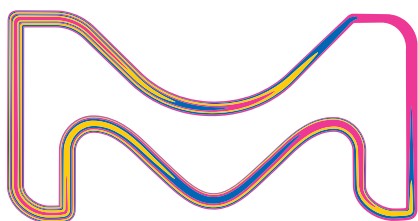
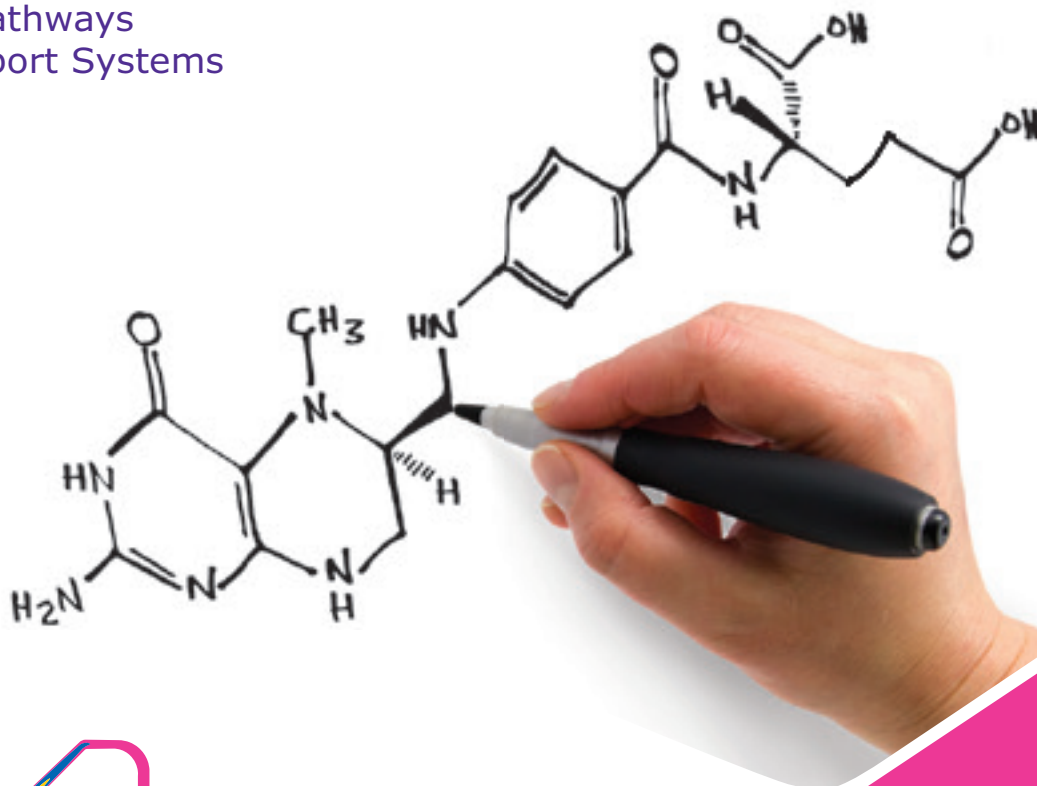


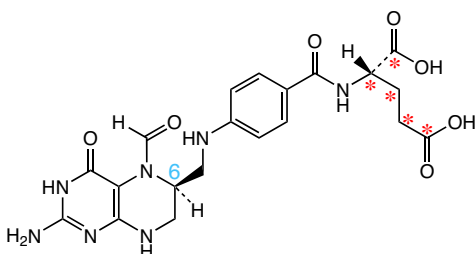
Folate Compounds

From The Leading Folate Chemistry Experts

Folate Substance List
 Natural Folates
 Biochemical Pathways
 Cellular Transport Systems
 Folate History



Folate Substance List



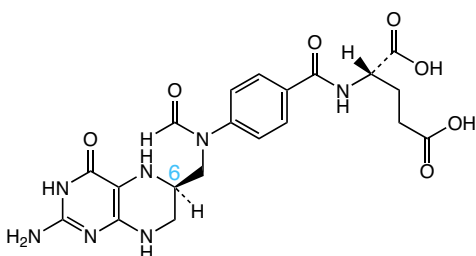
5-Formyltetrahydrofolate (Leucovorin)

$C_{20}H_{23}N_7O_7$
MW 473.44

(6R,S)-, (6R)-, (6S)-5-CHO-H₄PteGlu,
Ca- or Na₂-salt

$C_{15}^{13}C_5H_{23}N_7O_7$
MW 478.44

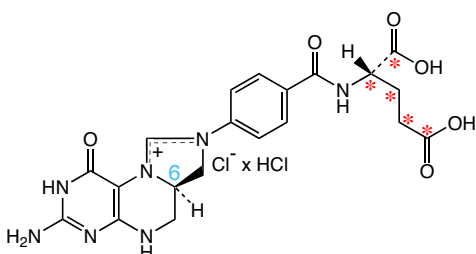
(6S)-5-CHO-H₄Pte[¹³C₅]Glu, Ca-salt



10-Formyltetrahydrofolate

$C_{20}H_{23}N_7O_7$
MW 473.44

(6R,S)-, (6S)-, (6R)-10-CHO-H₄PteGlu,
Ca- or Na₂-salt



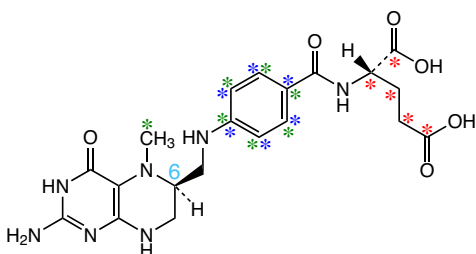
5,10-Methenyltetrahydrofolate (Anhydroleucovorin)

$C_{20}H_{23}Cl_2N_7O_6$
MW 528.35

(6R,S)-, (6R)-, (6R)-5,10-CH⁺-H₄PteGlu-
Cl x HCl (Cl x HCl salt)

$C_{15}^{13}C_5H_{23}Cl_2N_7O_6$
MW 533.35

(6R)-5,10-CH⁺-H₄Pte[¹³C₅]Glu-
Cl x HCl



5-Methyltetrahydrofolate

$C_{20}H_{25}N_7O_6$
MW 459.46

(6R,S)-, (6R)-, (6S)-5-CH₃-H₄PteGlu,
Ca- or Na₂-salt

$C_{15}^{13}C_5H_{25}N_7O_7$
MW 464.46

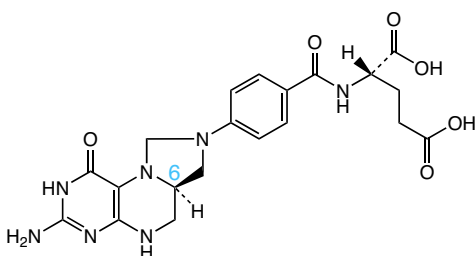
(6S)-5-CH₃-H₄Pte[¹³C₅]Glu, Ca-salt

$C_{13}^{13}C_6H_{25}N_7O_7$
MW 465.46

(6S)-5-CH₃-H₄[¹³C₆]PteGlu, Ca-salt

$C_{13}^{13}C_7H_{25}N_7O_6$
MW 466.46

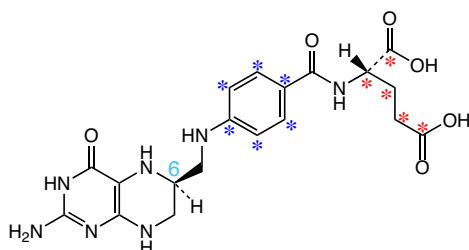
(6S)-5-¹³CH₃-H₄[¹³C₆]PteGlu, Ca-salt



5,10-Methylenetetrahydrofolate

$C_{20}H_{23}N_7O_6$
MW 457.44

(6R,S)-, (6S)-, (6R)-5,10-CH₂-H₄PteGlu,
Ca- or Na₂-salt



Tetrahydrofolate

$C_{19}H_{23}N_7O_6$
MW 445.43

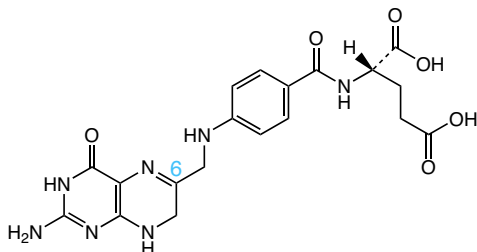
(6R,S)-, (6R)-, (6S)-H₄PteGlu,
Ca- or Na₂-salt

$C_{14}^{13}C_5H_{23}N_7O_6$
MW 450.43

(6S)-H₄Pte[¹³C₅]Glu, free acid form

$C_{13}^{13}C_6H_{23}N_7O_6$
MW 451.43

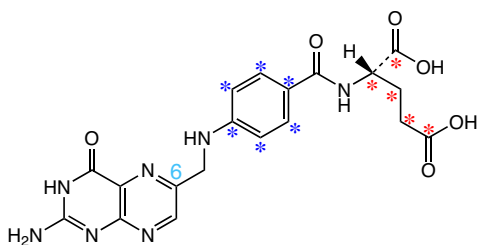
(6S)-H₄[¹³C₆]PteGlu, free acid form



7,8-Dihydrofolate

$C_{19}H_{21}N_7O_6$
MW 443.41

7,8-H₂PteGlu, free acid form



Folic acid (Pteroylglutamic acid)

$C_{19}H_{19}N_7O_6$
MW 441.40

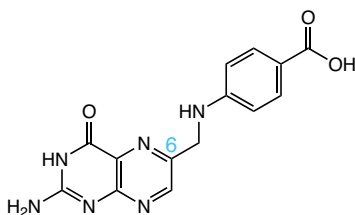
PteGlu, free acid form, Na₂-salt

$C_{14}^{13}C_5H_{19}N_7O_6$
MW 446.40

Pte[¹³C₅]Glu, free acid form

$C_{13}^{13}C_6H_{19}N_7O_6$
MW 447.40

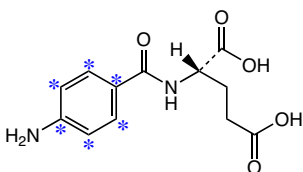
[¹³C₆]PteGlu, free acid form



Pteric acid

$C_{14}H_{12}N_6O_3$
MW 312.28

Pte, free acid form



4-Aminobenzoylglutamic acid (PABGA)

$C_{12}H_{14}N_2O_5$
MW 266.25

7-(4-Aminobenzoyl)-L-glutamic acid,
free acid form

$C_6^{13}C_6H_{14}N_2O_5$
MW 272.25

7-(4-Amino[¹³C₆]benzoyl)-L-glutamic
acid, free acid form

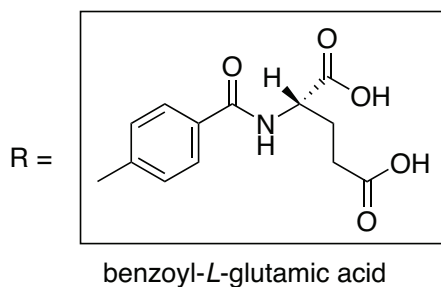
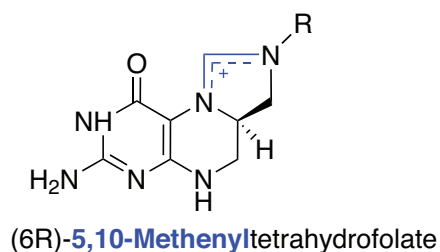
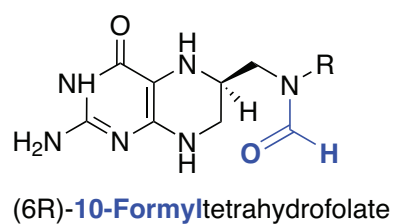
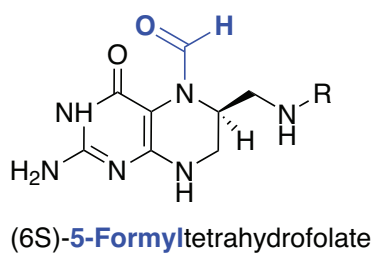
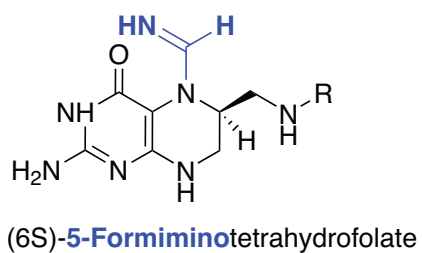
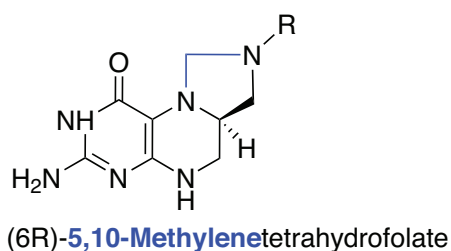
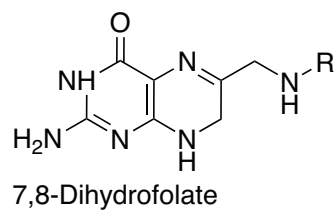
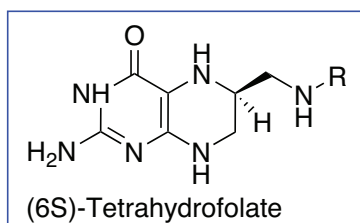
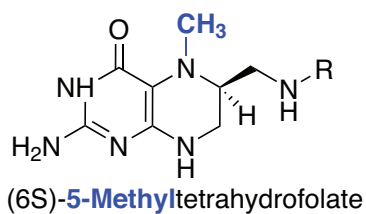
Structure and nomenclature of natural diastereoisomer

*¹³C₅-labeled form available

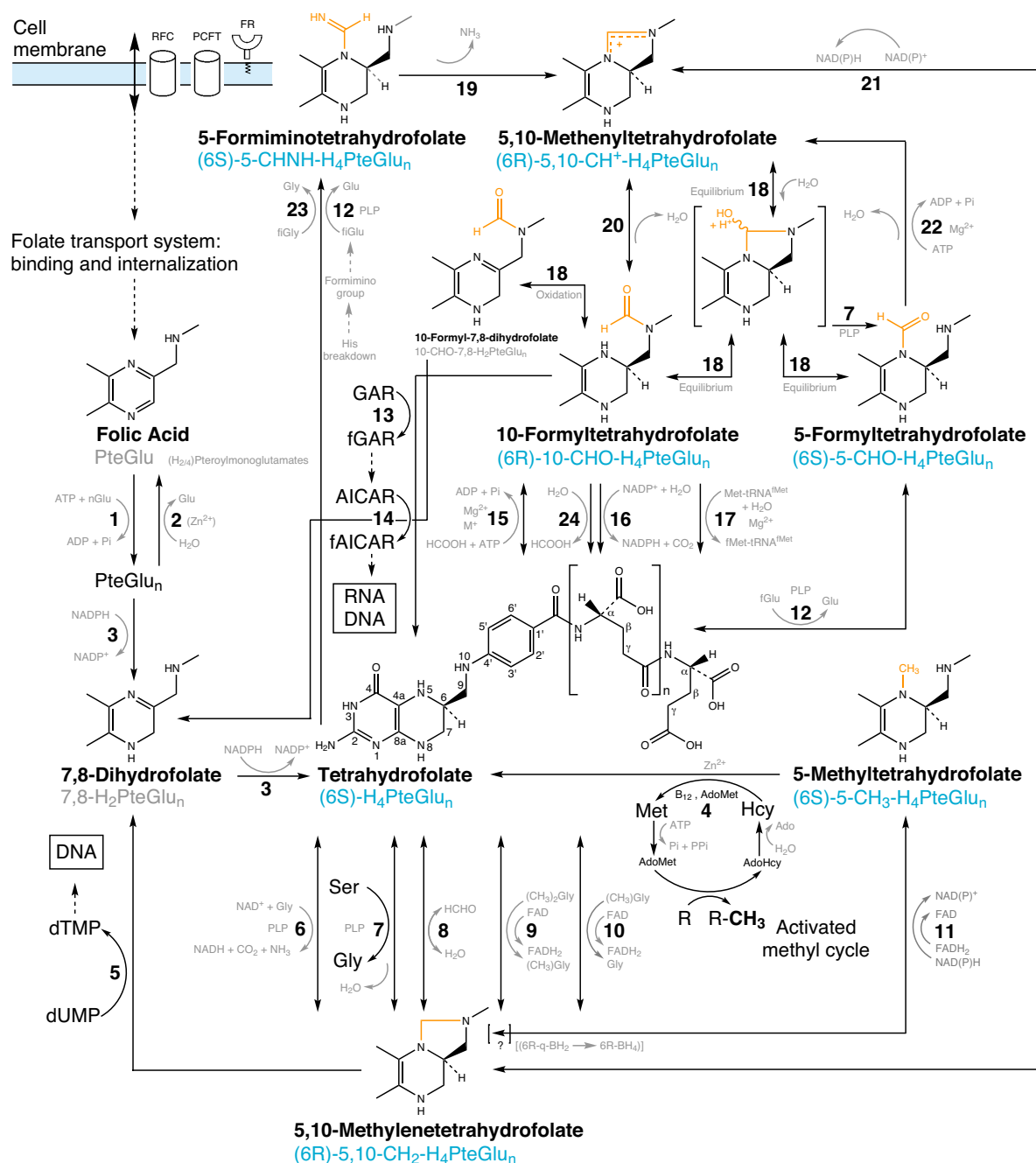
*¹³C₆-labeled form available

*¹³C₇-labeled form available

Natural Folates



Biochemical Pathways of Folates



- | | | | |
|----|--|----|--|
| 1 | Folypolyglutamate synthase EC 6.3.2.17 (FPGS , FPGS) | 15 | C-1-tetrahydrofolate synthase, EC 6.3.4.3 ³⁾ (MTHFD1 , MTHFD1) |
| 2 | γ-Glutamyl hydrolase EC 3.4.19.9 | | Monofunctional C1-tetrahydrofolate synthase EC 6.3.4.3 (MTHFD1L) |
| 3 | Dihydrofolate reductase EC 1.5.1.3 (DHFR , DHFR , DHFR2) | 16 | 10-Formyltetrahydrofolate dehydrogenase EC 1.5.1.6 (ALDH1L1 , ALDH1L2) |
| 4 | Methionine synthase EC 2.1.1.13 (MTR) | 17 | Methionyl-tRNA formyltransferase EC 2.1.2.9 (MTFMT) |
| 5 | Thymidylate synthase EC 2.1.1.45 (TYMS , TYMS , TYMS) | 18 | Non-enzymatic reactions |
| 6 | Glycine cleavage system ¹⁾ (GLDC/DLD/AMT/GCSH) | 19 | Formimidoyltransferase-cyclodeaminase EC 4.3.1.4 ²⁾ (FTCD) |
| 7 | Serine hydroxymethyltransferase EC 2.1.2.1 (SHMT1 , SHMT2a , SHMT1 , SHMT2a , SHMT2) | 20 | C-1-tetrahydrofolate synthase EC 3.5.4.9 ³⁾ (MTHFD1 , MTHFD1) |
| 8 | Equilibrium (non-enzymatic reaction) | | Bifunctional methylenetetrahydrofolate dehydrogenase/cyclohydrolase EC 3.5.4.9 ⁴⁾ (MTHFD2 / MTHFD2L) |
| 9 | Dimethylglycine dehydrogenase EC 1.5.99.2 (DMGD) | 21 | C-1-tetrahydrofolate synthase EC 1.5.1.5 ³⁾ (MTHFD1 , MTHFD1) |
| 10 | Sarcosine dehydrogenase EC 1.5.8.3 (SARDH) | | Bifunctional methylenetetrahydrofolate dehydrogenase/cyclohydrolase EC 1.5.1.15 ⁴⁾ (MTHFD2 / MTHFD2L) |
| 11 | 5,10-Methylenetetrahydrofolate reductase EC 1.5.1.20 (MTHFR) | 22 | 5-Formyltetrahydrofolate cyclo-ligase EC 6.3.3.2 (MTHFS) |
| 12 | Formimidoyltransferase-cyclodeaminase EC 2.1.2.5 ²⁾ (FTCD) | 23 | Glycine formimidoyltransferase EC 2.1.2.4 |
| 13 | Trifunctional purine biosynthetic protein adenosine-3 EC 2.1.2.2 (GART) | 24 | 10-Formyltetrahydrofolate deformylase EC 3.5.1.10 |
| 14 | Bifunctional purine biosynthesis protein PURH EC 2.1.2.3 (ATIC) | | |

cytosolic
mitochondrial
nuclear

- 1) Glycine cleavage system (EC 1.4.4.2, EC 1.8.1.4, EC 2.1.2.10)
- 2) Bifunctional enzyme in eukaryotes (EC 2.1.2.5, EC 4.3.1.4)
- 3) Trifunctional enzyme in eukaryotes (EC 6.3.4.3, EC 1.5.1.5, EC 3.5.4.9)
- 4) Bifunctional enzyme in eukaryotes (EC 1.5.1.15, EC 3.5.4.9)

Cellular Folate Transport Systems

Reduced Folate Carrier (RFC)

- Organic anion antiporter
- Major folate transport system

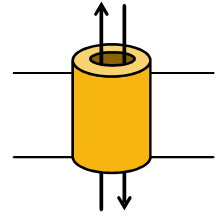
Type: Integral membrane glycoprotein, 12 transmembrane domains

pH Optimum: 7.4

Tissues: Placenta, liver, leukocytes, kidney, lung, bone marrow, intestine, CNS, brain

Affinities: (6S)-5-CH₃-H₄PteGlu (~ 5 μM) >> PteGlu (~ 200 μM)

Synonym: Solute carrier family 19 member 1



Folate Receptor (FR)

- Transport via receptor mediated endocytosis
- Expressed on the cell surface

Type: Anchored to cell surface by a glycosylphosphatidylinositol (GPI) domain

Isoforms: FR-α, FR-β, FR-γ (secreted), FR-δ

Tissues: FR-α: Epithelial tissues (e.g. placenta, proximal renal tubular cells, choroid plexus)
FR-α levels are greatly elevated in malignant tissues

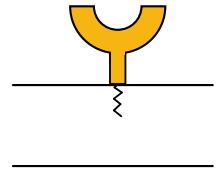
FR-β: Hematopoietic tissues (e.g. spleen, thymus, bone marrow), macrophages, fetal brain

FR-γ: Hematopoietic tissues (e.g. spleen, thymus, bone marrow)

FR-δ: Regulatory T-cells, Oocytes

Affinities: (6S)-5-CH₃-H₄PteGlu (≥ 1 nM) < PteGlu (< 1 nM)

Synonym: Folate binding protein (FBP)



Proton-Coupled Folate Transporter (PCFT)

- Proton symporter
- Major transport system for intestinal folate absorption

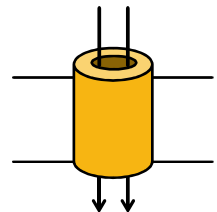
Type: Integral membrane glycoprotein, 12 transmembrane domains

pH Optimum: 5.5

Tissues: Intestine, kidney, liver, placenta, spleen, brain, testis, lung

Affinities: (6S)-5-CH₃-H₄PteGlu (~ 1 μM) = PteGlu (~ 1 μM)

Synonym: Heme carrier protein 1 (HCP1), solute carrier family 46 member 1



ATP-Binding Cassette (ABC) Transporters

- ATP dependent translocation of a wide variety of substances across membranes
- Export of folates (e.g. MRP1-5 and BCRP)

Type: Transmembrane protein

Affinities: Low-affinity, high-capacity pumps

Folate History

- 1931** Lucy Wills discovers that macrocytic anaemia can be prevented by adding yeast to a diet otherwise lacking B vitamins
- 1941** Herschel K. Mitchell and colleagues suggest the name "Folic acid" (folium, Latin for leaf) for the factor responsible for growth stimulation of *Streptococcus lactis* isolated from spinach
- 1945** Tom D. Spies demonstrates that Folic acid cures megaloblastic anaemia during pregnancy
- 1946** Robert B. Angier and co-workers report the structure and synthesis of the *Lactobacillus casei* factor (Folic acid) isolated from liver
- 1950** Emanuel B. Schoenbach and colleagues observe that the toxic side effects of amethopterin (methotrexate) cancer therapy can be reversed by treatment with "citrovorum factor" (Leucovorin)
- 1962** Victor Herbert consumes a folate-deficient diet for several months, documenting the development of deficiency symptoms
- 1968** Martin C. Carey and colleagues report that oral Folic acid therapy significantly reduces homocysteine excretion in the urine of mentally-handicapped children with homocystinuria
- 1981** Richard W. Smithells and co-workers report a preventive effect on neural tube defects by a periconceptional vitamin supplementation containing Folic acid
- 1982** David Machover and colleagues demonstrate that (6R,S)-5-Formyltetrahydrofolate or "Folinic acid" (Leucovorin) increases the therapeutic efficacy of 5-fluorouracil in the treatment of advanced colorectal and gastric adenocarcinomas
- 1991** Nicholas Wald demonstrates in a randomised double-blind prevention trial that Folic acid supplementation before pregnancy reduces the risk of neural tube defects by 70% in women who had previously given birth to a child with a neural tube defect
- 1991** Eprova AG succeeds in producing (6S)-5-Formyltetrahydrofolate (Levoleucovorin), the natural isomer of (6R,S)-5-Formyltetrahydrofolate (Leucovorin), in commercial quantities
- 1992** Andrew E. Czeizel finds that first occurrence of neural tube defects may be prevented by periconceptional Folic acid supplementation
- 1992** The U.S. Public Health Service recommends women of childbearing age to consume 0.4 mg of Folic acid daily
- 1997** Mary Ward and colleagues demonstrate that plasma homocysteine can be lowered by physiological doses of Folic acid
- 1998** The U.S. Food and Drug Administration (FDA) introduces mandatory fortification of flour, rice, pasta, and other grain products with Folic acid
- 2001** The U.S. Food and Drug Administration (FDA) accepts (6S)-5-Methyltetrahydrofolate (Metafolin®) for use in dietary supplements
- 2004** The European Food Safety Authority (EFSA) considers (6S)-5-Methyltetrahydrofolate (Metafolin®) safe
- 2005** The "Joint FAO/WHO Expert Committee on Food Additives" (JECFA) considers (6S)-5-Methyltetrahydrofolate (Metafolin®) safe
- 2008** The U.S. Food and Drug Administration (FDA) approves (6S)-5-Formyltetrahydrofolate (Fusilev®) for rescue after high-dose methotrexate therapy
- 2010** The U.S. Food and Drug Administration (FDA) approves Beyaz® a combination oral contraceptive containing (6S)-5-Methyltetrahydrofolate (Metafolin®) to prevent neural tube defects
- 2015** The Center for Radiopharmaceutical Sciences (CRS), a joint endeavor between the Paul Scherrer Institute, the ETH Zurich and the University Hospital Zurich starts the first-in-man clinical trial with a novel folate-based [¹⁸F]-PET tracer for imaging of folate receptor-positive tumors
- 2018** Isofol Medical AB reports positive efficacy data for (6R)-5,10-Methylenetetrahydrofolate (Modufolin®) from patients treated for metastatic colorectal cancer

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Lit. No. MK_BR2972EN Ver. 1.0
2018-11753 05/2018