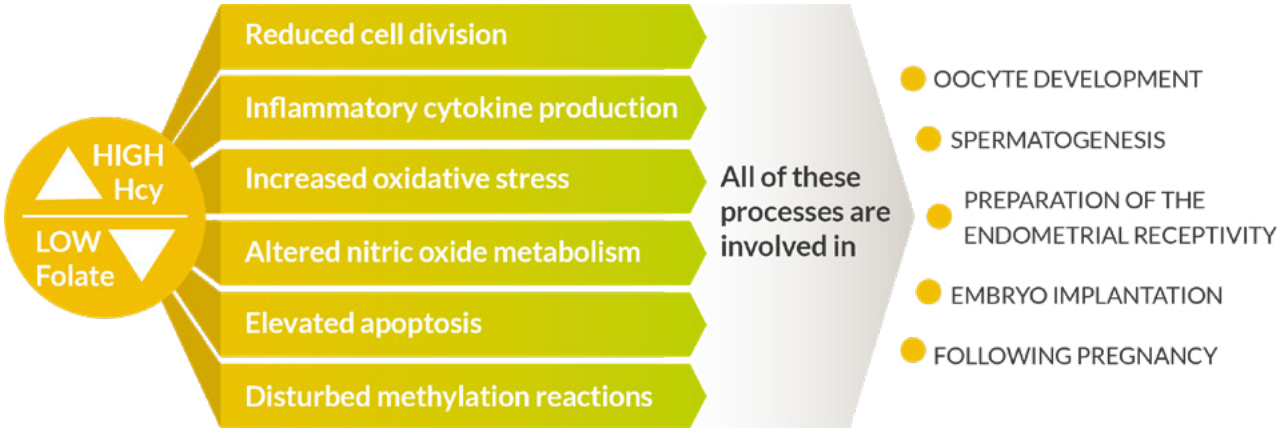
Homocysteine and Human Fertility

High levels of homocysteine and low folate can make more difficult to get pregnant for women and increase the risk of early miscarriage. Investigation of the role of homocysteine metabolism in patients with unexplained female sterility or secondary sterility due to recurrent pregnancy loss shows positive association. In men, folate deficiency results in homocysteine overproduction with subsequent excessive oxidative stress, chaotic methylation reactions, protein synthesis, and spermatogenesis deficiency.

Homocysteine is an inhibitor of the methylation process and a powerful pro-oxidant. It has a negative effect on spermatogenesis, and its concentration in the ejaculate is inversely correlated with fertility outcome.
Also, the polymorphisms of the MTHFR enzyme have been related to higher plasma homocysteine levels. Study has found that the polymorphisms would result in fetal non-viability rates of 4–7%, thus playing a significant role in human fertility.

The choice of right folate, such as Quatrefolic®, is recommended for right level of folate in *healthy pregnancy.*

Quatrefolic® provides the right folate dosage, it does not require metabolism of MTHFR enzyme, it is ready to enter human metabolism and, if necessary, normalizes homocysteine plasma levels.



Quatrefolic® provides the right folate dosage, normalizing homocysteine plasma levels.

Quatrefolic®

Jacquesson-Fournols L. et al. J Gynecol Women's Health 2017; Goyco Ortiz LE. et al. J Assist Reprod Genet 2019; Menezo, Y.J.R et al. Reprod Biomed Online 2016; Forges T. et al. Hum Reprod Update 2007; D'Elia P.Q. et al. Reprod Biomed Online. 2014; Altmäe S. et al. Fertil Steril. 2010; D'Uva M. et al. Thromb J. 2007; Reyes-Engel A. et al. Mol Hum Reprod. 2002

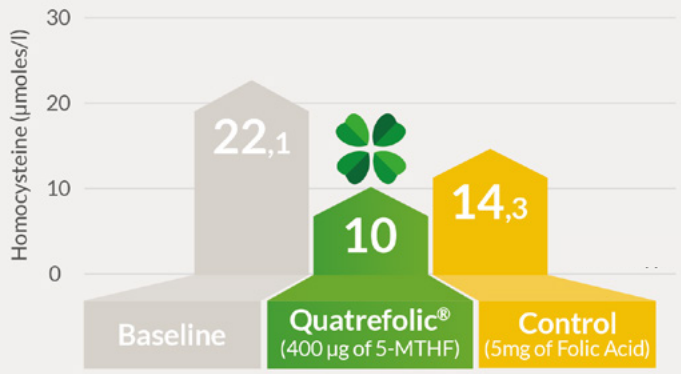
Quatrefolic® normalizes Homocysteine

Published results (Mazza et al. 2016) demonstrate the capacity of Quatrefolic® (400 µg plus B6 and B12) to lower homocysteine serum level better than conventional vitamin supplementation with highly dosed folic acid (5 mg/day).

Quatrefolic® was tested on hypertensive subjects at low cardiovascular risk (104 patients with HCys ≥15 µmol/L). The result shows significant HCys reduction in comparison with baseline from 21.5 µmol/L to 10.0 µmol/L.

Moreover, the treatment was significantly effective and the ideal HCys level was reached in 55.8% of cases in the Quatrefolic® group, and it was significantly higher than in the control one.

Polymorphisms in folate pathway genes could be one reason for fertility complications in some women with unexplained infertility.



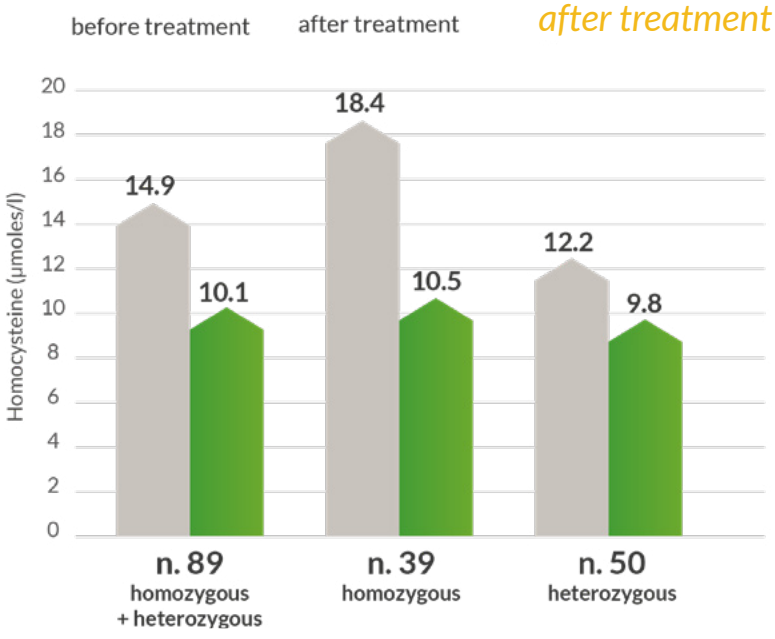
More recently, *Clement et al.* has recommended to analyze couples with a long history of infertility for MTHFR polymorphism and homocysteine, before starting Assisted Reproductive Technology (ART) attempts.

Volunteers with a history of repeated miscarriages (>3) or were facing infertility for >3 years and had at least 3 ART failures, were tested for MTHFR polymorphism (C677 T) and treated for at least 3 months with Quatrefolic® (600 µg per day complemented with B components and zinc).

In the 89 couples included, Quatrefolic® reduced rapidly the Hcy level of 42.7% in homozygous and 20.1% in heterozygous subjects, with no significant difference (p = 0.12) between the normalized homocysteine level of homozygous and heterozygous (10.5 µmol/L vs. 9.8 µmol/L).

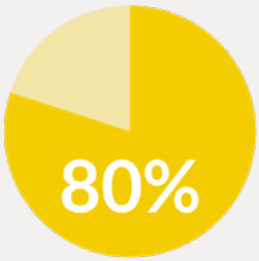
Quatrefolic® bypasses folate metabolism disturbances and may improve male and female fertility.

Variations in Hcy concentrations before and after treatment

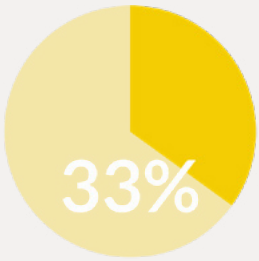


Active Folate and Menopause

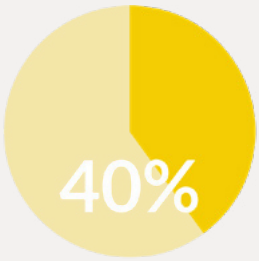
Millions of women go through menopause every year, with the majority experiencing a severe impact on everyday life.
The global menopause market size is expected to reach USD 24.4 billion by 2030, registering a CAGR of 5.37% from 2024 to 2030.
The growth of the market is influenced by the increasing aging of the female menopausal population and the increasing demand for new solutions, such as nature-based supplements.
Folate supplementation is recommended as an affordable and accessible way to help in the treatment of *menopausal* symptoms.



of women have MENOPAUSE symptoms



of PERIMENOPAUSAL women take vitamins or supplements



of women's lives are spent POST-MENOPAUSAL

Adapted from Bani S. et al., J Caring Sci. 2013

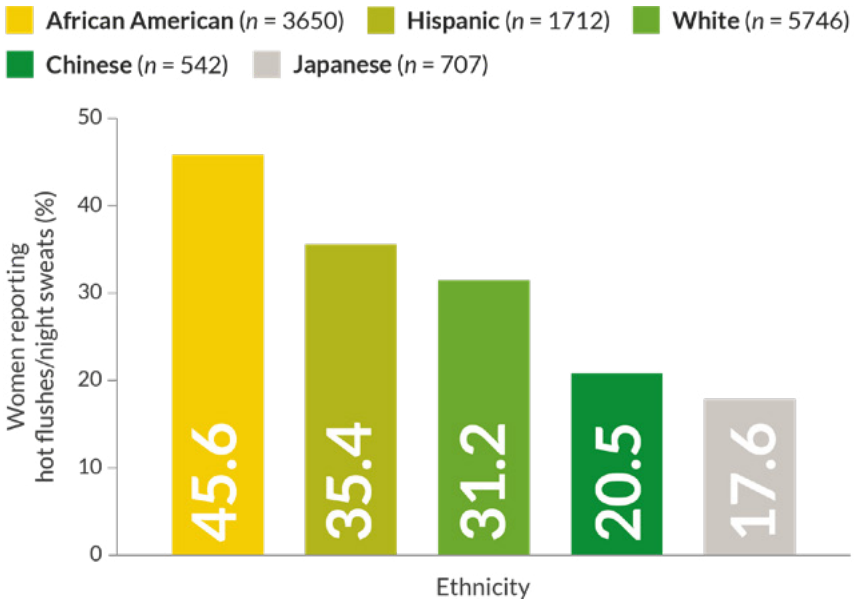
What are HFs?

Hot flashes (HFs) are the most characteristic and frequent symptoms of perimenopause and menopause, experienced by up to 75% of menopausal women, and in half of them, symptoms are severe enough to seek medical advice.

Some women find that hot flashes interrupt their daily lives. The earlier in life hot flashes begin, the longer they may experience them.
Hot flashes are temporary among the majority of women. The average duration of these symptoms ranges from 5 to 10 years, regardless of treatment. The severity and frequency of symptoms are peaking during late perimenopause and early postmenopause. They may last:

- **From 6 months to 2 years for the majority of women**
- **From 6 to 10 years for 26% of women**
- **Over 10 years for 10% of women**

Transient sensations of heat, sweating, flushing, anxiety, and chills lasting for 1–5 min.



SWAN study: reported prevalence of vasomotor symptoms in perimenopausal women
Ages 40 - 55 years
Research has found that African American and Hispanic women get hot flashes for more years than white and Asian women.

n = 12,357; SWAN = Study of Women's Health Across the Nation

Adapted from Gold EB. et al., Am J Public Health 2006

Common simptoms

- Flushing
- Abrupt sweating
- Rapid influx of heat
- Rapid or accelerated pulse

Classification of hot flashes

HIGH (feeling too much heat and sweating with disruption of daily activity).

MODERATE (feeling heat with sweating, no disruption of daily activity),

MILD (feeling heat without sweating),

Associated disturbances in HFs

INSOMNIA ▶ fatigue

BEHAVIORAL CHANGE ▶ anxiety and depression

MEMORY & CONCENTRATION PROBLEMS ▶ decrease in libido

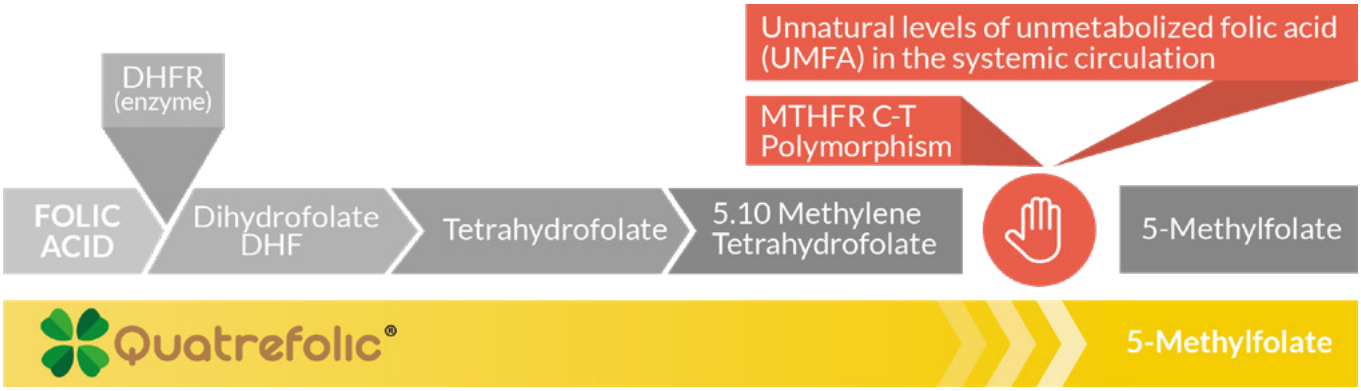
Quatrefolic® the Folate helps in Hot Flashes

Hormone replacement therapy (HRT) is the first treatment option in hot flashes. Estrogens stimulate the production of serotonin and endorphins: after menopause, there is a 50% decrease in levels of serotonin corresponding to declining estrogen levels.

Nevertheless, recently serious risk of hormone therapy has been recognized with prompted researchers to seek new alternatives. **Folate supplementation, like Quatrefolic®, is today recommended as an affordable and accessible method.**

Studies have shown that folate* supplementation significantly ameliorates hot flashes in postmenopausal women, with a mechanism similar to HRT, by interacting with monoamine neurotransmitters in the brain. The 5-Methyltetrahydrofolate (Methylfolate or 5-MTHF), the metabolically active and tissue-usable form of folic acid, is essential for the biosynthesis of the monoamine neurotransmitters serotonin and noradrenaline, and in hot flashes, it lowers noradrenaline and increases serotonin activities.

*in the form of folic acid



5-MTHF is also the form in which both folic acid and food folates are converted in the body through enzymatic biotransformation: this *enzymatic multi-step process* of conversion can be impacted by polymorphism of the methylenetetrahydrofolate reductase (MTHF), reducing the availability of active folate in the body.

Quatrefolic® is the 5-MTHF, doesn't need to be metabolized, and is ready to exert the body's biological effect. In hot flashes, it is a safe and helpful alternative option.

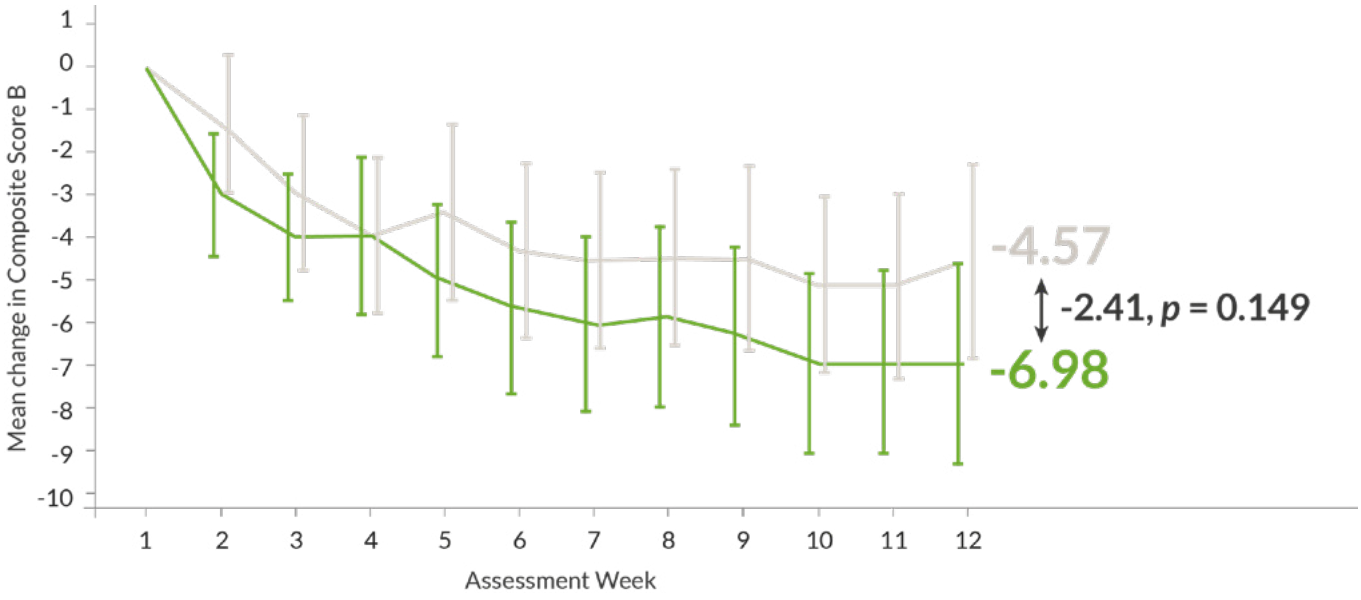
Other effective alternative therapies – like herbs and drugs including antidepressants - have not been accepted by women because have some side effects, which may reduce the patients' tolerance, other drug interactions may limit their use, and they are not effective for all patients.

The vitaminic supplementation, as Quatrefolic®, is a well-known option for all women, helpful not only for hot flashes but for general women's health.

Folate and Hot Flashes: Clinical Evidence

STUDY 1	2 GROUPS (N=46) *(5 mg folic acid/daily - 4 weeks vs placebo)*	**65%** of improvement of hot flashes* *and 16% improvement with placebo *Gaweesh SS et al., Gynecol Endocrinol. 2010*
STUDY 2	2 GROUPS (N =40) *(5 mg/daily folic acid - 4 weeks when compared with no treatment)*	**57%** reduction in the frequency in hot flushes with daily *Grodnitskaya E. et al., Climacteric. 2011 Jun 2011*
STUDY 3	2 GROUPS (N= 70) *(1 mg/daily - 4 weeks)*	Significant improvement in severity, duration, and frequency * *as well as with placebo tablets, however, there was more improvement in the folic acid group. *Bani S. et al., J Caring Sci. 2013*
STUDY 4	3 GROUPS (N =80) *(1 mg /daily folate, omega-3 1000 mg, or placebo tablets -12 weeks)*	Significant improvement in severity, duration, and frequency of hot flushes *Jahdi F. et al., Complementary Medicine Journal. 2018*
STUDY 5	2 GROUP WOMEN (N =164) *(5 mg/daily folic acid vs placebo - 12 weeks)*	Folic acid has greater benefit in reducing Hot Flush Score in postmenopausal women when compared with placebo* *Ewies AAA. et al., BJOG. 2021*

***The Study 5*, with strict inclusion and exclusion criteria, shows that folic acid had a greater benefit in reducing Hot Flush Score over 12 weeks in postmenopausal women when compared with placebo; however, the difference did not reach statistical significance*.**



Bani S. et al., J Caring Sci. 2013; Ewies AAA. Et al., World J Obstet Gynecol. 2013; Bansal R, Aggarwal N. J Midlife Health. 2019; Gold EB. et al., Am J Epidemiol 2000; Gold EB. et al., Am J Public Health 2006; Gast GC. et al., Maturitas 2010; Freedman RR. Am J Med. 2005; Women's Health Initiative Trial. JAMA. 2002; Ewies AAA. et al., BJOG. 2021; Gaweesh SS et al., Gynecol Endocrinol. 2010; Grodnitskaya E. et al., Climacteric. 2011; Jahdi F. et al., Complementary Medicine Journal. 2018; Li L. et al., Menopause. 2017; Sturdee DW et al., Climacteric. 2017; https://www.menopause.org/.
*The placebo effect in hot flashes treatment is largely documented. Specifically, a higher response rate was observed in trials of hormonal drugs than in trials of nonhormonal drugs, indicating a subjective expectations that may affect the treatment efficacy. Quantitative analysis of placebo response and factors associated with menopausal hot flashes - PubMed (nih.gov)

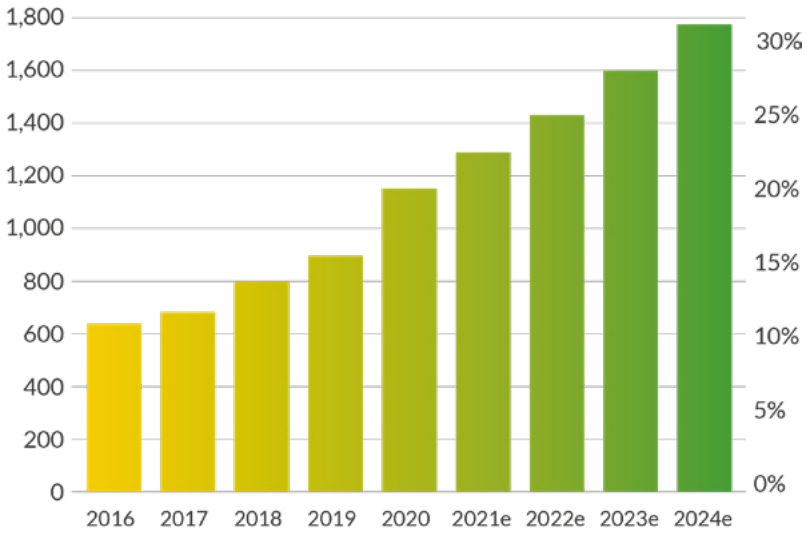
Active Folate and Mood Health

Worldwide, an estimated **264 million** people are affected by depression.
Only in the U.S.A, **42% of adults** reported symptoms of anxiety and depression, up from **11% in previous years**. As the need grows, the search for a meaningful response and a long-term solution is among the top priorities in the supplementation health trend.

The 2022 FMCG Gurus report has highlighted that **48% of global supplement consumers** had a strong desire to take care of their mental wellbeing for the next 12 months.
People recognize their mental health as important as physical health (immunity, digestive) as a levier to improve their overall wellbeing.

In reaction to these trends, the growth of sales of supplementation dedicated to mental health and mood has increased.

Mood and Mental health supplements sales



Source: Nutrition Business Journal ($mil, consumer sales)

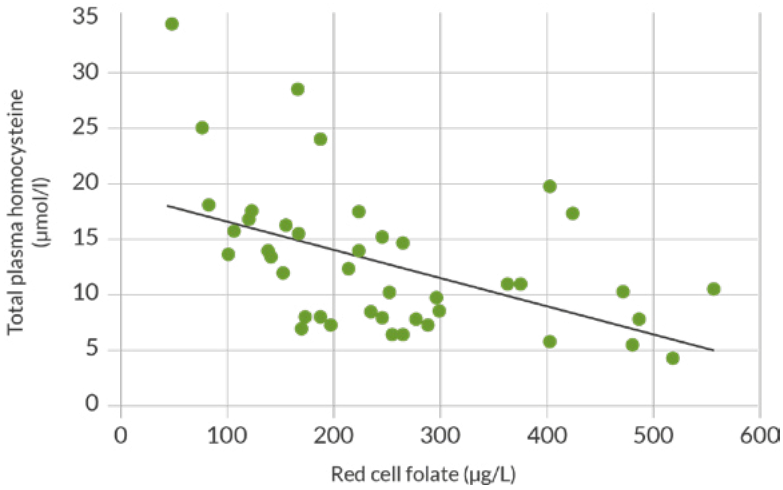
FMCG GURUS TOP TEN TRENDS FOR 2022
https://www.nature.com/articles/d41586-021-00175-z

The connection of Active Folate with Mood Impairment

Folate deficiency has been suggested as a risk factor for mood impairment as well as for potential inadequate response to treatment. Along with vitamin B12, the biologically active form of folate (methylfolate or 5-MTHF) serves as a donor of methyl groups in the critical pathway of the synthesis of monoamine neurotransmitters. **Low levels of 5-MTHF critically contribute to reduce the neurotransmitter levels in the Cerebral Spinal Fluid*, whilst to increase homocysteine levels, some of the main factors that lead to the onset of the mood impairments.**

In 2000, Bottiglieri et al. studied the relationship between homocysteine, folate and monoamine metabolism in patients with severe depression, finding that red cell folate concentrations significantly correlate with plasma homocysteine and that nearly one third of severely depressed patients has folate deficiency.

1/3 of severely depressed patients has folate deficiency



Bottiglieri T. Am J Clin Nutr 2002; Nguyen B. et al. J Affect Disord 2017; Almeida O.P. et al. Arch Gen Psychiatry 2008; Miller A.L. Altern Med Rev 2008; Jacques P.F. et al. Circul 1996.

The same data have been confirmed by *Nguyen et al. in 2017*, highlighting the link between folate concentration in the blood and the increased risk of moderate depression.

In older adults, high tHcy increases the risk of mood impairment as shown by a meta-analysis published in 2008 by *Almeida et al.* It has been demonstrated that supplementation of folate lowers the buildup of homocysteine and benefits mood balance and brain health.

5-MTHF such as Quatrefolic® reduces homocysteine more effectively than folic acid because it is the only folate crossing the "Blood-Brain Barrier" (BBB) and can straightly enter the brain without metabolism or genetic MTHFR impairment.**

*Cerebrospinal fluid (CSF) is a clear, colorless body fluid found within the tissue that surrounds the brain and spinal cord of all vertebrates.
**Blood Brain Barrier (BBB) is one of the most complex and selective barriers in the human organism, with the role to protect the brain and preserve the homeostasis of the central nervous system (CNS).

Quatrefolic® advantages in MTHFR Polymorphism

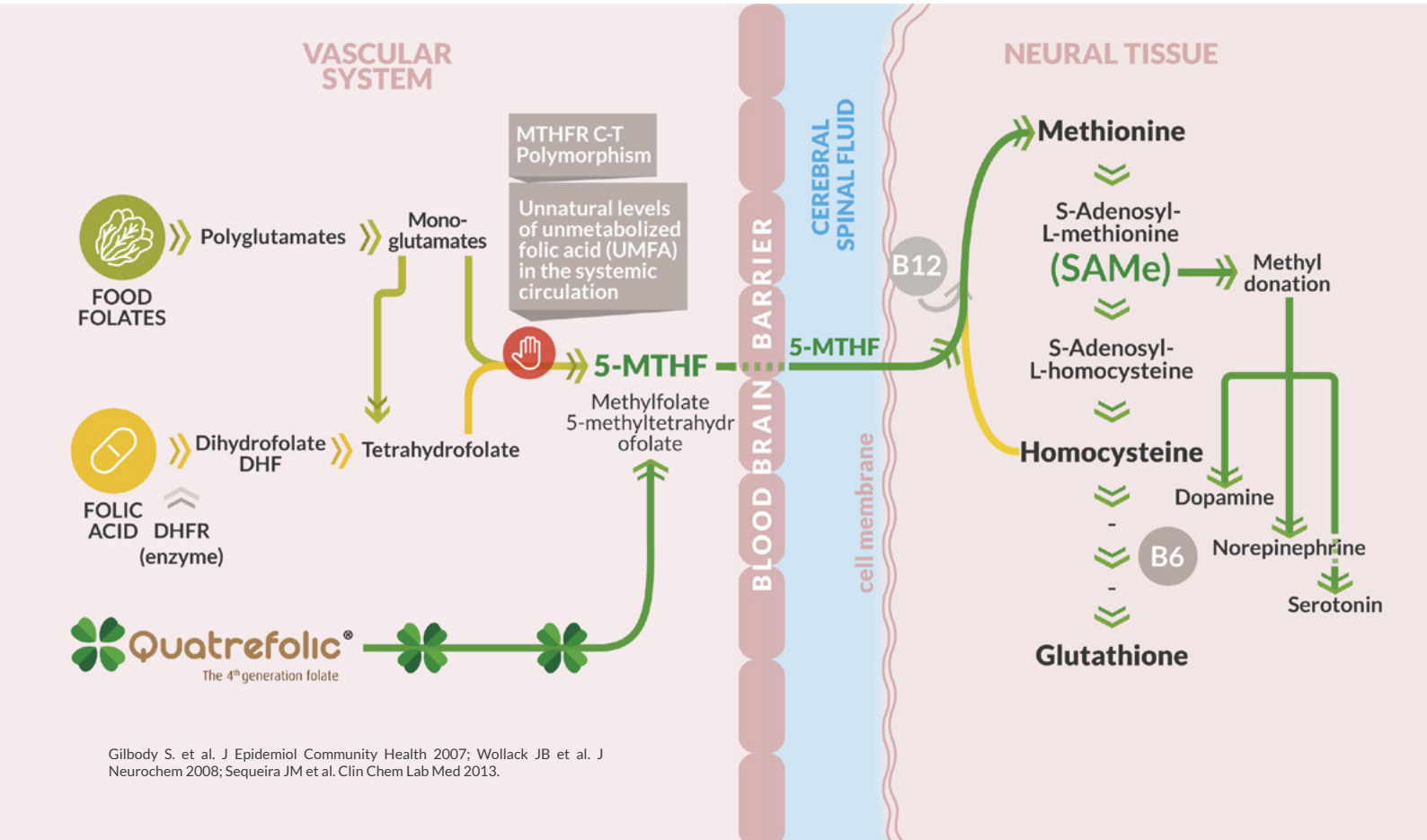
Lower systemic levels of folate can result from poor dietary intake, various gastrointestinal disorders, drug use, chronic diseases, and a particular and serious genetic polymorphism related to the methylenetetrahydrofolate reductase enzyme (MTHFR), a key regulatory enzyme in folate and homocysteine metabolism, which is responsible for a key step of folate cycle where 5-MTHF is irreversibly produced.

People with polymorphism have a reduced ability to convert folate into the centrally active compound, 5-MTHF. MHTFR polymorphism impairs the biochemical transformation of other folates and of the synthetic man-made folic acid, making their intake or supplementation low effective. This mutation is the most common genetic cause of increased homocysteine levels.

In 2007, *Gilbody et al.* published a meta-analysis finding a significant correlation between this polymorphism, folate levels and mood disorders, including 11 relevant studies with 15,315 participants. Gilbody found that patients with the homozygote (TT) variant of MTHFR over the normal subjects with wild-type (CC) genotypes have a 1.36 times greater chance of developing depression. The odds of having the TT genotype is twice as great in depressed patients.

5-MTHF, such as Quatrefolic®, can support mood functions because it is not affected by this common impairment of folate metabolism and is able to bypass the critic enzymatic step. As it is, 5-MTHF crosses the BBB and enters the central nervous system where it sustains the release of monoamines neurotransmitters – serotonin, norepinephrine and dopamine – through the participation in one-carbon metabolism cycle.

In this key network of interrelated reactions, that involves the transfer of one-carbon methyl groups from one compound to another, 5-MTHF lets the homocysteine levels decreasing through its remethylation to methionine and the next S-adenosylmethionine (SAMe) production.



Gilbody S. et al. J Epidemiol Community Health 2007; Wollack JB et al. J Neurochem 2008; Sequeira JM et al. Clin Chem Lab Med 2013.

Quatrefolic® and Mood in Pregnancy and Postpartum

The depletion of nutrient reserves throughout pregnancy and a lack of postpartum recovery may increase the risk of mood impairment in women. The most notable nutritional deficiencies[G5] associated with *postpartum depression* include folate.

Disturbances in methyl metabolism, such as those associated with the MTHFR 677TT genotype and the perturbed methionine–homocysteine metabolism, have been described as a factor that may accentuate the deficiency of folate.

While folate supplementation may be useful to reduce disturbances in folate metabolism, Quatrefolic® might offer protection against mood impairment during pregnancy and postpartum through the normalization of the level of folate in all women, including those with MTHFR polymorphism.

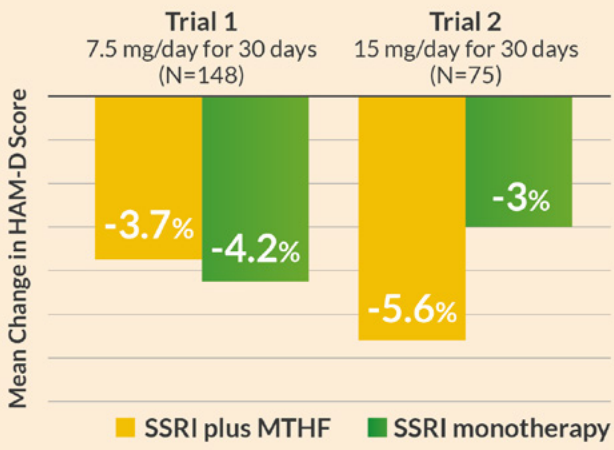
Yan et al. Nutrients. 2017; Smits L.J., Essed G.G. Lancet 2001; Devlin A.M. et al. PLoS One 2010; Aishwarya S. et al. Asian J Psychiatr 2013; Lewis S. et al. Eur J Clin Nutr. 2012; Knudson-Martin C, Silverstein R. J Marital Fam Ther. 2009.

Quatrefolic® and contribution in Non-responders to antidepressant

Studies suggest that low levels of 5-MTHF critically contribute to reduce neurotransmitters production and increase homocysteine levels, some of the main factors that lead to onset of the mood impairment and depression.

Partial- or non-responders to antidepressant drugs may be gravely folate deficient, caused by the polymorphism MTHFR, which is quite common among patients with depression. Up to 70% of patients with depression test positive for the polymorphism rendering folic acid supplements ineffective for helping in depression.

These individuals have impaired capacity to convert food folate/ folic acid into the metabolic active form, the 5-MTHF.



Adjunctive L-Methylfolate may constitute an effective, and safe strategy for patients with major depressive disorder who have a partial response or no response to antidepressant drugs (SSRI), has been reported by Papakostas in 2012.

In 2021 a meta-analysis published by *Altaf et al.* has provided supporting evidence for the addition of methylfolate to antidepressant drugs **to be effective and safe in major depressive disorders, highlighting the likely correlation of folate with the synthesis of monoamines which play an essential role in symptoms of depression.** The study has concluded that folate supplementation is a safe treatment that improves response, remission, and overall depression scale scores.

Papakostas GI et al. Am J Psychiatry. 2012; Altaf R. et al. Complement Ther Med. 2021; Page R et al. Am J Clin Nutr 2017 21; Knowles L et al. JIMD reports 2016.

Quatrefolic® at the brain level: 5-MTHF vs. Folic Acid

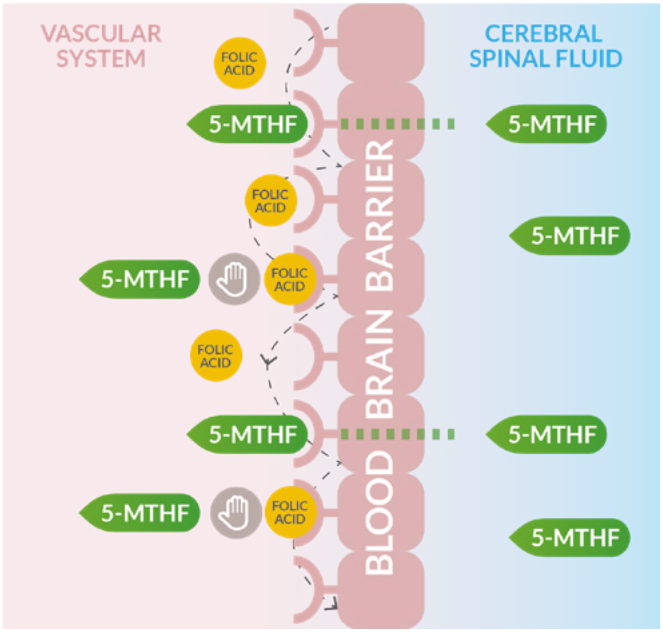
Quatrefolic®, as being the active folate form 5-MTHF, does not require metabolization and is immediately available in the brain to support SAMe release and consequently the production of neurotransmitters, avoiding the presence of unmetabolized folic acid (UMFA) and protecting the body from its potential injurious effects.

Quatrefolic® offers proven advantages over folic acid by allowing more 5-MTHF to cross the blood-brain barrier.

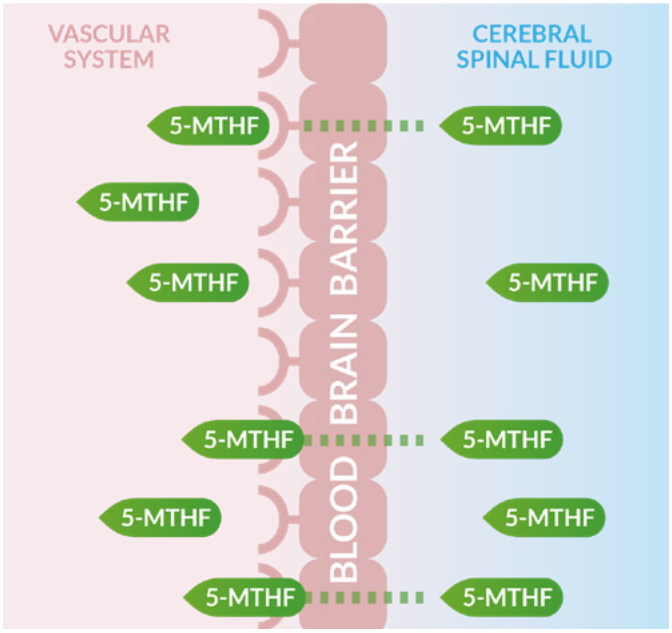
In normal subjects, the intake of folic acid (FA) higher than 200-300 µg/day leads to the direct appearance of UMFA in the plasma which can also persist for many years, depending on the levels of intake. Unmetabolized levels of FA may be amplified in subjects positive to MTHFR polymorphism where metabolism is impaired and/or slacked.

UMFA is a current concern for the potential toxic effects. In the brain UMFA is not able to cross the BBB and may become bound to receptors (folate binding protein) on the membrane competing for cellular transport, thereby blocking the 5–MTHF bound and its absorption. Consequently, the amount of 5-MTHF crossing the BBB into the cerebral spinal fluid may be reduced, with a folate deficiency that may lead to mood dysfunction.

FOLIC ACID BLOCKS L-METHYLFOLATE TRANSPORT ACROSS THE BLOOD BRAIN BARRIER



UNHINDERED TRANSPORT OF L-METHYLFOLATE ACROSS THE BLOOD BRAIN BARRIER



Papakostas G.I. et al. Am J Psychiatry, 2012; Page R et al. Am J Clin Nutr 2017 21; Knowles L. et al. JIMD Reports 2016; Ionescu D.F. et al. Dialogues Clin Neurosci 2015; Shelton R.C. et al. CNS Disord 2013; Papakostas, G.I. et al Can J Psychiatry 2012.

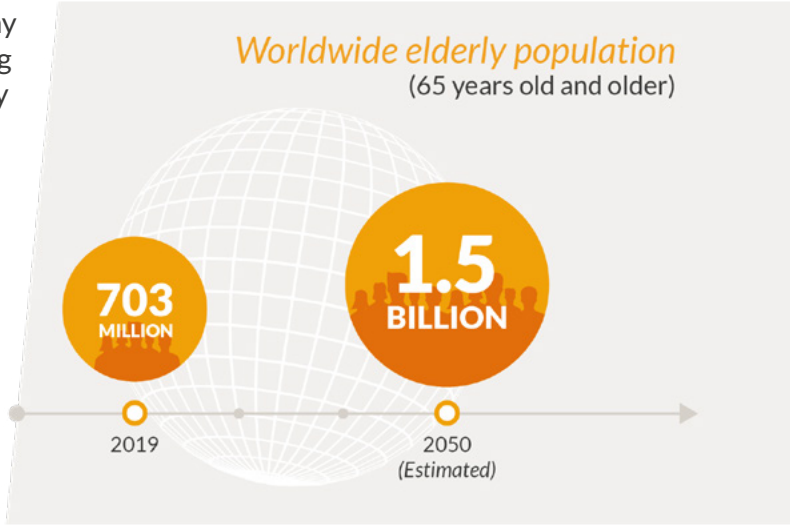
Active Folate and Cognitive Function

The Support over a Lifetime

Cognitive health describes the ability to learn, think, and remember. As global life expectancy increases, it is essential to optimize cognitive function.

The world's population is aging, and people over 65 are the fastest-growing age group: by 2050, one in six people in the world will be over age 65 (16%), up from one in 11 in 2019 (9%). Cognitive health is a key component of healthy aging as it is vital in maintaining a good quality of life and the independence to perform everyday activities. However, overall cognitive health tends to decrease with age, and enhancing healthy habits through proper nutritional intakes, including active folate, can help keep the brain optimally operating.

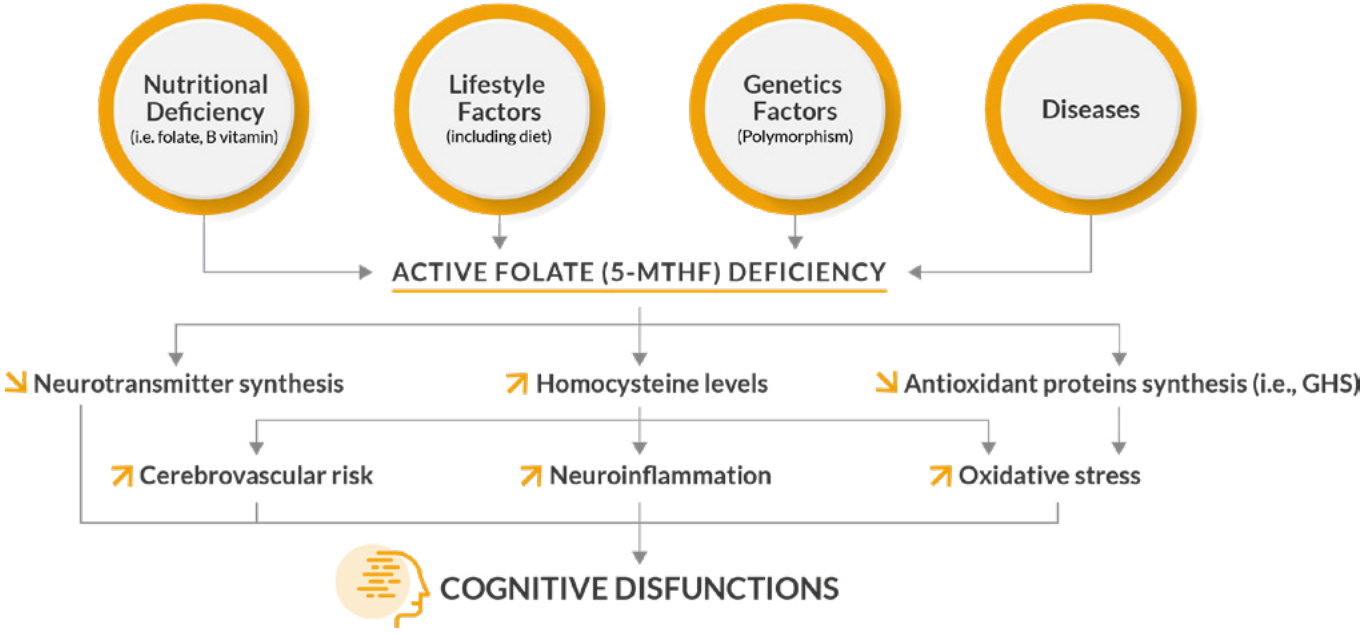
Brain Health is among the most promising and dynamic segments in the total supplement market (+8% growth vs PY, vs +4% market), with 75% of the oldest adults saying that healthy aging is extremely or very important.



The Essential Role of Active Folate in Cognition

Folate (vitamin B9) plays an important role in *cognitive functions* and nerve signaling in the brain, and its deficiency can potentially elevate the risks of cognitive impairment. In its biological form 5-methyltetrahydrofolate, folate is required for working the one-carbon cycle essential for the synthesis of neurotransmitters, key proteins as well as antioxidants in the brain.

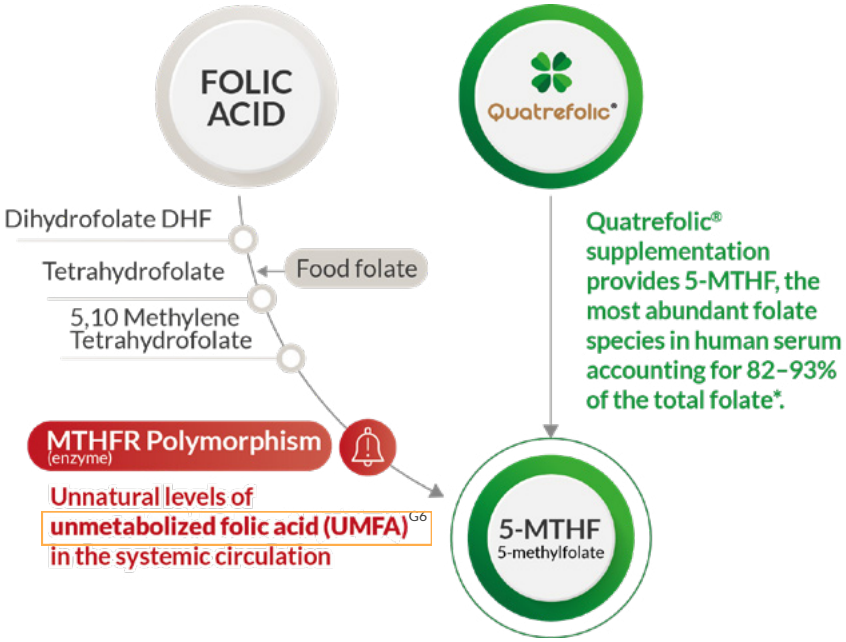
Relationship between folate deficiency, poor cognitive function, and homocysteine



Supplementation with Quatrefolic®, the glucosamine salt of 5-MTHF, can maximize *cognitive benefits*, providing the active form of folate without requiring metabolization:

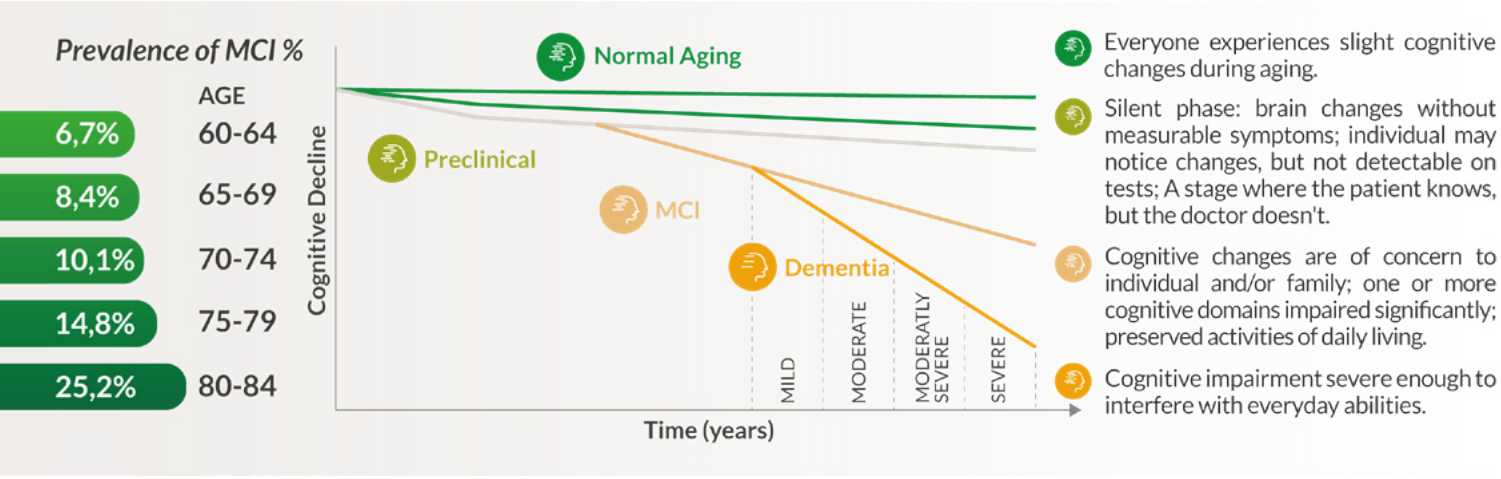
Both food folate and folic acid need to be converted into active folate 5-MTHF through a multistep enzymatic conversion, where the enzyme methylenetetrahydrofolate reductase (MTHFR) plays a key role.

MTHFR polymorphism impairs the ability of the MTHFR to properly convert dietary folate and folic acid into active form of folate (5-MTHF).



*Pfeiffer C.M., Fazili Z., McCoy L., Zhang M., Gunter E.W. Determination of folate vitamers in human serum by stable-is tope-dilution tandem mass spectrometry and comparison with radioassay and microbiologic assay. Clin. Chem. 2004; Kirsch S.H., Knapp J.-P., Herrmann W., Obeid R. Quantification of key folate forms in serum using stable-isotope dilution ultra-performance liquid chromatography–tandem mass spectrometry. J. Chromatogr. B. 2010

Normal vs. Abnormal Brain Aging



Researchers found **that low serum folate levels may increase the risk of dementia by 68%**, suggesting that its supplementation is a nutritional intervention that may play an important role in the progression of cognitive impairment and dementia.

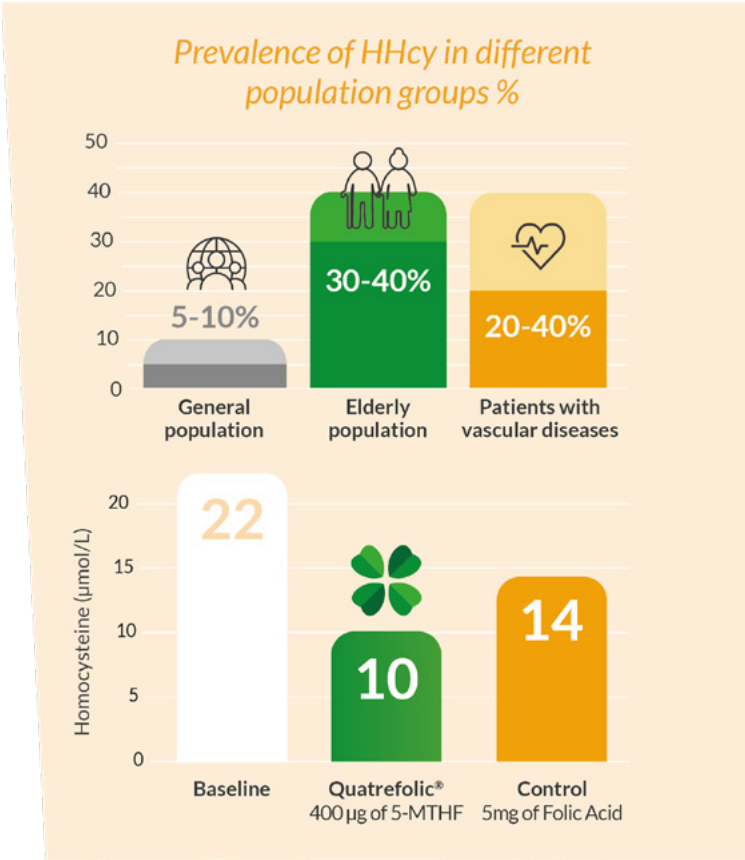
In 2016, *Mazza et al.* showed that supplementation of folate for 12 months to individuals with MCI was associated with significant improvements in global cognitive function compared to individuals in the conventional-treatment group, particularly in memory tasks.

Quatrefolic®, Homocysteine, and Cognitive Function

Hyperhomocysteinemia (HHcy) is considered a strong modifiable risk factor for both cognitive decline and dementia, and their progression in Alzheimer's disease (AD). Increased Hcy reflects the functional status of three B vitamins (folate, B12, and B6). Older people are more prone to vitamin B deficiencies due to inadequate intakes, malabsorptions, drug interactions, and/or polymorphisms.

400 µg of Quatrefolic® plus B6 and B12 was tested versus a conventional vitamin supplementation with 5 mg/day of folic acid in patients with stage 1 hypertension and serum hyperhomocysteinemia (Hcys >15 µmol/L) at low cardiovascular risk.

A normal physiologic Hcy level (i.e. less than 10 µmol/L) was reached in 55.8% of cases in the Quatrefolic® group, and it was significantly higher than in the control.

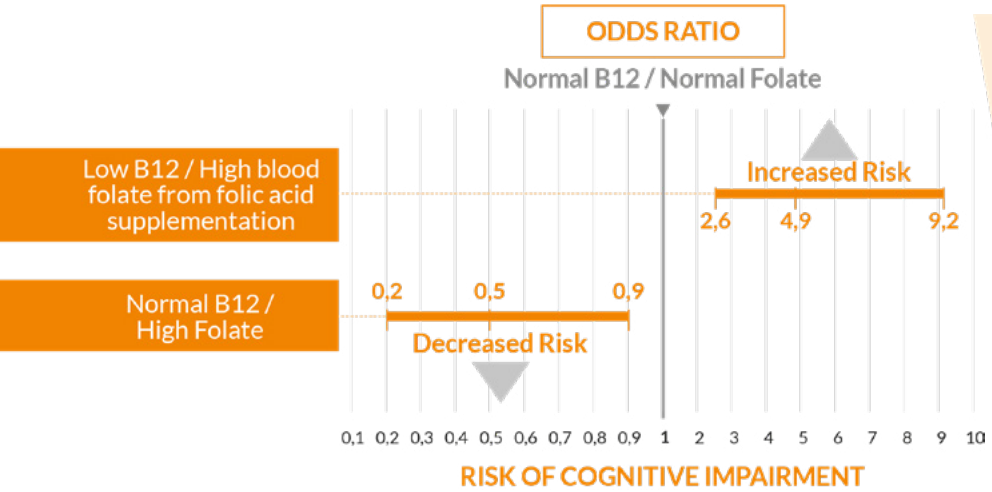


Offers tangible advantages over folic acid, especially for people with MTHFR polymorphism.

World Health Organization. May 17, 2017; Source: Nutrition Business Journal; ($mil, consumer sales);
FMGC GURU, TOP TEN TRENDS FOR 2022, JANUARY 2022

Quatrefolic® vs. Folic Acid: Masking the Vit B12 Effects

The prevalence of vitamin B12 deficiency increases with age and for those focuing on a vegetarian diet. In the elderly, it is approaching to 20% in the global population, and it is difficult to recognize.
Having the low vitamin B12 status is associated with a significantly increased prevalence of both anemia and cognitive impairment. Folate and vitamin B12 deficiency have the same hematologic symptom, megaloblastic anemia, which disappears after supplementation with large amounts of folic acid, through the stimulatory effect of unmetabolized folic acid on DNA synthesis.



The *study* found a higher prevalence of cognitive impairment in association with normal/high serum folate, from folic acid fortification, in older Americans with a low vitamin B12 status.

Adapted from Morris 2007: "Folate and vitamin B-12 status in relation to anemia, macrocytosis, and cognitive impairment in older Americans in the age of folic acid fortification"; Moore E.M. et al. J Alzheimers Dis. 2014; Morris M.S. J Clin Nutr. 2007; Selhub J. et al. Am J Clin Nutr. 2009; Smith A.D. Am J Clin Nutr. 2007

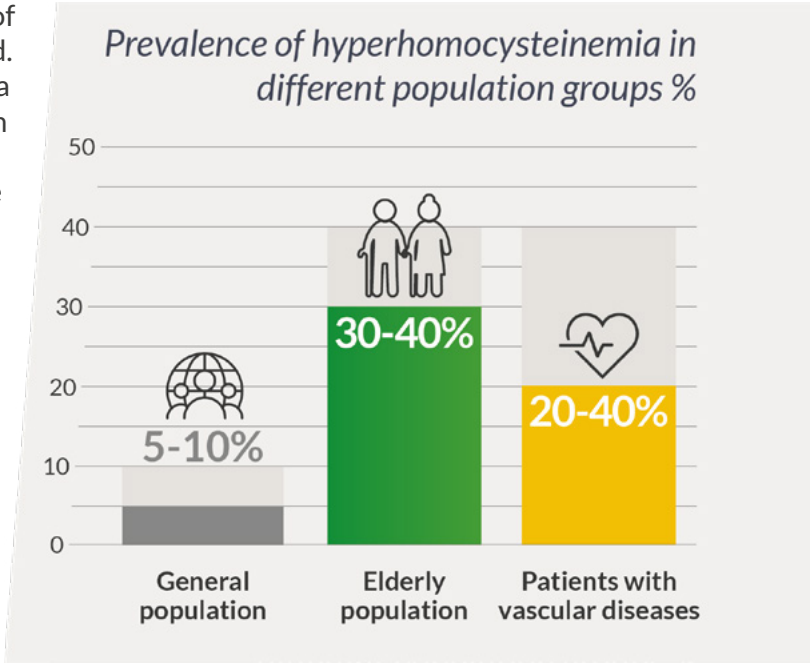
In comparison, *Quatrefolic*® is unlikely to mask the B12 deficiency because, if B12 is absent, 5-MTHF remains "metabolically trapped". This situation produces a "pseudo folate deficiency" because, although the cells have adequate levels of folate, this is trapped as the 5-MTHF form, allowing doctors to diagnose B12 deficiency.

Bailey RL, Carmel R, Green R, Pfeiffer CM, Cogswell ME, Osterloh JD, Sempos CT, Yetley EA. Monitoring of vitamin B-12 nutritional status in the United States by using plasma methylmalonic acid and serum vitamin B-12. Am J Clin Nutr. 2011;94(2):552–61. [PMC free article] [PubMed] [Google Scholar]; Rizzo G, Lagana AS, Rapisarda AM, La Ferrera GM, Buscema M, Rossetti P, Nigro A, Muscia V, Valenti G, Sapia Fet al.. Vitamin B12 among vegetarians: status, assessment and supplementation. Nutrients. 2016;8(12):767. [Google Scholar]; Moore EM, Ames D, Mander AG, et al. Among vitamin B12 deficient older people, high folate levels are associated with worse cognitive function: combined data from three cohorts. J Alzheimers Dis. 2014;39(3):661-668. doi:10.3233/JAD-131265; Morris MS, Jacques PF, Rosenberg IH, Selhub J. Folate and vitamin B-12 status in relation to anemia, macrocytosis, and cognitive impairment in older Americans in the age of folic acid fortification. Am J Clin Nutr. 2007;85(1):193-200. doi:10.1093/ajcn/85.1.193; Selhub J, Morris MS, Jacques PF, Rosenberg IH. Folate-vitamin B-12 interaction in relation to cognitive impairment, anemia, and biochemical indicators of vitamin B-12 deficiency [published correction appears in Am J Clin Nutr. 2009 Jun;89(6):1951]. Am J Clin Nutr. 2009;89(2):702S-6S. doi:10.3945/ajcn.2008.26947C; A David Smith, Folic acid fortification: the good, the bad, and the puzzle of vitamin B-12, The American Journal of Clinical Nutrition, Volume 85, Issue 1, January 2007, Pages 3–5, https://doi.org/10.1093/ajcn/85.1.3

Active Folate and Cardiovascular Health

Cardiovascular diseases (CVD) continue to be one of the main causes of mortality in the western world. Elevated plasma Hcy concentration is considered a risk factor for CVD and may also be associated with hypertension.
Not surprisingly, heart health has become one of the top five concerns of supplement consumers, second only to general health. The search for solutions to prevent and reduce risks has led to new discoveries about the factors that aggravate CVD.

In 2022, FMGC GURU reported that 53% of the consumer's panel they studied claimed they would take a proactive approach to heart health in 2022. Several studies demonstrated that the mild hyperhomocysteinemia (HHcy) is an independent risk of factor of cardiovscular health.



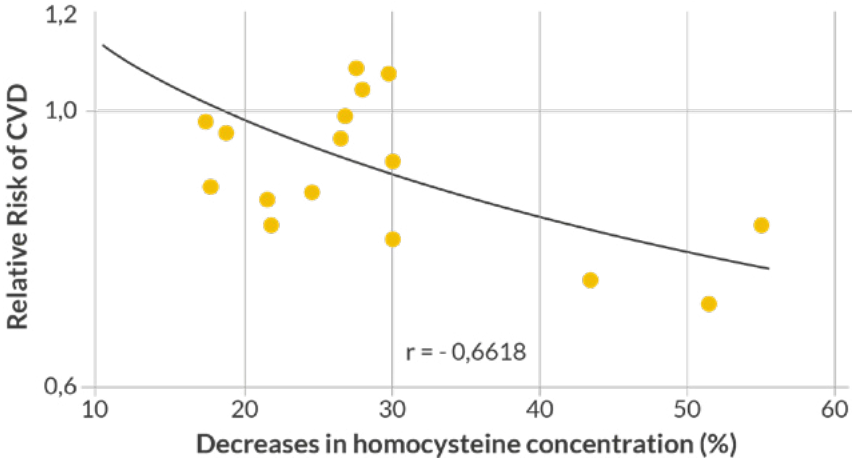
World Health Organization. May 17, 2017 Source: Nutrition Business Journal ($mil, consumer sales); FMGC GURU, TOP TEN TRENDS FOR 2022, JANUARY 2022; Selhub J. et al. JAMA 1993; Peng H. et al. J Zhejiang Univ Sci B. 2015

The Interconnection between Folate, Hcy and CVD

Extensive researches are trying to identify strategies to promote *heart health* and prevent the related risk factors for cardiovascular disease. Of particular interest is the role of folate and homocysteine (Hcy), causally related to Coronary Heart Disease (CHD) and stroke as reported by many observational *studies*. Indeed, the evidence was deemed strong enough to recommend folate supplements as part of a healthy lifestyle.

Hcy is a common amino acid found in the bloodstream and produced as a metabolite of methionine metabolism in the one carbon cycle. The plasmatic levels of Hcy are predictive for cardiovascular risk and determined by several factors (lifestyle, genetics and diet).

Relation between homocysteine reduction and relative risk of CVD (significant dose-response association)



Hcy is responsible for 10% of the total risk for atherothrombotic vascular disease; furthermore, a meta-analysis has highlighted that each increase of 5 µmol/L in homocysteine level raises the risk of CHD events by approximately 20%, independently of traditional CHD risk factors.

Adapted from Li et al. 2016.

Normal
5-15 µmol/l

Mild
15-30 µmol/l

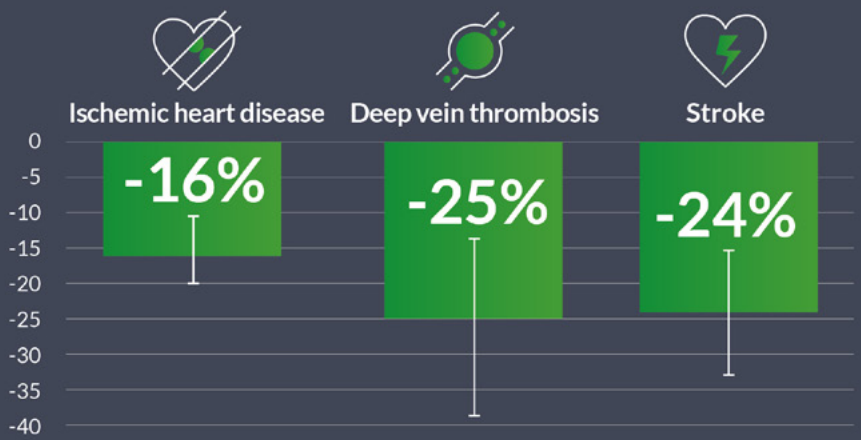
Moderate
30-100 µmol/l

Severe
>100 µmol/l

The Classification of Homocysteinemia

Meta-analysis (72 studies) has demonstrated significant associations between blood total homocysteine and the risk of ischemic heart disease, deep vein thrombosis & pulmonary embolism, and stroke. **It was estimated that lowering blood Hcy by 3µmol/L would reduce an individual's risk of ischemic heart disease by 16 %, deep vein thrombosis by 25 %, and stroke by 24 %.**

Decrease in risk per 3 µmol/L reduction in blood total homocysteine (%)



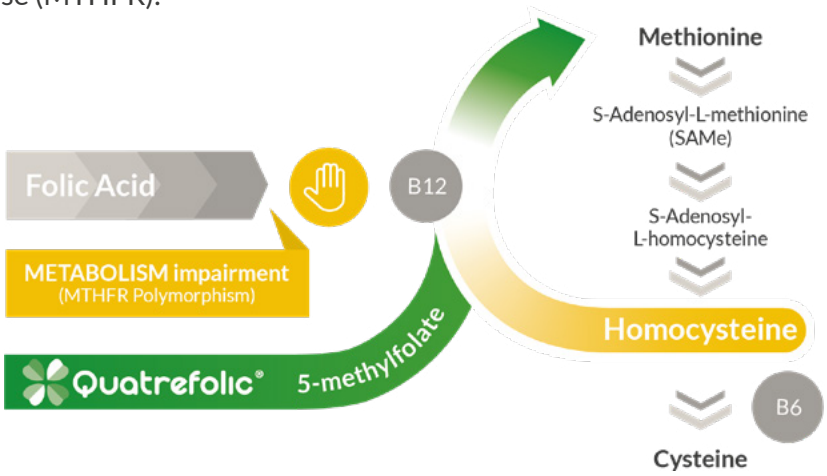
Adapted from Wald D.S. et al. BMJ 2002

Smith G.D., Ebrahim S. The Lancet. Vol. 366, Issue 9498. 2005; Li Y. et al. J Am Heart Assoc. 2016; Clarke R et al. Arch Intern Med. 2010; Toole J.F. et al. JAMA 2004; Blom H.J. et al. Nat Rev Neurosci. 2006; Homocysteine Studies Collaboration. JAMA 2002; Wald D.S. et al. BMJ 2002.

Quatrefolic® and Heart Support

Folate is an important regulator of Hcy metabolism and helps to maintain Hcy normal plasma concentrations. Clinical studies report evidence that folate supplementation can support cardiovascular health.
The most common cause of genetic hyperhomocysteinemia (HHcy) is the 677CT gene polymorphism of the enzyme methylenetetrahydrofolate reductase (MTHFR).

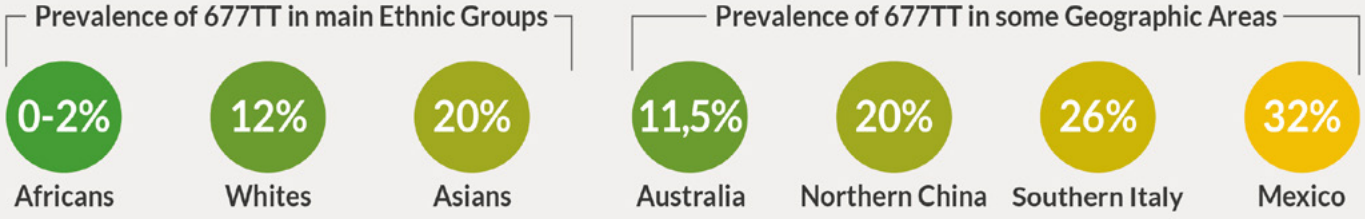
The MTHFR polymorphism[G7] is responsible of the reduced activity in the metabolization of folate in 5-MTHF, that works as a methyl donor for homocysteine remethylation to regenerate methionine in the one carbon metabolism cycle.
Low 5-MTHF in turn leads to an increase of Hcy.



> ***Quatrefolic®*** **completely bypasses the impaired MTHFR conversion step, overstepping the problem of people with polymorphisms enzyme, and consequently is a valid healthy option to control Hcy levels and support heart health.**

Hcy and the 677CT Polymorphism of the MTHFR Enzyme

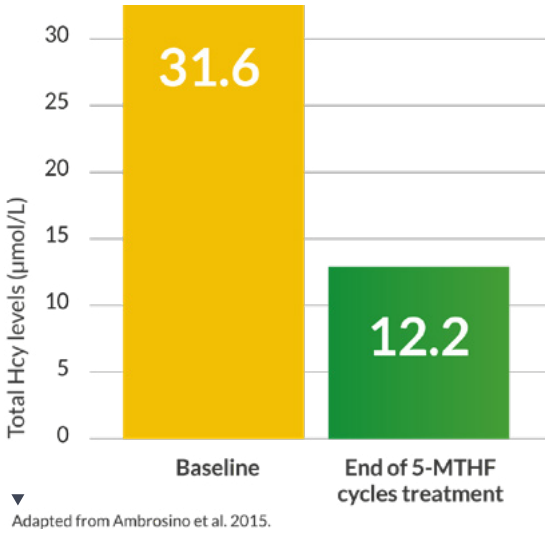
It is estimated that with homozygous mutation of MTHFR, up to 70% of the enzyme function is lost; and with heterozygous mutation, there is a 35% loss of enzymatic function. In these subjects the immediate use of the bioavailable form of folate 5-MTHF Quatrefolic® is a tangible benefit. Subjects with homozygotes polymorphism have 25% higher homocysteine levels (about 2,5 µmol/l) due to the reduced enzyme activity.



World variability of the prevalence of MTHFR polymorphism (homozygous 677TT genotype) ***among different ethnic groups and regions:*** *the incidence is higher in Mediterranean countries, Latin Americans and lower in African-Americans*

Ambrosino et al. clearly shows that a cyclic schedule (1 month of therapy followed by 2 months of withdrawal) of 5-MTHF supplementation is able to significantly reduce tHcy levels in patients with mild/moderate HHcy.

> **During the entire study period, there was a reduction of tHcy levels from 31.6 to 12.2 µmol/L. The repeated folate cycles normalized levels in 86.8% of patients treated.**



Adapted from Ambrosino et al. 2015.

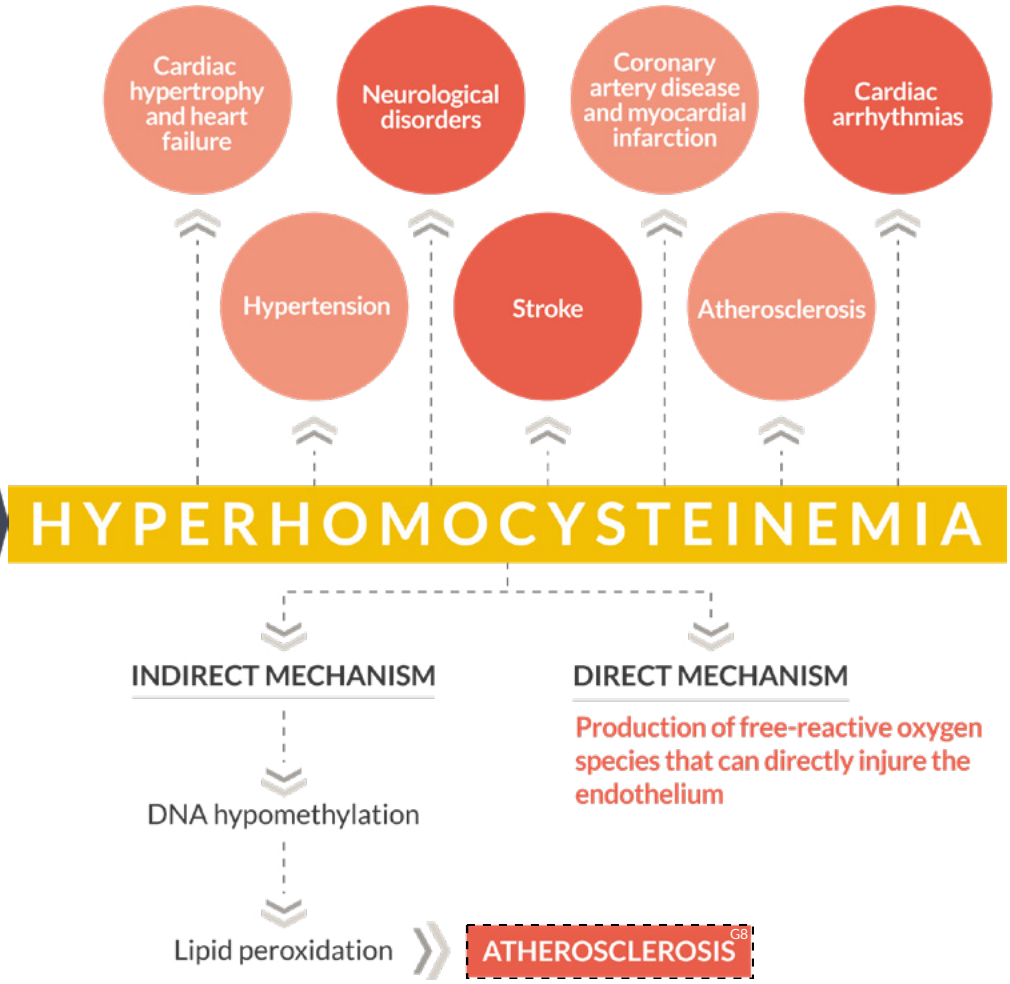
Quatrefolic®, already in the form of active folate, may be a valid nutritional support for people and that can support heart health

Ilhan N. et al. Arch Med Res. 2008; Kiseljaković E. et al. Bosn J Basic Med Sci. 2008; Wilcken D.E. J Cardiovasc Risk. 1998; Ambrosino et al. Nutr Res. 2015; Miller A.L.Altern Med Rev. 2003; Resfum M.D., Ueland P.M. Annu Rev Medicine 1998; Caruso A. et al. J Cardiovasc Pharmacol 2006; Scott J.M., Weir DG. J Cardiovasc Risk. 1998

Behind the mechanisms of Homocysteine

The mechanisms by which high levels of homocysteine exert disease effects may be either indirect or direct. In the first case, Hcy is a simple marker; in the second case, it is a true risk factor of cardiovascular disease. Possible mechanisms by which homocysteine causes vascular injury include *endothelial injury*, DNA dysfunction, proliferation of smooth muscle cells, increased oxidative stress, reduced activity of glutathione peroxidase and promoting inflammation.

Risk factor and molecular mechanisms of homocysteine induced cellular dysfunction



New Potential Benefits of the Active Folate Quatrefolic®

The impact of folate administration in patients with cardiovascular disease does not end with the reduction of Hcy but involves wider effects that lead to an improvement in endothelial function and oxidative stress. The mechanisms responsible for these effects include direct antioxidant properties of 5-MTHF and an interaction with the nitric oxide synthase enzyme (eNOS).

> ***Stanhewicz et al.*** **have recently published a review highlighting the role of the active folate 5-MTHF in the cascade of events leading to increased production and bioavailability of nitric oxide (NO) at the endothelial level.**

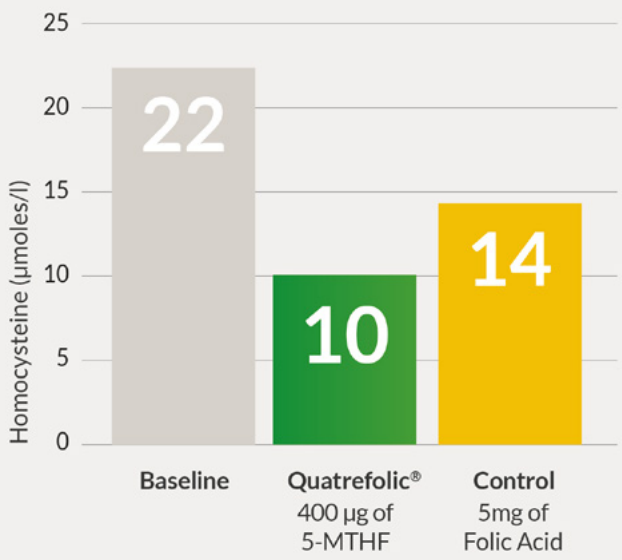
Endothelial Dysfunction: The Link Between Homocysteine and Hydrogen Sulfide (nih.gov); Stranger O. et al. Clin Chem Lab Med. 2003; Boushey C.J. et al. J. Am Med Assoc. 1995; Ganguly P. et al. Nutr. J. 2015; Djuric D. et al. Indian J Chest Dis Allied Sci. 2008; Cacciapuoti F. Ann Vasc Med Res. 2017; Stanhewicz A.E., Kenney W.L. Nutr Rev. 2017; Pushpakumar S. et al. Curr Med Chem. 2014.

Quatrefolic® and Hcy Levels

The homocysteine-lowering and -normalizing effect of Quatrefolic®, in comparison with high dosage of folic acid in hypertensive subjects at low cardiovascular risk, has been tested by *Mazza et al in 2016.*
400 µg of Quatrefolic® plus B6 and B12 was tested versus a conventional vitamin supplementation with 5 mg/day of folic acid, in patients with stage 1 hypertension and serum hyperhomocysteinemia (Hcys >15 µmol/L) at low cardiovascular risk.

The results of Quatrefolic® group showed a significant Hcys reduction in comparison with baseline from 21.5 µmol/L to 10.0 µmol/L versus the control group (highly dosed folic acid).

The ideal Hcys level (i.e. less than 10 µmol/L) was reached in 55.8% of cases in the Quatrefolic® group, and it was significantly higher than in the control one.



Quatrefolic® offers tangible advantages versus folic acid and can promote healthier heart life, especially for people with MTHFR polymorphism.

Epidemiological data - the Importance of Reduced Folate

Recently, a *randomized study* that included 20,702 adults without a history of stroke or myocardial infarction (MI) has revealed that the combined use of the hypertension medication enalapril and folic acid, compared with enalapril alone, significantly reduced the risk of first stroke among Chinese adults with hypertension.
Participants were stratified by variations in the polymorphism of the enzyme methylenetetrahydrofolate reductase (MTHFR) that affects folate levels.

The protective effect of folic acid was not the same in the different genotypes.
In patients with polymorphism (homozygous mutation) the preventive effect of folic acid therapy was lost, and the group had the highest stroke rates.

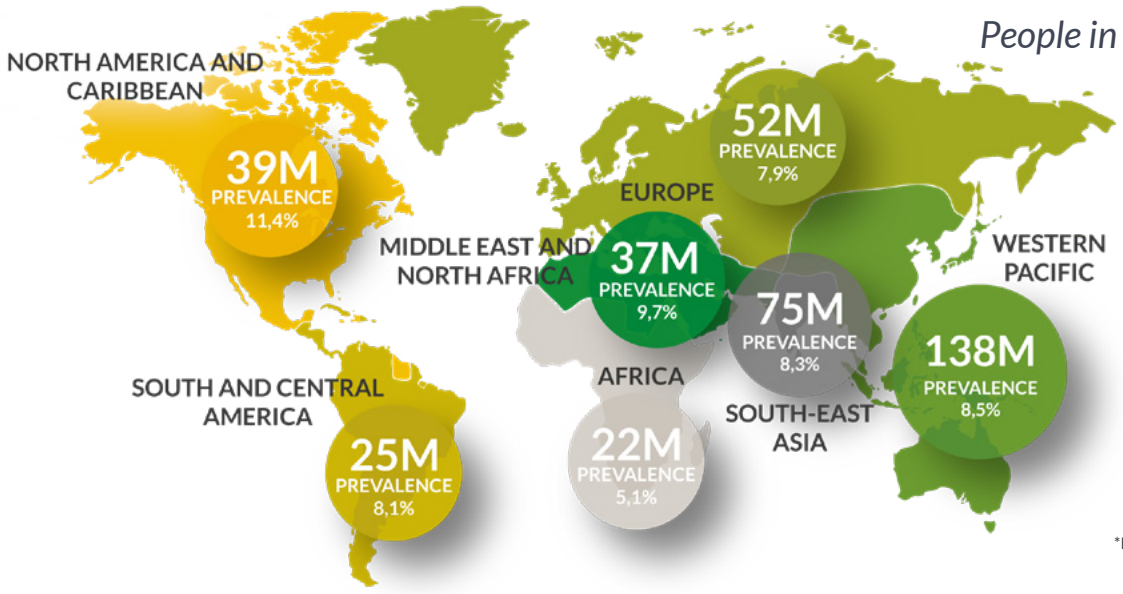
> **In these subjects, the supplementation of right folate such as *Quatrefolic*® bypasses the reduction of folate metabolism and, hence, lower circulating levels of folate.**

Mazza et al. Biol Regul Homeost Agents. 2016; Huo Y. et al. JAMA 2015

Active Folate and Glucose Metabolism

The global prevalence of **IGT (Impaired Glucose Tolerance)** was estimated at 7.3% of the adult population in 2017, equivalent to 352.1 million individuals. By 2045 the pervasiveness is anticipated to increase to **8.3% of the global adult population, equivalent to an estimated 587 million individuals.**
Prediabetes is a condition that can be considered an early, yet potentially reversible stage of the development of type 2 diabetes (15-30% of people with prediabetes will develop type 2 within 5 years).
Type 2 diabetes mellitus (T2DM) is on the rise worldwide. More than 400 million people were living with diabetes as of 2015* and 95% of people around the world that have Type 2 diabetes mellitus.**
The many risk factors for this type of diabetes include lifestyle decisions that result in excess body weight and physical inactivity. Consistent evidence shows that the **risk of developing type 2 diabetes mellitus** can be reduced by **adhering to a healthy diet** with the **right level of vitamins such as folate and regular physical activity.**

People in the world living with diabetes



*International Diabetes Federation
** World Health Organization

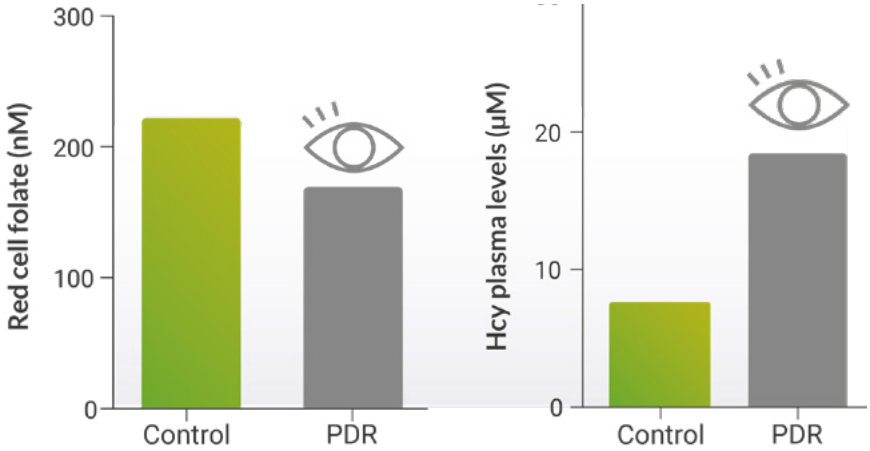
Folate's Interconnection with Glycemic Control

There is a growing interest in the considerable benefits of the glycemic profile.

In particular, folate supplementation (vitamin B9) might be beneficial for glucose homeostasis and lowering, insulin resistance (IR) with a possible role in diabetes deterrence, and reducing the risk of T2DM, and some diabetic conditions associated with it.

Recent studies have shown that circulating folate concencentrations are significantly lower in T2DM patients compared with healthy individuals.

Adapted from Malaguarnera et al. Clin Ophthalmol. 2015



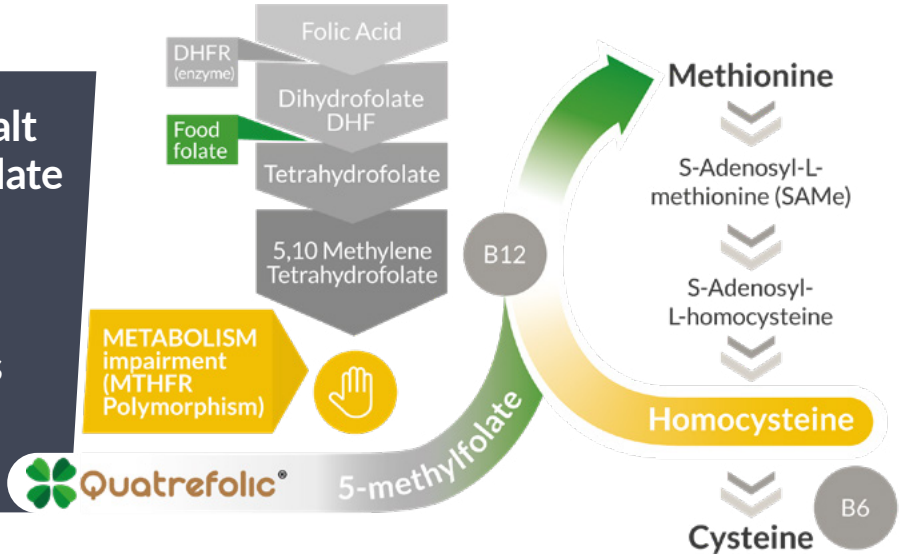
Lower levels of red blood cell folate, and increased Hcy level, were observed in patients with T2DM especially with complications (PDR, proliferative diabetic retinopathy).

In a large prospective *cohort study (2020)*, folate intake (including both dietary and supplemental resources) was inversely associated with the incidence of diabetes among American adults during a 30-year follow-up. Presumably, this inverse association is explained by the pathways of folate with mechanisms related to homocysteine (Hcy) levels, IR, and systemic inflammation.
***Folate is crucial to one-carbon metabolism*, a specific pathway that is essential for cellular function, amino acid metabolism, both DNA and RNA biosynthesis, DNA methylation, and methylation reactions.**

FOCM, Folate, and Diabetes

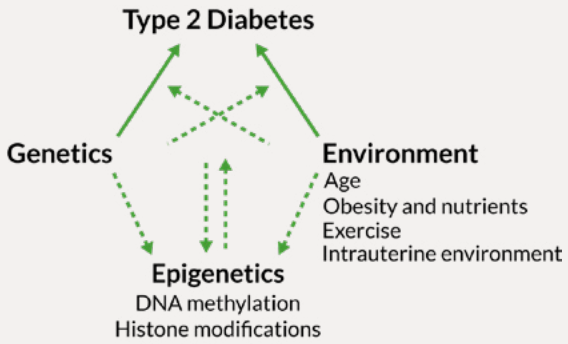
Folate enters the methionine cycle in the active form of 5-methyltetrahydrofolate (5-MTHF or methylfolate). **Both food folate and folic acid need to be transformed to this active folate through a multistep enzymatic conversion, where the enzyme MTHFR plays a key role.** MTHFR polymorphism prevalence in the general population is approximately 40%. Individuals can have low availability of 5-MTHF, independent of folic acid supplementation, with potential accumulation of Hcy in the blood, causing increased oxidative stress and vascular damage.

Quatrefolic®, glucosamine salt of the 5-methyltetrahydrofolate (5-MTHF or methylfolate), represents a natural-based approach to help manage glycemic problems, diabetes progression, and diabetes complications.



Asbaghi O et al. Nutrients. 2021; Malaguarnera G. et al. Clin Ophthalmol. 2015; Genquan JMD et al. Asia Pac J Clin Nutr 2021; Kido Y. Diabetology International. 2017; Hayden MR et al. Medicina (Kaunas). 2021; Galicia-Garcia U at al. Int J Mol Sci. 2020; Eguchi, N. et al. Int. J. Mol. Sci. 2021; Shang Y et al. J Intern Med. 2019; Ling C et al., Diabetes. 2009.

Folate's Benefits on Glucose Levels



Adapted from Ling et al. Diabetes. 2009

T2DM is a metabolic disease characterized by high blood glucose, insulin resistance, and a relative lack of insulin. It is also associated with inflammatory profiles and altered DNA methylation patterns. Coexisting disorders, including obesity, hypertension, and dyslipidemia, contribute to the severity of T2DM.

This metabolic disease is characterized by a combination of two primary factors: **defective insulin secretion by pancreatic β-cells and the inability of insulin-sensitive tissues to respond appropriately to insulin.**

Defects in any of the mechanisms involved in these processes can lead to a metabolic imbalance responsible for the development of the disease.
Before people develop T2DM, they almost always have prediabetes—where blood glucose levels are higher than normal, but not yet high enough to be diagnosed as diabetes.

Early nutritional treatment as well as moderate lifestyle changes can actually return blood glucose (blood sugar) levels to a normal range, effectively preventing or delaying T2DM.

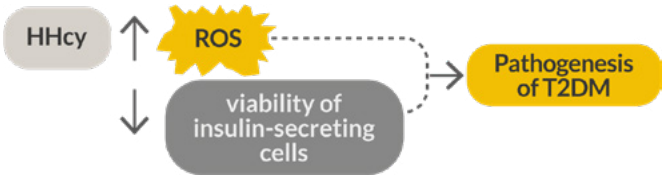
Benefits of folate supplementation

- ↑ **Ameliorate DNA methylation**
- ↑ **Improve glycemic control**
- ↑ **Improve endothelial function**
- ↓ **Decrease inflammation and oxidative stress**
- ↓ **Reduce fasting blood glucose**
- ↓ **Reduce serum insulin**
- ↓ **Decrease insulin resistance**

Behind the mechanisms of T2DM and Folate

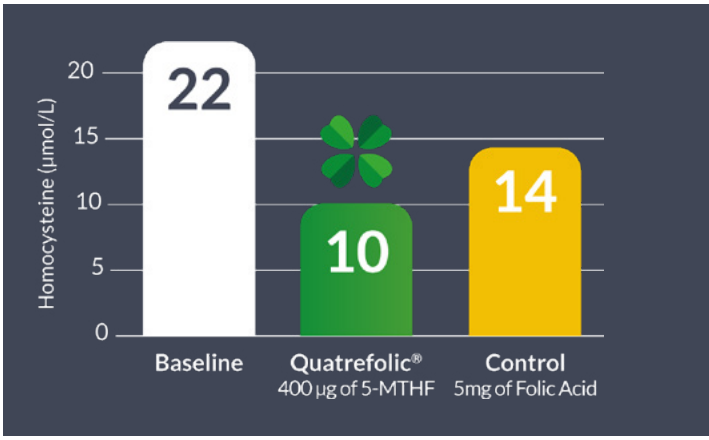
Hcy-Dependent Pathway

Folate deficiency contributes to elevated Hcy (HHcy) concentrations, a recognized risk factor for the development of cardiovascular disease and T2DM. In T2DM development, acute and prolonged exposure to Hcy has adverse effects on the cell viability of pancreatic β-cells.



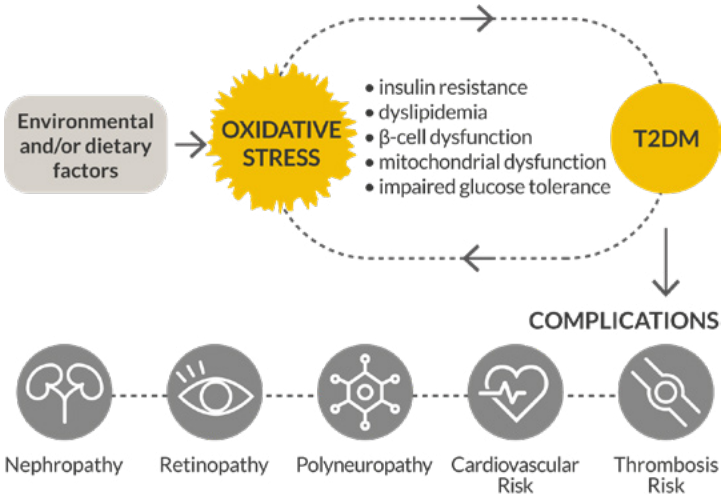
MTHFR enzyme is responsible for converting dietary folate into reduced active folate. Consequently, this enzyme is crucial for maintaining adequate levels of Hcy. MTHFR polymorphisms that affect this conversion may hence affect Hcy concentration. Although the results are still conflicting, a meta-analysis published in 2013 has investigated the **associations of MTHFR-linked Hcy and T2DM, providing evidence that polymorphic TT genotype of MTHFR C677T contributes to susceptibility to T2DM**, and supported the hypothesis that elevated Hcy is causally related to increased risk of T2DM.

Folate supplementation is widely accepted and recommended to any patient/person who has an elevated Hcy level for its normaliazing effect.
The efficacy of Quatrefolic® (400 mcg plus B6 and B12) versus a conventional high-dose folic acid supplement (5 mg/day) has been evaluated in hypertensive subjects at low cardiovascular risk (104 patients with Hcys ≥15 μ mol/L).
The *treatment* was significantly effective with the ideal Hcys level reached in 55.8% of cases in the Quatrefolic® group, and it was significantly higher than in controls.



Hcy-Dependent Pathway

Pancreatic β-cells are particularly susceptible to oxidative stress due to their high endogenous production of reactive oxygen species (ROS) and their low antioxidant capacity, suggesting that oxidative stress may play an important role in β-cell failure and death. If β-cell functioning is impaired, it results in the underproduction of insulin, fasting hyperglycemia, and eventually the development of T2DM.
On the other side, chronic exposure to a glucose-rich environment and consequent hyperglycemia can induce oxidative stress[G9], creating several physiological and pathophysiological changes, exacerbating its toxic effect on cells, tissues, and organ systems.
As T2DM begins to develop, the body becomes less sensitive to insulin and the resulting IR also leads to inflammation. A vicious cycle can result, with more inflammation causing more IR and vice versa.



Folate is a well-known antioxidant molecule protecting cells from oxidative degradation. Additionally, the actions of folate in DNA methylation, repair, and synthesis processes may influence the inflammatory phenotype via epigenetic changes and modulation of cell proliferation.

Globally, folate supplementation seems to be able to significantly decrease fasting blood glucose, serum insulin, and insulin resistance.

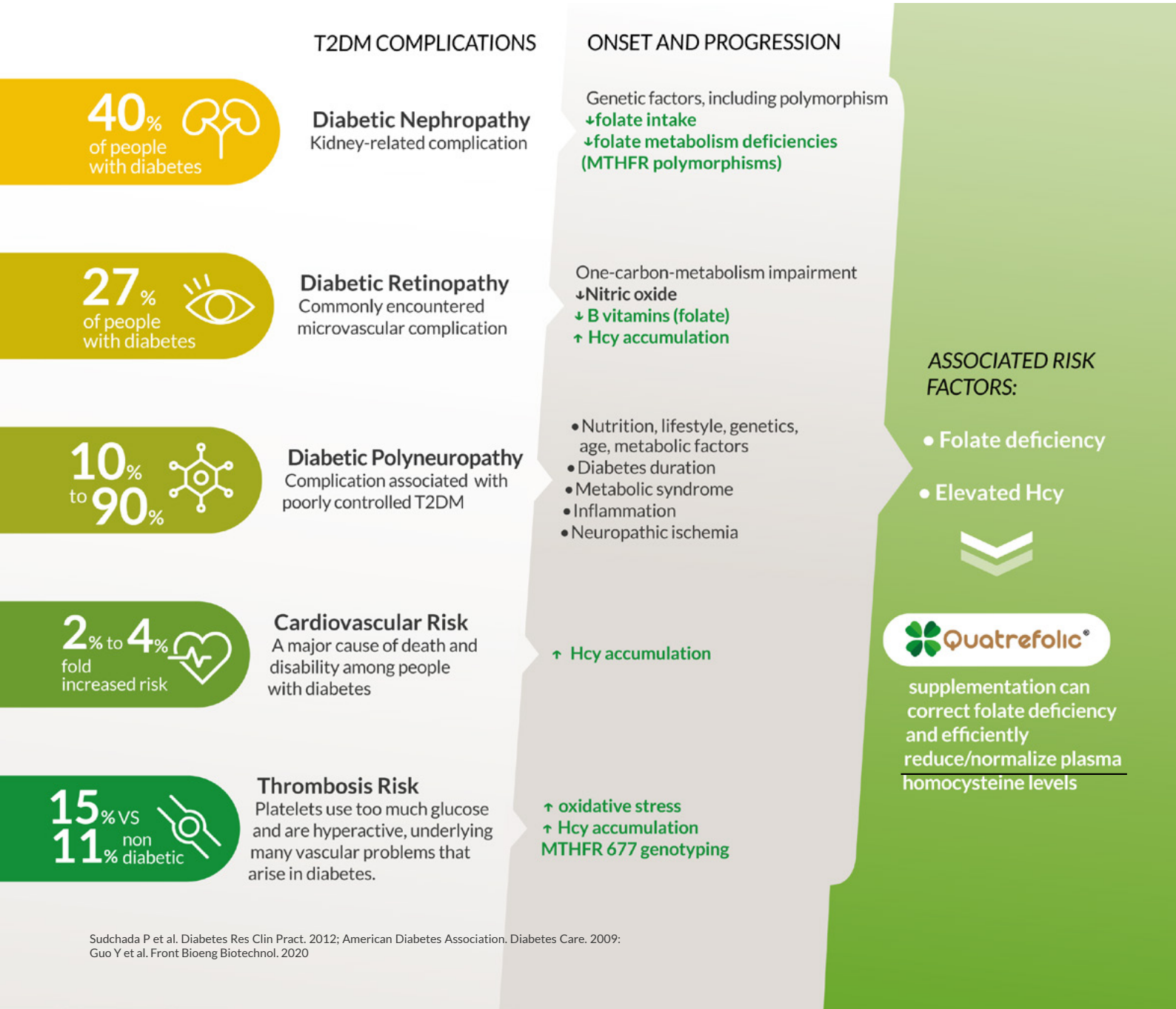
Cheng Y et al.Front Cardiovasc Med. 2022; Huang T et al. BMC Genomics. 2013; Eguchi, N. et al. Int. J. Mol. Sci. 2021; Tangvarasittichai S. World J Diabetes;

T2DM and Complications: How Folate Can Help

Chronic hyperglycemia associated with diabetes can result in end-organ dysfunction and failure, and will trigger chronic metabolic syndrome, including obesity, cardiovascular disease, retinopathy, nephropathy, dyslipidemia, and hypertension. T2DM now represents a coronary heart disease risk, and nearly 80% of diabetic mortality is the result of diabetes-induced cardiovascular disease.

The increased risk of CVD in T2DM patients is associated with high Hcy levels that promote endothelial dysfunction and increase the likelihood of atherosclerosis development.
T2DM with high Hcy levels are associated with increased incidence of venous thrombosis.

> **The antioxidant and anti-inflammatory properties of folate - in particular of Quatrefolic®, biological active form 5-MTHF not dependent on MTHFR action for activity – can contribute to combating the inflammation and the vicious cycle of T2DM-related disease aggravation. Specifically, folate can promote micro-vessel (diabetic retinopathy) and nerve growth (diabetic polyneuropathy).**



Sudchada P et al. Diabetes Res Clin Pract. 2012; American Diabetes Association. Diabetes Care. 2009:
Guo Y et al. Front Bioeng Biotechnol. 2020

Active Folate and Anemia

Lifetime Support

Anemia is a global public health problem, affecting 571 million women and 269 million young children worldwide.

It is a serious global public health problem that particularly affects young generations, menstruating adolescent girls and women, and pregnant and postpartum women. WHO estimates that 40% of children 6–59 months of age, 37% of pregnant women, and 30% of women 15–49 years of age worldwide should be anemic. It is more prevalent in low - and middle - income countries.

The WHO Global Nutrition Target 2025 aims to reduce anemia in women of reproductive age by 50% by 2025.

Prevalence of anemia in women of reproductive age (15-49)

Risk Factors
Certain factors can increase the risk for anemia, which includes a poor diet, intestinal problems such as celiac disease and Crohn's disease, menstruation and menopause, pregnancy, chronic disease - cancer and kidney failure, genetic factors, alcoholism, and exposure to toxic chemicals.

Symptoms
The signs and symptoms of anemia differ depending on the causes. Symptoms of anemia get worse as the condition becomes more severe.

www.who.int/data/gho/data/themes/topics/anaemia_in_women_and_children

The Different Kind of Anemia – Role of Folate

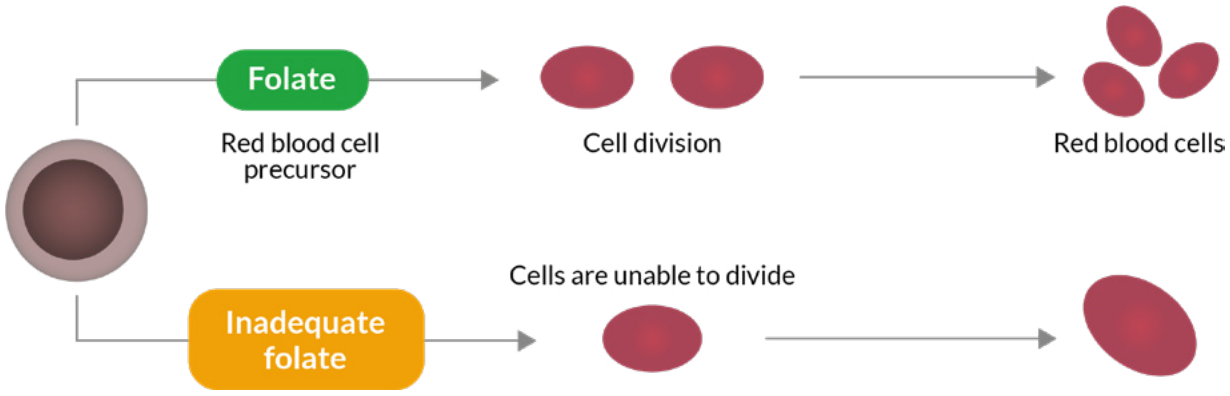
Anemia is a decrease in circulating red blood cells or hemoglobin, insufficient to meet physiologic needs. This leads to a consequent decrease in the amount of oxygen delivered to tissues.

The optimal hemoglobin concentration required to meet physiologic needs varies by age, sex, residence elevation, smoking habits, and pregnancy status. The causes of nutrient deficiencies include **inadequate dietary intake**, **increased nutrient losses** (blood loss from parasites, bleeding associated with childbirth, heavy menstrual losses), or **altered nutrient metabolism.**

Among the various types of anemia, the most common cause of anemia includes nutritional deficiencies, where concentrations of hematopoietic nutrients involved in Red Blood Cell production or maintenance – iron and vitamins - are insufficient to meet those demands.

Iron deficiency is the most common cause of anemia associated with **folate or vitamin B12 deficiencies, representing approximately 12% of all anemia.**

In folate-deficiency anemia, the red blood cells are abnormally large. Such cells are called macrocytes. They are also called megaloblasts when they are seen in the bone marrow. That is why this anemia is also called megaloblastic anemia.



Macrocytes typically carry oxygen but are not as pliable as normal red blood cells (RBCs), and their lifespan is decreased by 30-50%. The flexibility of RBCs is essential to pass through small capillaries during microcirculation.

The appearance of substantial red blood cells marks megaloblastic anemia. This disorder is caused by the incomplete formation of red blood cells, resulting in large numbers of immature and incompletely developed cells.

Folate plays a key role: without enough folate in the body, the number of red blood cells will be depleted and lead to anemia.

Deficiency of vit. B12 and folate → Diminished DNA synthesis → Failure of nuclear maturation → Proliferation of erythroblast fail → Macrocytes → Rapidly destructured → ANEMIA

Crucial Role of Folate in Anemia in Pregnancy

Recent studies show a high prevalence of anemia among pregnant women worldwide. The prevalence of anemia in the third trimester was higher than in the first and second trimesters. Anemia in pregnant women in developing countries is significantly higher than in developed countries due to pregnancy's economic, sociological, and health factors.

Pregnant women need more oxygen to support the growing baby and they are more likely to get iron-deficiency *anemia* during pregnancy.

Iron and folate deficiency are among the most common conditions among gynecological patients of reproductive age and in pregnant women. Low folate and iron levels can lead to several diseases, including pregnancy complications, fetal malformations, and delayed mental development of the neonate.

A recent *Russian study* confirms that the right combination of these components — active folate and iron — can make up for fundamental micronutrient deficiencies in women of reproductive age. Researchers assessed clinical and laboratory efficacy (the absence of objective anemia signs and the changes in hemoglobin level, folate, and ferritin blood tests over time) and tolerability (the rate of adverse events, tolerability of the taken product evaluated using the visual analog scale) at days 30, 60 and 90 after the beginning of treatment.

The result confirms maintaining iron and active folate, such as Quatrefolic®, homeostasis by oral intake during three months in the preconception period and early stages of pregnancy is a feasible option based on its high efficacy and good tolerability with the minimal risk of side effects.

Studies compared iron-folate supplementation* versus placebo or no supplementation during pregnancy.

Folate supplementation is recommended in pregnant women with iron-deficiency anemia, irrespective of the serum levels of folate.

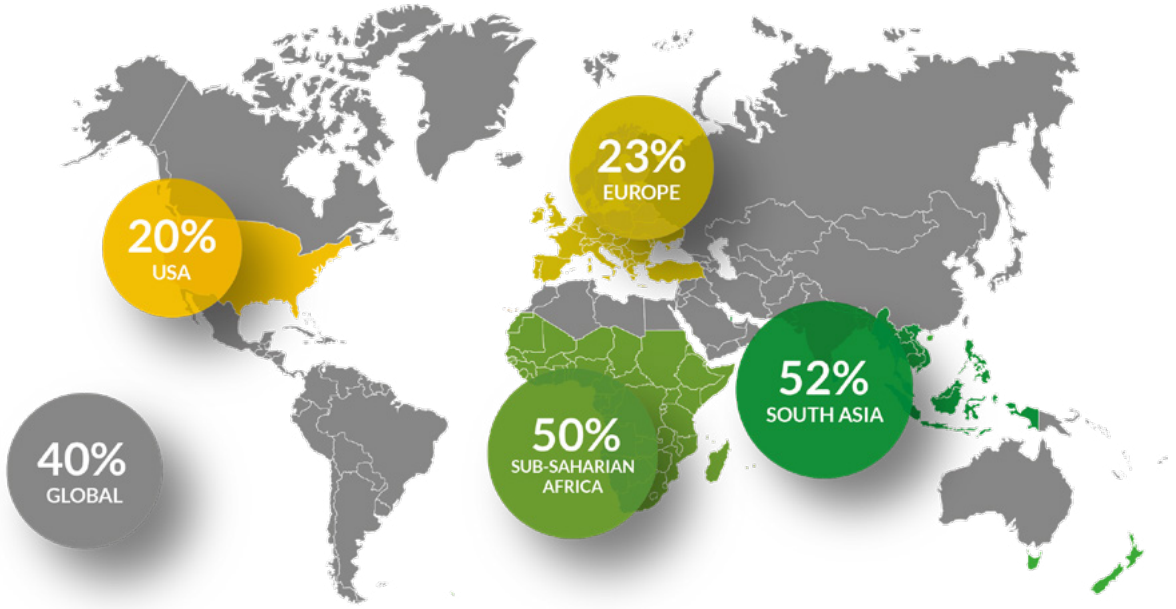
Iron and folate work together for the production of healthy Red Blood Cells that function properly, guaranteeing hemoglobin production and oxygen transport within the body.

- **Reduced maternal anemia by 69%**
- **Reduced risk of premature birth**
- **Greater mean birth weight**

Folate has been recognized by the European Food Safety Authority (EFSA) for its beneficial effects in supporting normal blood formation and in the reduction of fatigue and tiredness.

Anemia and Children - Folate Support

An estimated 600 million children worldwide suffer from anemia, which impairs physical and cognitive development and weakens the immune system. Anemia seriously affects child growth, development, and survival, and is associated with poor cognitive and motor development.



Many children in low- and middle-income countries have limited access to iron-rich foods, but rigorous research shows that weekly iron and **folic acid (IFA) supplementation is highly effective in reducing iron-deficiency anemia.**

Inexpensive, safe, and recommended by the World Health Organization, IFA supplementation has been shown to reduce the risk of anemia in children by 49%.

Folate as Quatrefolic® and Iron – the Importance of the Combination

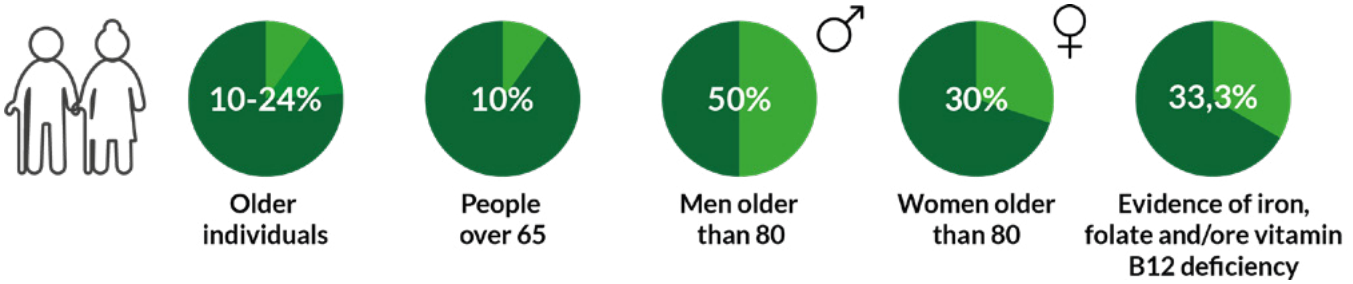
Rigorous research shows that intermittent iron and folic acid (IFA) supplementation is highly effective at reducing iron-deficiency anemia:

- **A meta-analysis from trials conducted in 20 countries** in Latin America, Africa, and Asia shows that intermittent supplementation **reduces the risk of anemia by 49% among children aged 0-12 years.**
- **A large-scale *cohort study*** in India combined weekly IFA supplementation with counseling and deworming for adolescent girls aged 10-19 years and found a **decrease in anemia rates from 73% to 25%.**
- **A *pilot project*** in Vietnam combined weekly IFA and deworming tablets for women aged 15 to 45. The prevalence of **iron-deficiency anemia fell from 18% at baseline to 3% at 12 months**, and remained at 4% at 54 months, confirming this condition had essentially been eliminated in this population.

Anemia and the Elderly: Why Quatrefolic® doesn't mask vitamin B12 deficiency

Vitamin B12 deficiency is common in older adults and may be hard to recognize.

Folate and *vitamin B12 deficiency* have common symptoms caused by either folate or vitamin B12 deficiency. The most common hematologic symptom is megaloblastic anemia, which disappears after supplementation with large amounts of folate, particularly in the elderly, where vitamin B12 deficiency remains undiagnosed.



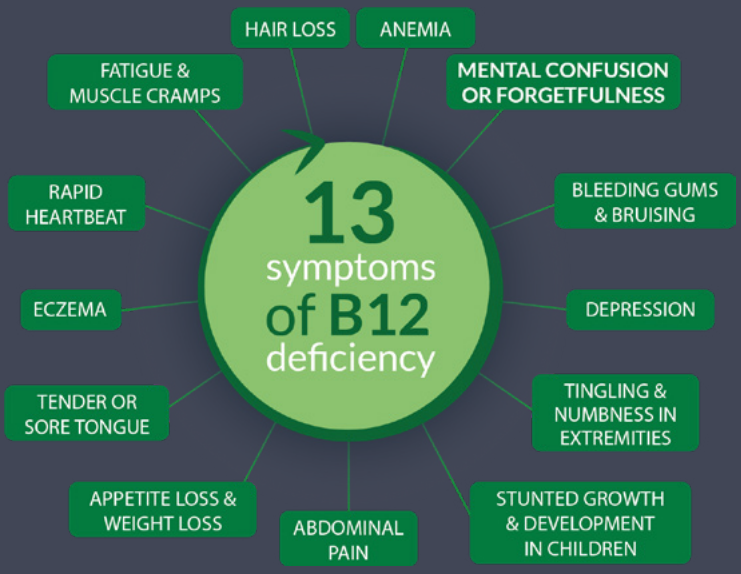
Anemia in older persons is common and relevant, with a complex etiology. Anemia will likely increase dramatically in the coming years because of an aging population in Western societies.

The National Institute of Health has found that large amounts of folic acid can mask the damaging effects of vitamin B12 deficiency. This means that current blood tests may not reveal the actual level of B12. Furthermore, folic acid can correct anemia caused by vitamin B12 deficiency without correcting the neurological damage that also occurs due to the vit B12 deficiency.

Concerns have been raised that high intake of folic acid from fortified food and dietary supplements might mask the macrocytic anemia of vitamin B12 deficiency, thereby eliminating an important diagnostic sign while allowing the progression of neurological symptoms.

Quatrefolic® supplementation is unlikely to mask vitamin B12, resulting in tangible advantages and a safer profile than folic acid.

Supplementation with the active form of folate, 5-MTHF, may be an interesting alternative to folic acid in vitamin-B12 deficient individuals as its metabolic pathway is vitamin-B12 dependent. It is also noted that in comparison to folic acid, 5-MTHF would be unlikely to mask the clinical symptoms of vitamin-B12 deficiency.



Chaparro CM, Suchdev PS. Anemia epidemiology, pathophysiology, and etiology in low- and middle [1] income countries. Ann N Y Acad Sci 2019; 1450:15–31 • WHO/CDC. Assessing the iron status of populations: Including Literature Reviews. WHO/CDC. Assessing the Iron Status of Populations: Including Literature Reviews. WHO/CDC; Geneva, Switzerland: 2004. p. 112: Geneva, Switzerland, 2004. • Metz J. A high prevalence of biochemical evidence of vitamin B12 or folate deficiency does not translate into a comparable prevalence of anemia. Food Nutr Bull 2008; 29:S74-85; • Socha DS, DeSouza SI, Flagg A, Sekeres M, Rogers HJ. Severe megaloblastic anemia: Vitamin deficiency and other causes. Cleve Clin J Med 2020; 87:153–64; • Smelt et al. 2018 (20) Meta-analysis; Zec et al. 2020 ; Willoughby et al. 1967; Lassi et al. 2013 ; Mahomed 2006; • Quatrefolic® and iron form, Lipofer® article review: RMJ. • Mother and child. T. 5,nº 1, 2022 / Russian Journal of Woman and Child Health. Vol. 5, nº 1, 2022 • www.evidenceaction.org/programs/equal-vitamin-access

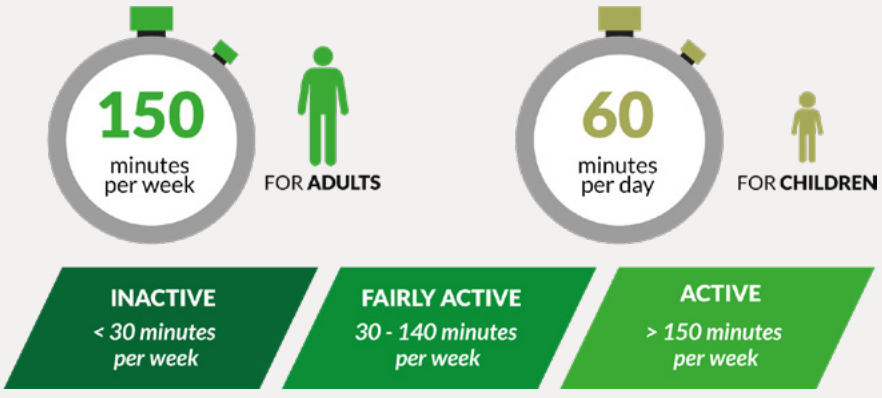
Active Folate and Sports

The Importance of Physical Activities

Since the COVID-19 pandemic, 60% of global consumers have proactively improved their physical and mental health.

The global sports nutrition market size was estimated at USD 45.24 billion in 2023 and is expected to grow at a compound annual growth rate (CAGR) of 7.5% from 2024 to 2030. Nutrition and diet are essential for maximum performance, and regular physical activity can alter the requirements for some micronutrients.

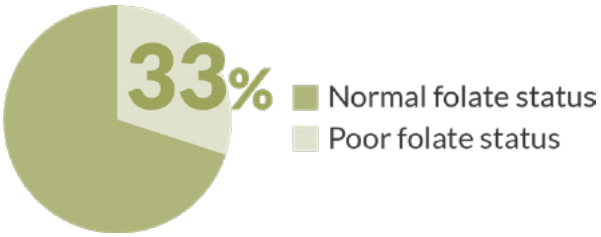
World Health Organization (WHO)
recommends moderate to vigorous intensity physical activity



Folate Status in Athletes

B vitamins are vital for exercise because they are involved in the regulation of *energy metabolism*.

Folate status in female marathon runners



The active folate 5-MTHF, such as Quatrefolic®, doesn't need to be metabolized, and it has a direct role in synthesizing new cells and building and repairing body tissues, including those injured due to physical activity.

Studies have shown that athletes, especially women, may be more at risk of poor folate intake and status. It is noted that severe folate insufficiency could result in anemia and, consequently, reduce endurance workout performance. Thus, it is recommended that all athletes, both males and females, increase their daily folate intake.

Food folate and folic acid need conversion via specific enzymes to provide *biologically active folate* (5-MTHF), where MTHFR (methylenetetrahydrofolate reductase) is the rate-limiting enzyme.

Folate Benefits for Athletes

Petito A, Altamura M, Iuso S, Padalino FA, Sessa F, D'Andrea G, Margaglione M, Bellomo A. The Relationship between Personality Traits, the 5HTT Polymorphisms, and the Occurrence of Anxiety and Depressive Symptoms in Elite Athletes. PLoS ONE 2016; 11:e0156601.

Active Folate's Mental-Function Effect

The improvement of exercise performance in sports not only involves the enhancement of muscle strength but also includes the development of *psychological* and cognitive functions.

Elite athletes may suffer from competition stress and anxiety, which are associated with a range of symptoms.

Quatrefolic®, the active folate 5-MTHF, is readily available for the athlete's body.

Athlete Cognitive Skills

- Decision-making
- Processing speed
- Working memory
- Perceptual processing
- Motor functioning
- Attention

Competition Stress & Anxiety

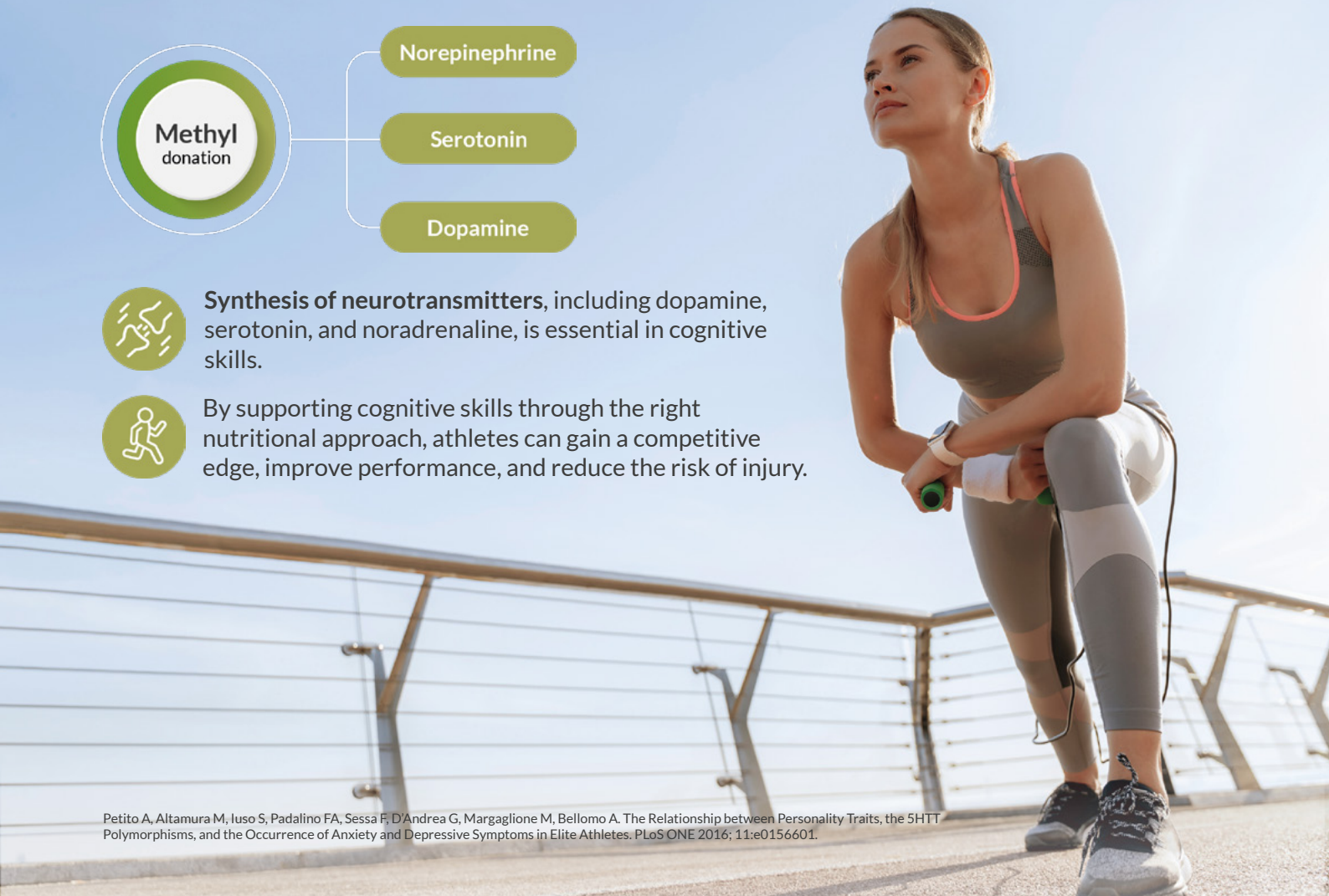
- Quality of sleep
- Level of energy
- Fatigue

Folate as Quatrefolic® is essential in the Methylation Cycle

Active folate, such as Quatrefolic®, not only supports physical health but also helps boost *mental function* and processing, increasing the production of neurotransmitters (through mono-carbon metabolism).

Synthesis of neurotransmitters, including dopamine, serotonin, and noradrenaline, is essential in cognitive skills.

By supporting cognitive skills through the right nutritional approach, athletes can gain a competitive edge, improve performance, and reduce the risk of injury.



Petito A, Altamura M, Iuso S, Padalino FA, Sessa F, D'Andrea G, Margaglione M, Bellomo A. The Relationship between Personality Traits, the 5HTT Polymorphisms, and the Occurrence of Anxiety and Depressive Symptoms in Elite Athletes. PLoS ONE 2016; 11:e0156601.

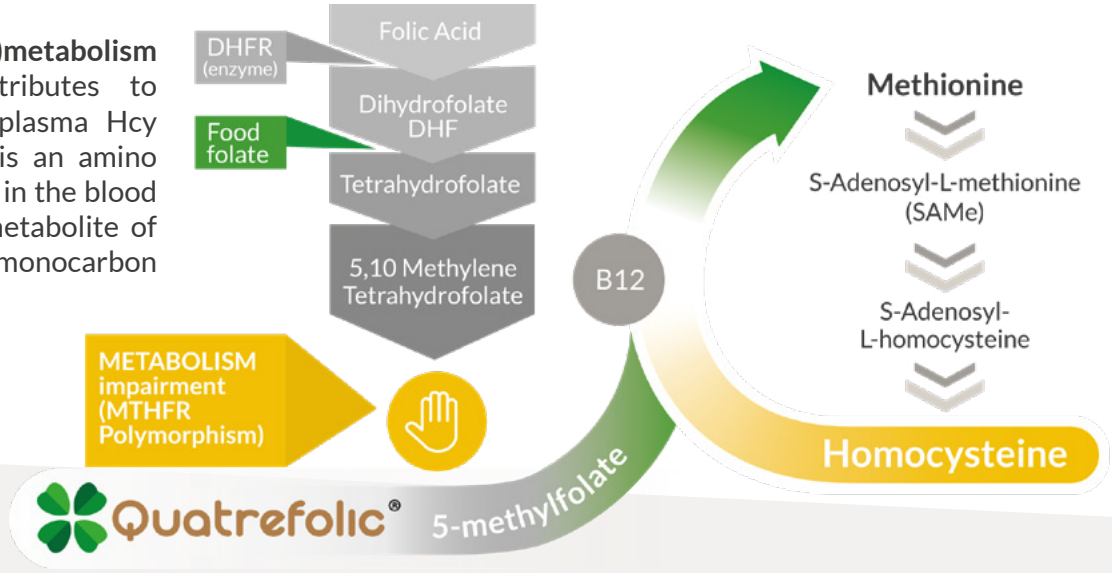
Active Folate's Cardiac/Cardiovascular Functions

Physical activity is a key component of health recognized to reduce the risk of cardiovascular disease, and the link between physical activity, blood folate, and homocysteine levels depends on the level and duration of exercise.

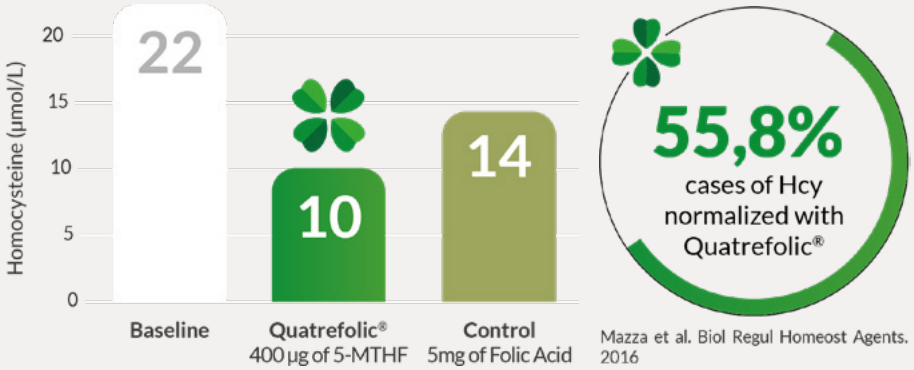
Intense endurance physical activity (e.g., a triathlon) negatively impacts Hcy levels (+Hcy) due to high folate consumption following exercise-induced metabolic demand.

Intense endurance activity can lead to increased homocysteine levels.

Folate is an essential homocysteine (Hcy)metabolism regulator and contributes to maintaining normal plasma Hcy concentrations. Hcy is an amino acid commonly found in the blood and produced as a metabolite of methionine in the monocarbon cycle.



Quatrefolic® supplementation has shown to be effective in lowering/normalizing homocysteine blood levels, supporting both the nutritional status of athletes and *cardiovascular health.*



Mazza et al. Biol Regul Homeost Agents. 2016

Active Folate, Muscle Strength and Recovery

Hwang et al. have reviewed the research on folate and its biological functions in the growth and differentiation of muscle cells and how folate deficiency affects muscle cell development. The results highlight that the higher blood folate levels are associated with:

- Improved muscle strength, which is crucial for better physical performance, stimulated by myogenesis (formation of skeletal muscular tissue);
- Increased muscle blood flow, which improves physical performance and muscle tissue regeneration after physical exercise.

Quatrefolic®

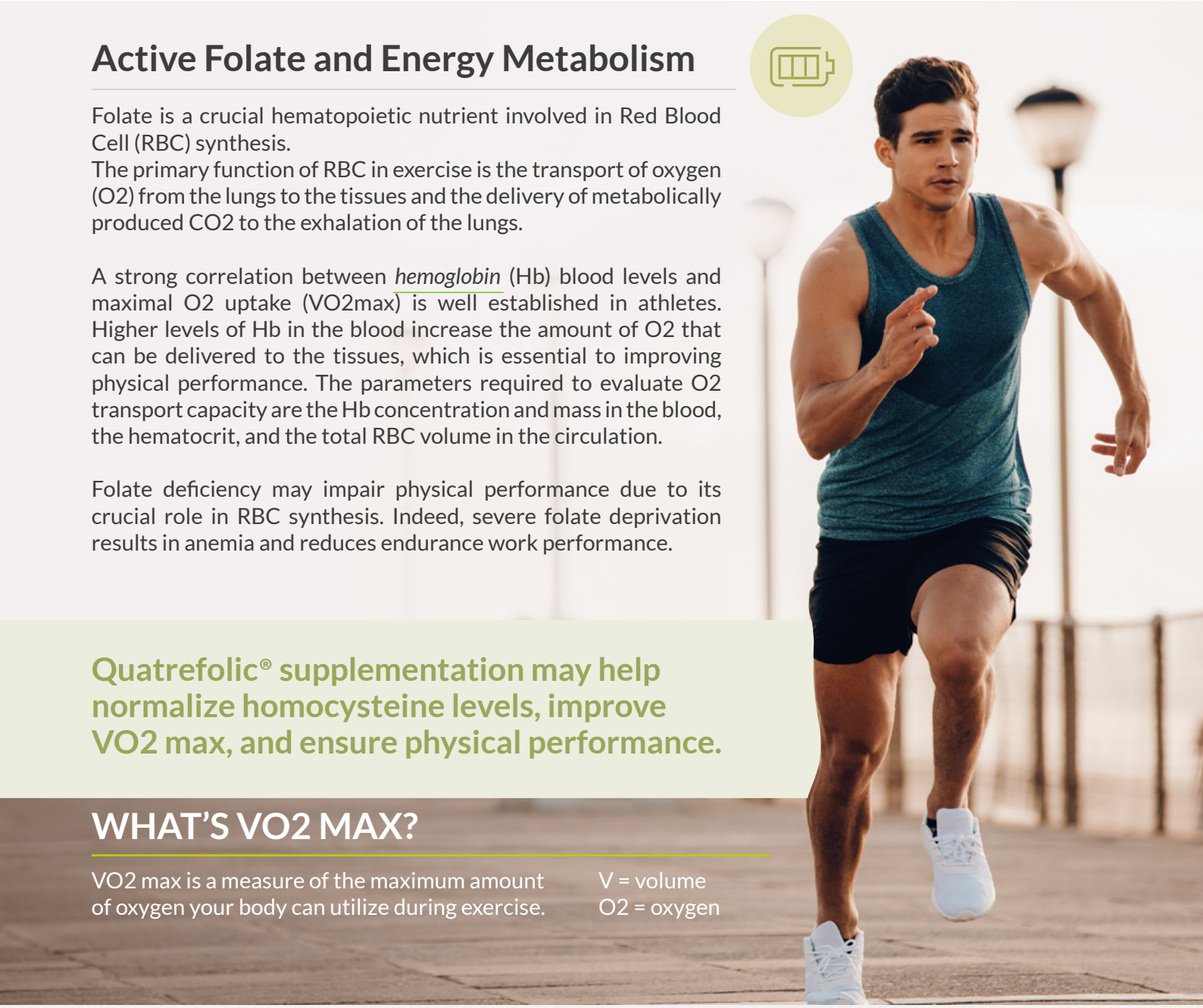
- **Helps improve muscle strength**
- **Regenerates muscle tissue quickly**
- **Helps faster recovery after exercise**

Active Folate and Energy Metabolism

Folate is a crucial hematopoietic nutrient involved in Red Blood Cell (RBC) synthesis.
The primary function of RBC in exercise is the transport of oxygen (O2) from the lungs to the tissues and the delivery of metabolically produced CO2 to the exhalation of the lungs.

A strong correlation between *hemoglobin* (Hb) blood levels and maximal O2 uptake (VO2max) is well established in athletes. Higher levels of Hb in the blood increase the amount of O2 that can be delivered to the tissues, which is essential to improving physical performance. The parameters required to evaluate O2 transport capacity are the Hb concentration and mass in the blood, the hematocrit, and the total RBC volume in the circulation.

Folate deficiency may impair physical performance due to its crucial role in RBC synthesis. Indeed, severe folate deprivation results in anemia and reduces endurance work performance.



Quatrefolic® supplementation may help normalize homocysteine levels, improve VO2 max, and ensure physical performance.

WHAT'S VO2 MAX?

VO2 max is a measure of the maximum amount of oxygen your body can utilize during exercise.

V = volume
O2 = oxygen

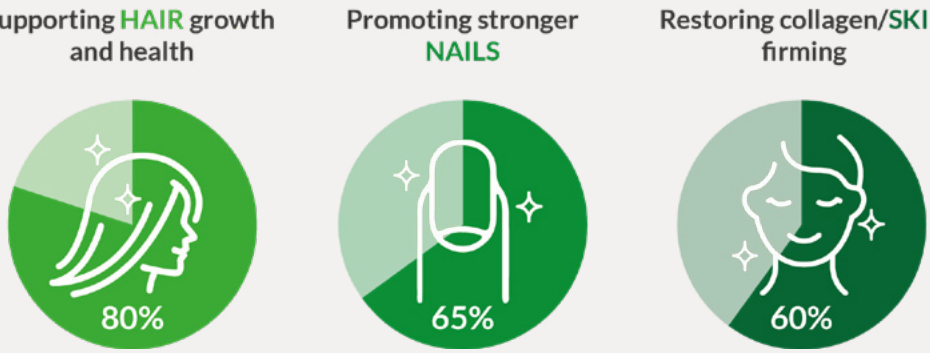
References: Hwang SY, Sung B, Kim ND. Roles of folate in skeletal muscle cell development and functions. Arch Pharm Res 2019; 42:319–25; Mairbäurl H. Red blood cells in sports: effects of exercise and training on oxygen supply by red blood cells. Front Physiol 2013; 4:332;Novack V, Finestone AS, Constantini N, Shpilberg O, Weitzman S, Merkel D. The prevalence of low hemoglobin values among new infantry recruits and nonlinear relationship between hemoglobin concentration and physical fitness. Am J Hematol 2007; 82:128–33;Wagner C. BIOCHEMICAL ROLE OF FOLATE IN CELLULAR METABOLISM*. Clinical Research and Regulatory Affairs 2001; 18:161–80

Active Folate and Beauty

Beauty Supplements Market was valued at around USD 2.90 billion in 2023 and is projected to grow at a 13.6% CAGR between 2024 and 2032.

Nutricosmetics, which emphasizes that beauty originates from the inside, emerged due to the intersection of nutrition and cosmetics. Supplements and other nutricosmetic items are designed to improve skin, hair, and nails by focusing on specific ingredients.

An October 2022 survey of more than 2,000 US female beauty buyers showed that consumer interest in beauty supplements is driven by:*



*conducted by The Benchmarking Company

Folate Status and Beauty

Folate is an essential water-soluble vitamin that plays a crucial role in various physiological processes, including DNA synthesis and cell division and repair.

> **5-MTHF is the biologically active form of folate and represents the predominant physiologic form of folate found in blood.**

Indeed, food folate and folic acid, the synthetic form used in supplements, need to undergo cellular metabolization to become active.

Supplementation with active folate 5-MTHF, such as Quatrefolic®, delivers a "finished" folate the body can immediately use without any transformation to support skin, hair, and nail health.

Quatrefolic®

Skin Health

Skin elasticity and firmness

Active folate is involved in DNA synthesis and cell division, which are crucial for the production and turnover of skin cells.

Active folate can boost collagen metabolism and support skin firmness.

Photoaging and UV protection

Folate, as Quatrefolic®, is involved in DNA repair, antioxidant activity, collagen production, and melanin synthesis, indirectly contributing to skin health and protection against sun-induced damage.

Quatrefolic®

Nail Health

Brittle nails may be a symptom of folate deficiency.

Quatrefolic® promotes healthy nail growth and plays a vital role in developing red blood cells, which carry oxygen throughout the body, including extremities such as fingernails and toenails.

Quatrefolic®

Hair Health

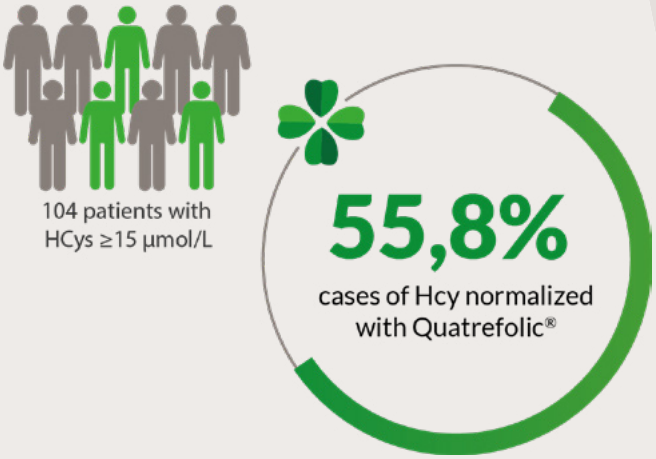
Active folate, such as Quatrefolic®, supports cell division and DNA synthesis in the hair follicles. It contributes to the overall strength and vitality of the hair, and it aids in tissue repair and growth, which is essential for maintaining a healthy scalp.

Active Folate, Skin Elasticity, and Firmness

> ***Hyperhomocysteinemia*** **is associated with skin aging, probably due to the oxidative stress leading to collagen breakdown and a decreased elasticity and firmness of the skin.**

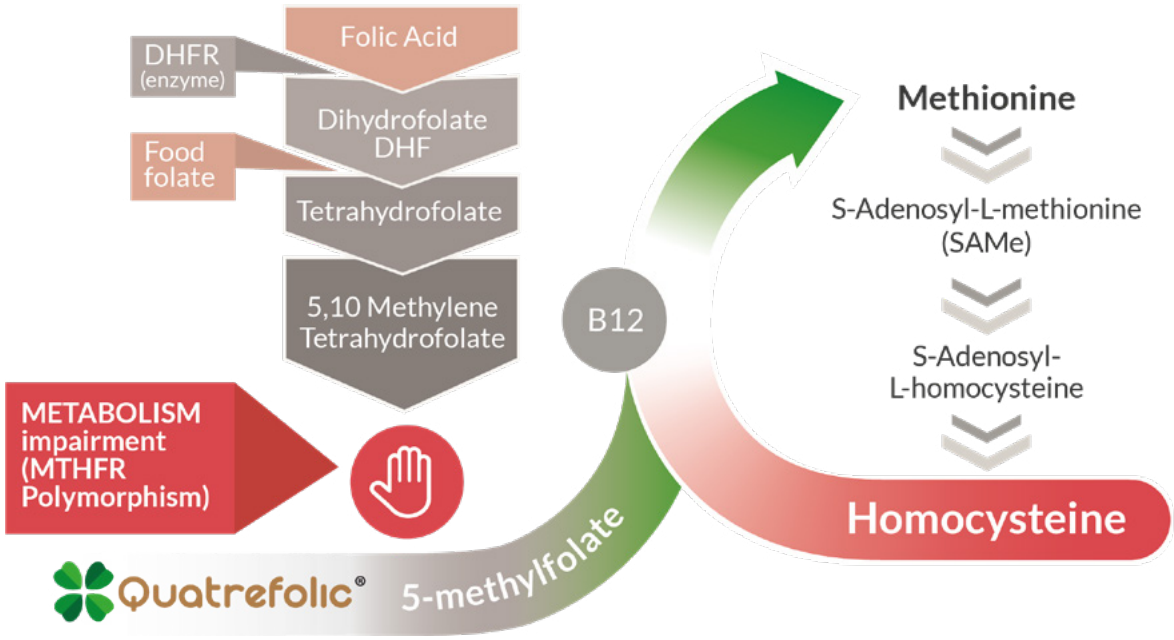
Two *clinical studies* reported the efficacy of folate in a blend with creatine for treating photo-aged skin. Beneficial effects were an improvement of skin elasticity and firmness, reduced wrinkle volume, tactile roughness, and laxity.

Folate is an essential homocysteine (Hcy) metabolism regulator that contributes to maintaining normal plasma Hcy concentrations. Blood folate deficiency can affect skin health, which is supported by the association between skin conditions and hyperhomocysteinemia.



Quatrefolic®, the Active Folate:

- **Provides the right active folate dosage, normalizing homocysteine plasma levels**
- **Improves collagen metabolism associated with skin elasticity and firmness**



Food folate and *folic acid* need conversion via specific enzymes to provide biologically active folate (5-MTHF), where MTHFR (methylenetetrahydrofolate reductase) is the rate-limiting enzyme.

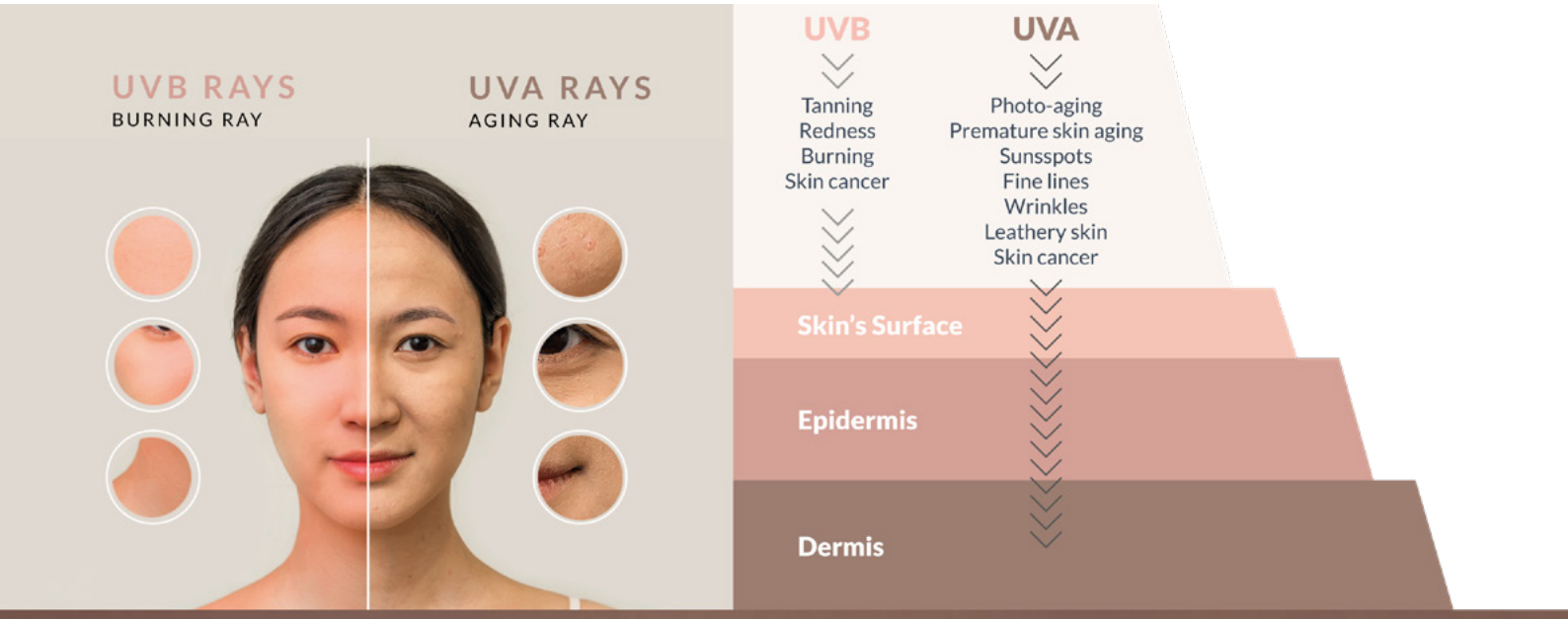
> **5-MTHF, such as Quatrefolic®, bypasses the enzymatic step since it doesn't need to be converted, supporting cardiovascular health and skin regeneration.**

Scaglione F, Panzavolta G. Folate, folic acid and 5-methyltetrahydrofolate are not the same thing. Xenobiotica 2014; 44:480–8. ; Blom HJ, Smulders Y. Overview of homocysteine and folate metabolism. With special references to cardiovascular disease and neural tube defects. J Inherit Metab Dis 2011; 34:75–81. ; Watson RR, Zibadi S, Preedy VR. Dietary components and immune function. Humana Press: New York, Dordrecht, Heidelberg etc., 2010. ; 10. Williams JD, Jacobson EL, Kim H, Kim M, Jacobson MK. Folate in skin cancer prevention. Subcell Biochem 2012; 56:181–97. Lucock M, Beckett E, Martin C, Jones P, Furst J, Yates Z, Jablonski NG, Chaplin G, Veysey M. UV-associated decline in systemic folate: implications for human nutrigenetics, health, and evolutionary processes. Am J Hum Biol 2017; 29. - 23. Lucock MD. The evolution of human skin pigmentation: A changing medley of vitamins, genetic variability, and UV radiation during human expansion. Am J Biol Anthropol 2023; 180:252–71.

Active Folate, Photoaging , and UV protection

Skin, the body's largest, most exposed organ, provides a protective interface between humans and the environment. One of its primary roles is protecting against exposure to sunlight, such as ultraviolet radiation (UVR).

Optimized folate levels support many biochemical processes necessary for the maintenance and function of *healthy skin.*



Folate Levels and UV Irradiance

Skin pigmentation serves to balance the interaction of micronutrients with solar UVR.
Studies show that chronic exposure can reduce long-term systemic folate levels.

In particular, 5-MTHF, the active form of folate, can be degraded by UVR, which results in the loss of vitamin activity.

A large cross-sectional *clinical study* (N=649) showed that cumulative UV irradiance reduces long-term systemic folate levels, which is influenced by the C677T-MTHFR gene polymorphism.
More specifically, **UV irradiance lowers red blood cell folate** in an MTHFR genotype-specific way, which has implications for human health and the evolution of skin pigmentation.

Quatrefolic® supplementation replenishes the essential folate levels required to protect the skin from the harmful effects of UVA and UVB rays.

Active Folate and the Elderly

Aging population and folate levels

In 2050, the over 60 years will represent 22% of the world's population. As aging comes with new challenges for us bodies, this reality implies an increase in health needs.
Dietary deficiencies, Vitamin D, Magnesium, Calcium, and vitamin B are often prevalent in the elderly. **Studies show that approximately 30% of individuals aged ≥ 65 years old experience a deficiency of folate.** Notable works, done in healthy aging, have focused on ingredients that reduce oxidative stress (such as active folate) and consequences in health. They suggest that some ingredients could also be related with immunity. Indeed, poor folate levels are associated with an increased risk of diseases which may imply hyperhomocysteinemia, vascular issues and cognitive dysfunction.

Thanks to its various health claims (EFSA APPROVED) related to psychological functions, reduction of tiredness and fatigue, cell division, normal homocysteine metabolism and well function of the immune system, folate is a good solution for elderly people.

For older adults and the *elderly*, the supplementation of the proper form of folate is essential to quickly re-establish the plasma folate levels and counter the variety of physiologic changes that influence folate status and the need for medical care.

By ensuring the right dosage of biologically active folate, Quatrefolic®, the salt of glucosamine 5-MTHF, allows the full enjoyment of the golden years.

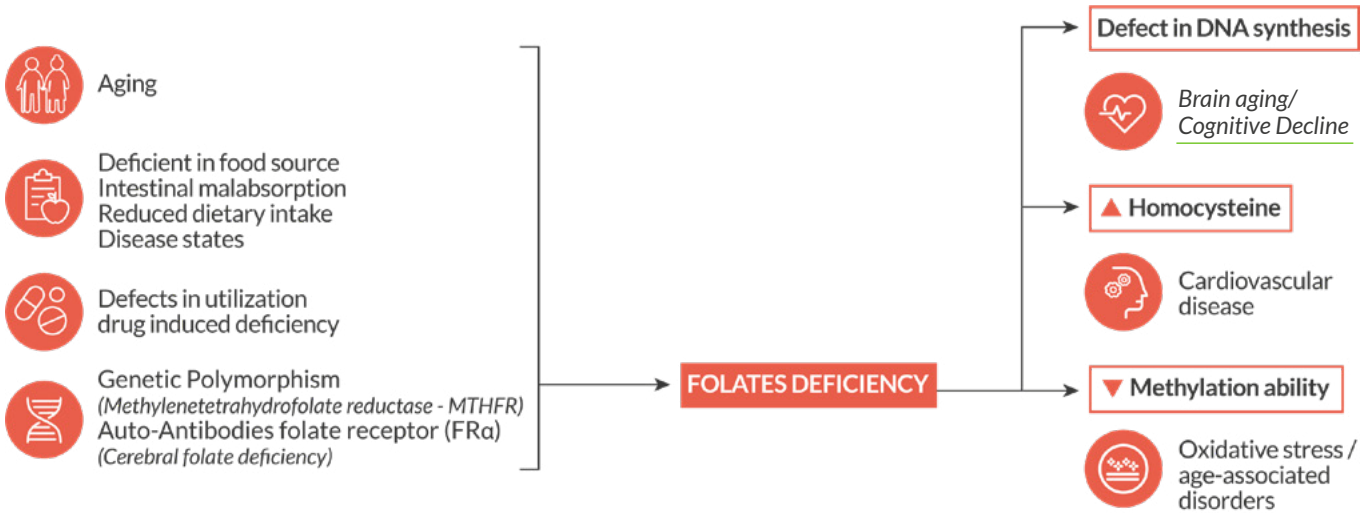
Bani S. et al. J Caring Sci. 2013; Zhang FF. et al. BMJ. 2020; EFSA Panel on Dietetic Products, Nutrition and Allergies (NDA). "Scientific Opinion on the substantiation of health claims related to folate and blood formation (ID 79), homocysteine metabolism (ID 80), energy-yielding metabolism (ID 90), function of the immune system (ID 91), function of blood vessels (ID 94, 175, 192), cell division (ID 193), and maternal tissue growth during pregnancy (ID 2882) pursuant to Article 13 (1) of Regulation (EC) No 1924/2006." EFSA Journal 7.10 (2009): 1213.

Quatrefolic® and the CAUSES of folate deficiencies

The lack of folate (vitamin B9) derives mainly from the reduced dietary intake and the impairment of metabolism and may require supplementation.

The metabolic availability of biological active folate - methylfolate, 5-methyltetrahydrofolate or 5-MTHF - is affected by multi-steps process of conversion of both folic acid and food folate, where the enzyme MTHFR (methylenetetrahydrofolate reductase) owns a key role. Some individuals, due to their unique genetic patterns and expression, have a polymorphic form [G10] with impaired folate metabolism and with exacerbation of folate deficiency.

External supplementation of the naturally occurring 5-MTHF, like Quatrefolic® has important advantages over synthetic folic acid - it is well absorbed, and its bioavailability is not affected by metabolic defects.



Quatrefolic® supplementation has important advantages over synthetic folic acid

The 5-MTHF is:

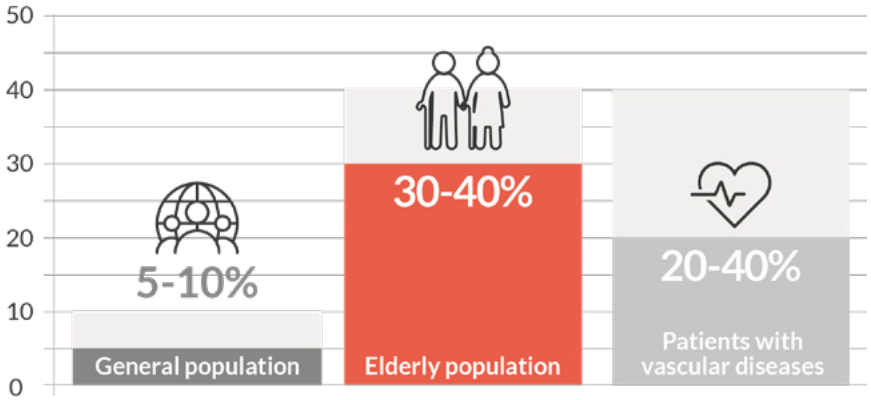
- the well absorbed folate form even when gastrointestinal pH is altered
- the folate form normally found in the circulation
- the folate form that is transported into peripheral tissues to be used for cellular metabolism
- the folate form able to cross the BBB (Blood Brain Barrier)

Quatrefolic® bioavailability[G11] is not affected by multi-steps process of conversion and by metabolic defects typical of folic acid, such as the polymorphism of the enzyme MTHFR.

Araújo J. et al. Ageing research reviews 2015; Mitchell E.S. et al. Neurosci Biobehav Rev 2014; Ostrakhovitch S. et al. Ageing Res Rev. 2019; Selhub J. et al. JAMA 1993; Selhub J. et al. Am J Clin Nutr 2000; Rampersaud G.C. et al. J Am Coll Nutr. 2003; Patanwala I. et al. Am J Clin Nutr 2014; Scaglione F., Panzavolta G. Xenobiotica 2014; Ulrich C.M., Potter J.D. Cancer Epidemiol Biomarkers Prev 2006; Gillies N. et al. Br J Nutr. 2020; Lökk J.et al. J Gerontol A 2003; Tinelli C et al. Front Nutr. 2019.

Homocysteine in Health

Prevalence of hyperhomocysteinemia in different population groups %



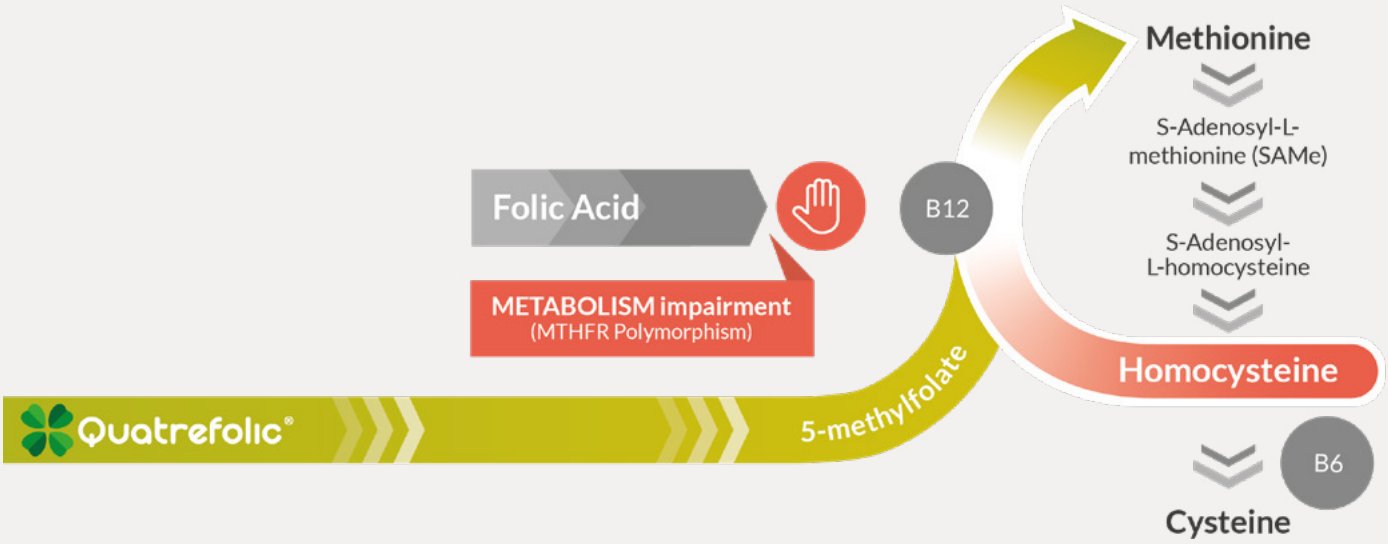
The best folate to help to maintain ideal levels of homocysteine

Homocysteinemia plays a crucial role for disease development determining longevity and health throughout a person's life and is seen as a predictor of potential health problems. The incidence of hyperhomocysteinemia in elderly population is significant.
Folate deficiency is one of the causes of hyperhomocysteinemia.

Quatrefolic® may be also a valid nutritional support for people with alterations in the metabolism of folate and, consequently, of homocysteine. In these subjects, the most prevalent *polymorphism of the MTHFR* enzyme may impair the availability of 5-MTHF.

The Classification of Homocysteinemia

TEST RESULTS	DIAGNOSIS
Normal	5 - 15 µmol/l
Mild	15 - 30 µmol/l
Moderate	30 - 100 µmol/l
Severe	> 100 µmol/l



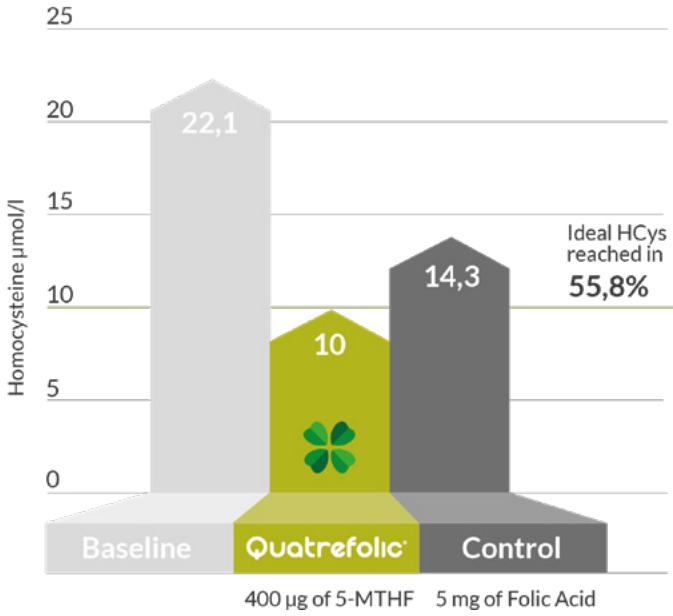
Miller A.L. Altern Med Rev. 2003; Refsum M.D., Ueland P.M. Annu Rev Medicine 1998; Nygard O. et al. Annu Rev Medicine 1992; Tavares J.R. et al. Arquivos brasileiros de cardiologia 2002; Caruso A. et al. J Cardiovasc Pharmacol 2006; Scott J.M., Weir DG. J Cardiovasc Risk.1998; Selhub J, Jacques PF, Wilson PWF, et al. JAMA 1993; Peng H. et al. Journal of Zhejiang University Science B, 201; Djuric D. et al. Can J Physiol Pharmacol. 2018

Quatrefolic® and Cardiovascular Health

In 2015, a meta-analysis showed that the elevated HCys level is an independent predictor for cardiovascular good health in the general population, and among elderly persons.

Mazza et al. investigated the efficacy of Quatrefolic® (400 µg of Quatrefolic® plus B6, and B12) in lowering homocysteine serum levels (HCys) versus a conventional vitamin supplementation with highly dosed folic acid (5 mg/day), in hypertensive subjects at low cardiovascular risk (104 patients with HCys ≥15 µmol/l).

The result shows significant HCys reduction in comparison with baseline from 21.5 µmol/l to 10.0 µmol/l with the product containing Quatrefolic®. The treatment was significantly effective, and the ideal HCys level was reached in 55.8% of cases in the Quatrefolic® group, and it was significantly higher than in controls.

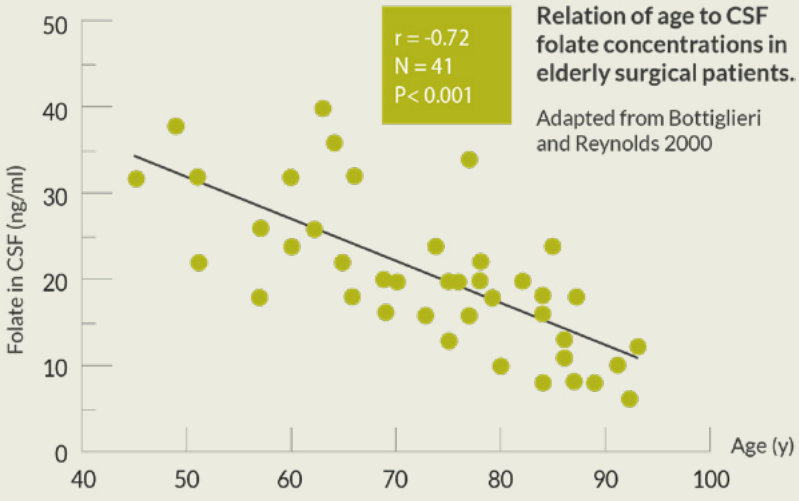


Quatrefolic® offers a homocystne -lowering and -normalizing effect clinically proven.

Quatrefolic® Brain Aging and Cognitive Decline

In 2000, *Bottiglieri and Reynolds* showed that the concentration of folate in the cerebrospinal fluid (CSF) decreases with age, particularly in older adults over 70 of age. This provided further evidence of a link between folate concentration and age, and for the first time, highlighted this connection directly in the nervous system.

Quatrefolic® supplementation is suggested to guarantee the right dosage of biologically active folate and to efficiently support brain function, since it is the only folate form able to cross the blood-brain barrier.



Relation of age to CSF folate concentrations in elderly surgical patients.

Adapted from Bottiglieri and Reynolds 2000

Mazza A. et al. Biol Regul Homeost Agents 2016 ; Fu Y. et al. Br J Pharmacol 2018; Peng H. et al. J Zhejiang Univ Sci B. 2015; Ambrosino P. at al. Nutr Res. 2015; Bottiglieri T., Reynolds E. et al. J Neurol Neurosurg Psychiatry 2000; Seshadri S. at al. N Engl J Med 2002; Stover PJ et al. Curr Opin Biotechnol. 2017: Rima Obeid R., Herrmann W. FEBS Letters 580, 2006.

Quatrefolic® E-Book

As a biochemist specialized in the field of nutrition, I explore the profound impact of nutrition on genetic expression and health, and it is with great enthusiasm that I introduce this e-book on Quatrefolic®, a cutting-edge development in folate supplementation by Gnosis by Lesaffre.

This resource is crafted to serve as a comprehensive guide on Quatrefolic®, synthesizing decades of scientific research into an accessible, interactive format. The e-book aims to explore the manifold benefits of this revolutionary form of folate, suitable for healthcare professionals, researchers, and health-conscious individuals alike.

Quatrefolic® represents the latest innovation in folate technology, offering the most bioavailable form of folate—5-methyltetrahydrofolate (5-MTHF) in the form of a glucosamine salt. This enhances both its stability and bioavailability, ensuring that the body can utilize folate efficiently without the complex metabolic conversions required by traditional folic acid supplements.

Folate plays a critical role in many of the body's essential functions, including DNA synthesis and repair, cellular division, and amino acid metabolism. The traditional form of supplemented folate, folic acid, must undergo conversion in the body to its active form, 5-MTHF, a process mediated by the enzyme methylenetetrahydrofolate reductase (MTHFR). Genetic variations can impair this conversion, leading to suboptimal folate levels despite intake. Quatrefolic® bypasses this metabolic hurdle by delivering folate in its active form, directly accessible for biological action, which is a significant advantage for individuals with MTHFR gene variants.

Prof. Luca Tiano PhD
Coordinator of the MSc in Food and Nutrition
at the Polytechnic University of Marche

Quatrefolic®

Glossary

[G1] **folate requirements:** The requirement is of 5-10 fold higher compared to non-pregnant ones.

[G2] **elevated homocysteine:** The excess homocysteine may damage the lining of your arteries (blood vessels that carry oxygen-rich blood throughout your body). High levels of homocysteine can also lead to blood clots or blood vessel blockages. Artery damage or blood clots significantly raise your risk of heart attack.

[G3] **polymorphic forms:** The MTHFR genetic polymorphisms related to the folate pathway have been shown to be associated with functional implications and specific conditions including NTDs (van der Linden et al. 2006), cardiovascular disease (Klerk et al. 2002).

[G4] **polymorphic MTHFR:** Nowadays a total of 9 common variants have been reported. The two most common ones are C677T and A1298C. The numbers refer to their location on the gene.

[G5] **nutritional deficiencies:** Without adequate supplementation, levels of maternal folate decrease gradually from the fifth month of pregnancy onwards and remain low for several months after childbirth. This decline in folate levels is further accentuated in women with shorter inter-pregnancy intervals.

[G6] **UMFA** https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4336532/ Quatrefolic® UMFA (gnosisbylesaffre.com)

[G7] **MTHFR polymorphism:** The MTHFR genetic polymorphisms related to the folate pathway have been shown to be associated with functional implications and specific conditions including NTDs (van der Linden et al. 2006), cardiovascular disease (Klerk et al. 2002).

[G8] **atherosclerosis:** Atherosclerosis is the buildup of fats, cholesterol and other substances in and on the artery walls. This buildup is called plaque. The plaque can cause arteries to narrow, blocking blood flow. or it burst, leading to a blood clot.
Although atherosclerosis is often considered a heart problem, it can affect arteries anywhere in the body. Atherosclerosis can be treated. Healthy lifestyle habits can help prevent atherosclerosis.

[G9] **oxidative stress:** Oxidative stress is a phenomenon caused by an imbalance between production and accumulation of oxygen reactive species (ROS) in cells and tissues and the ability of a biological system to detoxify these reactive products.

[G10] **polymorphic form:** The MTHFR genetic polymorphisms related to the folate pathway have been shown to be associated with functional implications and specific conditions including NTDs (van der Linden et al. 2006), cardiovascular disease (Klerk et al. 2002).

[G11] **brain aging/cognitive decline:** Bioavailability is a subcategory of absorption and is the fraction (%) of an administered drug that reaches the systemic circulation. https://quatrefolic.com/what-is-quatrefolic/innovative-folate/

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