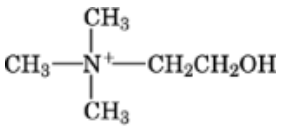
Kirk-Othmer Encyclopedia of Chemical Technology.

CHOLINE

Choline base [123-41-1], $[(CH_3)_3NCH_2CH_2OH]^+OH^-$, trimethyl(2-hydroxyethyl)-ammonium hydroxide, derives its name from bile (Greek *cholē*;) from which it was first obtained. This so-called free-choline is a colorless, hygroscopic liquid with an odor of trimethylamine. The quaternary ammonium compound **(1)** choline [62-49-7] or a precursor is needed in the diet as a constituent of certain phospholipids universally present in protoplasm.



(1)

This makes choline an important nutritional substance. It is also of great physiological interest because one of its esters, acetylcholine [51-84-3], appears to be responsible for the mediation of parasympathetic nerve impulses and has been postulated to be essential to the transmission of all nerve impulses. Acetylcholine and other more stable compounds that simulate its action are pharmacologically important because of their powerful effect on the heart and on smooth muscle. Choline is used clinically in liver disorders and as a constituent in animal feeds.

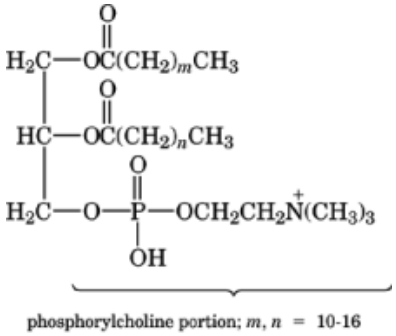
Choline was isolated from ox bile in 1849 by Strecker. During 1900 to 1920, observations led to interest in the vasodepressor properties of the esters of choline, and in the 1920s it was shown that acetylcholine was presumably the "vagus-substance." The nutritional importance of choline was recognized in the 1930s, when it was found that choline would prevent fatty infiltration of the liver in rats. Subsequent observations showed that choline deficiency could produce cirrhosis (1) or hemorrhagic kidneys (2) in experimental animals under various conditions.

1. Physical and Chemical Properties

Choline is a strong base ($pK_B = 5.06 \text{for} 0.0065 - 0.0403\ M \text{solutions}$) (3). It crystallizes with difficulty and is usually known as a colorless deliquescent syrupy liquid, which absorbs carbon dioxide from the atmosphere. Choline is very soluble in water and in absolute alcohol but insoluble in ether (4). It is stable in dilute solutions but in concentrated solutions tends to decompose at 100°C, giving ethylene glycol, poly(ethylene glycol), and trimethylamine (5).

2. Biological Functions

In nutrition, the most important function of choline appears to be the formation of lecithin (phosphatidylcholine) (**2**) and other choline-containing phospholipids.



(**2**)

Lecithin (qv) may be regarded as a triglyceride in which one of the fatty acid residues has been replaced by a phosphoric acid derivative of choline (phosphorylcholine) via cytidine diphosphate choline. The replacement changes the physical properties of the fat so that lecithin is readily dispersible with water. This property is important in the transport of fats in the blood. In choline deficiency, fats tend to accumulate in the liver presumably because they are not transformed into lecithin and hence are not carried away from the liver by the circulating blood. Owing to this effect, choline is said to have a lipotropic (fat-moving) action. Other known lipotropic substances, such as methionine, $CH_3SCH_2CH_2CH(NH_2)COOH$, and betaine, $(CH_3)_3N^+CH_2COO^-$;, furnish labile methyl groups that unite with 2-aminoethanol to form choline in the body. Choline itself can also yield labile methyl groups for the methylation of other organic compounds (2, 6–17).

Fatty infiltration of the liver has been observed to precede cirrhosis in experimental animals receiving diets low in choline and other substances that can furnish labile methyl groups, and can thus serve as precursors of choline.

Figure 1 shows some of the biological reactions involving labile methyl groups. The groups can originate from serine, formaldehyde, or formate by enzymatic reactions involving tetrahydrofolic acid, FAH_4, so that the compound N^5,N^{10}-methylenetetrahydrofolic acid, 5,10-CH_2FAH_4, is formed. This undergoes hydrogenation to form 5-methyltetrahydrofolic acid, 5-CH_3FAH_4, from which the methyl group is transferred to a vitamin B_{12} compound, shown in the diagram as CH_3B_{12}. This compound methylates homocysteine to produce methionine, which may become activated by adenosine triphosphate (ATP) with the formation of *S*-adenosylmethionine. The methyl group attached to sulfur in this compound can be transferred to various receptor molecules. One of these is 2-aminoethanol, $HOCH_2CH_2NH_2$, which is thereby converted to choline. Betaine is formed by dehydrogenation of choline, and can furnish a methyl group to homocysteine in an alternative pathway, catalyzed by methylpherase, for methionine biosynthesis.

3. Occurrence

Choline occurs widely in nature and, prepared synthetically, it is available as an article of commerce. Soybean lecithin and egg-yolk lecithin have been used as natural sources of choline for supplementing the diet. Other important natural-food sources include liver and certain legumes (18–22).

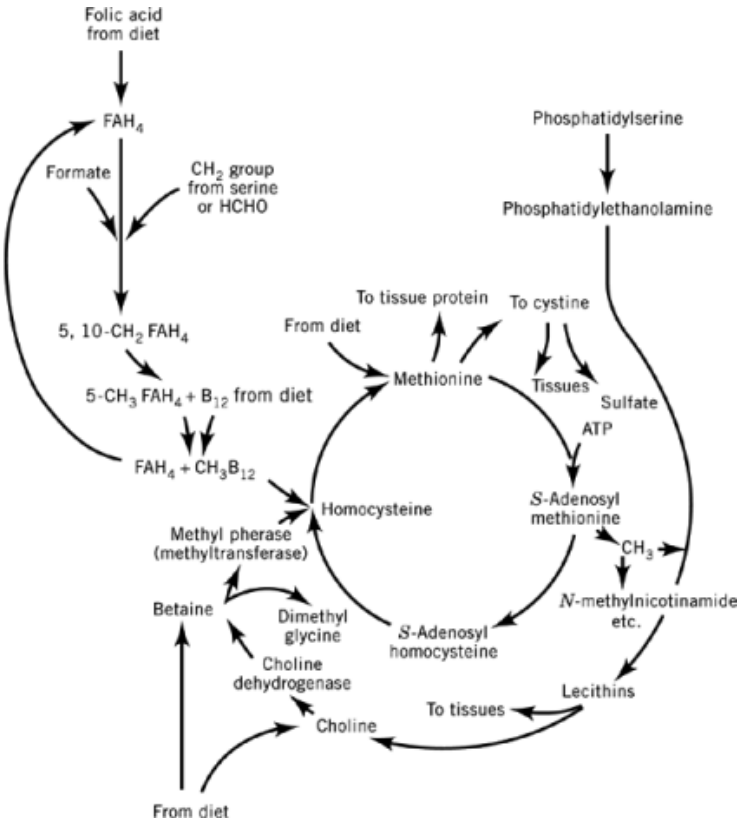


Fig. 1. The choline and methionine cycles showing the origin and disposition of labile methyl groups. FAH_4=tetrahydrofolic acid; CH_3B_{12}=methylated vitamin B_{12}; and ATP=adenosine triphosphate.

4. Preparation

Choline is not usually encountered as the free base but as a salt, most commonly, the chloride, $[(CH_3)_3N(CH_2CH_2OH)]^+Cl^-$;. As a quaternary ammonium hydroxide, choline reacts with hydrochloric acid to form the chloride and water, whereas primary, secondary, and tertiary amines combine with hydrochloric acid to form hydrochlorides.

An earlier procedure for the production of choline and its salts from natural sources, such as the hydrolysis of lecithin (23), has no present-day application. Choline is made from the reaction of trimethylamine with ethylene oxide [75-21-8] or ethylene chlorohydrin [107-07-3].

$$N(CH_3)_3 + ClCH_2CH_2OH \longrightarrow (CH_3)_3 N^+CH_2CH_2OH\ Cl^-$$

The chlorohydrin process (24) has been used for the preparation of acetyl-β-alkylcholine chloride (25). The preparation of salts may be carried out more economically by the neutralization of choline produced by the chlorohydrin synthesis. A modification produces choline carbonate as an intermediate that is converted to the desired salt (26). The most practical production procedure is that in which 300 parts of a 20% solution of trimethylamine is neutralized with 100 parts of concentrated hydrochloric acid, and the solution is treated for 3 h with 50 parts of ethylene oxide under pressure at 60°C (27).

5. Choline Salts

5.1. Choline Chloride

This compound [67-48-1] is a crystalline deliquescent salt, usually with a slight odor of trimethylamine (6). It is very soluble in water, freely soluble in alcohol, slightly soluble in acetone and chloroform, and practically insoluble in ether, benzene, and ligroin. Its aqueous solutions are neutral to litmus and are stable (4). The specific gravity of these solutions is a straight-line function between pure water and the value of 1.10 for the 80% solution, which represents the approximate limit of solubility. Choline chloride absorbs moisture from the atmosphere at relative humidities greater than 20% at 25.5°C.

5.2. Choline Dihydrogen Citrate

This compound [77-91-8] is a white, crystalline, granular substance possessing an acid taste, mp 105–107.5°C, and is freely soluble in water, very slightly soluble in alcohol, and practically insoluble in benzene, chloroform, and ether. The pH of a 25% solution is about 4.25.

Choline dihydrogen citrate $(CH_3)_3N(CH_2CH_2OH)C_6H_7O_7$, is prepared by methods similar to those for preparing choline chloride. It has the same pharmacological action as the chloride, but contains a lower proportion of choline. It is not as deliquescent as the chloride, and absorbs moisture from the atmosphere only at relative humidities greater than 56% at 25.5°C. It is more palatable than the chloride.

5.3. Tricholine Citrate Concentrate

This compound [546-63-4] is a clear, faintly yellow to light-green syrupy aqueous liquid containing $65.0 \pm 2.0\%$ tricholine citrate. It is usually has a slight amine odor. It should have a pH of 9.0–10.0 and should contain not more than 0.2% trimethylamine, 0.5% ethylene glycol, 10 ppm of formaldehyde, and 0.1% residue on ignition. Its limit for heavy metals is 20 ppm and it should contain more than 0.2% chlorides or sulfates.

5.4. Choline Bitartrate

This substance [87-67-2] is a white crystalline material possessing an acid taste. It melts at 149–153°C. Analysis by cobaltous chloride shows more than 99% as the bitartrate. Free ethylene glycol is less than 0.25%, with free alkali at 0.0%.

5.5. Others

Other choline salts available as commercial products include choline bicarbonate [78-73-9], choline salicylate [2016-36-6], and the bronchodilator choline theophyllinate [4499-40-5].

6. Analysis

In biological materials, various nonspecific precipitants have been used in the gravimetric determination of choline, including potassium triiodide, platinum chloride, gold chloride, and phosphotungstic acid (28). Choline may also be determined spectrophotometrically and by microbiological, enzymatic, and physiological assay methods.

Choline reineckate is used in the spectrophotometric determination of choline. Ammonium reineckate [13573-16-5] forms a water-insoluble complex with choline. The complex is soluble in acetone and a widely

used method for determination of choline is by light absorption of acetone solutions at 520 μm (8, 29–31). The sensitivity of the assay is as little as 100 ppm choline in plant and animal tissue (32).

The use of mutant 34486 of *Neurospora crassa* for the microbiological assay of choline has been described (8). A physiological method has also been used in which the choline is extracted after hydrolysis from a sample of biological material and acetylated. The acetylcholine is then assayed by a kymographic procedure, in which its effect in causing contraction of a piece of isolated rabbit intestine is measured (33).

Enzymatic methods have been described (34) as well as gc, lc, radiochemical, and fluorometric procedures for choline analysis (32).

7. Uses

Choline has a low toxicity, eg, LD_{50} (rat, oral) = 3 – 6 g/kg, (35, 36). It is used clinically and as a dietary supplement for poultry.

As a therapeutic agent, choline is administered orally in the form of syrups or elixers containing the chloride, citrates or bitartrate, or in the form of compressed tablets or capsules of the dihydrogen citrate. Choline is also given in small doses as a nutritional supplement in combination with a variety of other materials. In dry pharmaceutical-dosage forms, the dihydrogen citrate is usually preferred because of its lower tendency to absorb atmospheric moisture. Both salts have been used parenterally.

In the feeding of animals, choline is often added to chicken and turkey feeds as a dietary supplement (37–39). Its use in cattle feed has also been studied (40, 41).

Choline in the form of choline base (hydroxide) is a strong organic base with a pH of approximately 14. This product can have industrial applications where it is important to replace inorganic bases with organic materials. Choline base is currently used in the formulation of photoresist stripping products for use in the printed wire board industry. Dilute aqueous solutions (5%) of choline base that have very low concentrations of metallic ions have been utilized for applications in the semiconductor industry.

8. Economic Aspects

The world market for choline chloride used in animal feeds is estimated at 113,000 t on a 100% basis. The market for good grade choline chloride is a small market by comparison and is utilized mainly in the supplementation of infant formulas. Other choline salts are utilized solely in the human vitamin supplementation markets and are also small compared to animal feed usage.

There are nine primary producers of choline chloride within the world. These are listed in Table 1. There are also small producers located in Taiwan and the People's Republic of China.

Market prices for choline products in 1992 were as follows: 70% choline chloride(FeedGrade), $0.77/kg; choline dihydrogen citrate, $7.20/kg; and choline hydrogen tartrate, $9.50/kg.

9. Derivatives

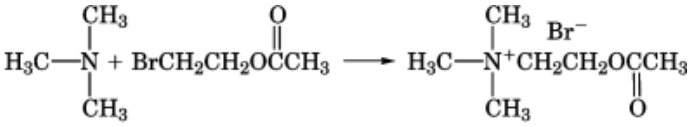
Important derivatives of choline are acetylcholine, acetyl-β-methylcholine, and carbamylcholine. Many other choline derivatives have been synthesized and studied, but have not been found satisfactory for clinical use.

Acetylcholine [51-84-3] occurs as the bromide [66-23-9] (Pragmoline) and the chloride [60-31-1] (Acecoline). The chloride is a hygroscopic, crystalline powder. It is very soluble in cold water and alcohol but is practically insoluble in diethyl ether. It is decomposed by hot water and alkalies.

Table 1. Choline Chloride Producers

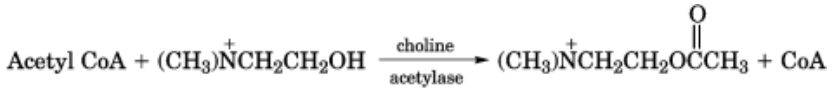
Company	Country
DuCoa	United States
Bioproducts, Inc.	United States
Chinook	Canada
I.C.I.	United Kingdom
U.C.B.	Belgium
Akzo	The Netherlands
BASF	Germany
Mitsubishi	Japan

Acetylcholine bromide can be prepared by **direct reaction of trimethylamine and β-bromoethyl acetate in benzene** (42).



acetylcholine bromide

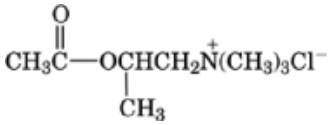
Acetylcholine is the product of the **reaction between choline and acetyl coenzyme A in the presence of choline acetylase** (41).



acetylcholine

Acetylcholine is a neurotransmitter at the neuromuscular junction in autonomic ganglia and at postganglionic parasympathetic nerve endings (see Neuroregulators). In the CNS, the motor-neuron collaterals to the Renshaw cells are cholinergic (43). In the rat brain, acetylcholine occurs in high concentrations in the interpeduncular and caudate nuclei (44). The LD_{50} (subcutaneous) of the chloride in rats is 250 mg/kg.

Acetyl-β-methylcholine chloride [62-51-1], **commonly called methacholine chloride**, is a parasympathomimetic bronchoconstrictor with clinical efficacy in bronchial asthma (45, 46).



Carbamylcholine chloride [51-83-2] **is also called carbachol** (47). Its principal use is as a parasympathomimetic in veterinary practice for large animals.

BIBLIOGRAPHY

"Choline" in *ECT* 1st ed., Vol. 3, pp. 919–927, by T. H. Jukes, American Cyanamid Co., Lederle Laboratories Div. and G. H. Schneller, American Cyanamid Co., Calco Chemical Div.; in *ECT* 2nd ed., Vol. 5, pp. 403–413, by T. H. Jukes, University of California, Berkeley; *ECT* 3rd ed., Vol. 6, pp. 19–28, by T. H. Jukes, University of California, Berkeley.

Cited Publications

1. C. L. Connor and I. L. Chaikoff, *Proc. Soc. Exptl. Biol. Med.* **39**, 356 (1938).
2. W. H. Griffith and D. J. Mulford, *J. Am. Chem. Soc.* **63**, 929 (1941).
3. C. W. Prince and W. C. M. Lewis, *Trans. Faraday Soc.* **29**, 775 (1933).
4. S. Budavari, ed., *The Merck Index*, 11th ed., Merck & Co., Inc., Rahway, N.J., 1989, p. 342.
5. E. Kahane and J. Lévy, *Biochimie de la Choline et de ses Dérivés*, Hermann, Paris, 1938.
6. *The United States Pharmacopeia*, 22nd rev. ed. (USP XXII), The United States Pharmacopeial Convention, Inc., Rockville, Md., 1990, p. 1736.
7. *Choline: Functions and Requirements*, DuCon, Highland, Ill., 1992.
8. W. H. Sebrell and R. S. Harris, eds., *The Vitamins*, 2nd ed., Vol. **3**, Academic Press, Inc., New York, 1971, 2–154.
9. H. Blumberg and E. V. McCollum, *Science* **93**, 598 (1941).
10. G. O. Broun and R. O. Muether, *J. Am. Med. Assoc.* **118**, 1403 (1942).
11. V. Du Vigneaud, S. Simmonds, J. P. Chandler, and M. Cohn, *J. Biol. Chem.* **165**, 639 (1946).
12. S. S. Kewar, J. H. Mangum, K. G. Scringeour, J. D. Brodies, and F. M. Huennekens, *Arch. Biochem. Biophys.* **116**, 305 (1966).
13. C. Entenman, I. L. Chaikoff, and M. Montgomery. *J. Biol. Chem.* **155**, 573 (1944).
14. J. V. Lowry, L. L. Ashburn, and W. H. Sebrell, *Quart. J. Studies Alc.* **6**, 271 (1945).
15. A. E. Schaefer and J. L. Knowles, *Proc. Soc. Exper. Biol. Med.* **77**, 655 (1951).
16. A. W. Moyer and V. Du Vigneaud, *J. Biol. Chem.* **143**, 373 (1942).
17. D. Stetten, Jr., *J. Biol. Chem.* **140**, 143 (1941).
18. A. Z. Hodson, *J. Nutr.* **29**, 137 (1945).
19. J. M. McIntire, B. S. Schweigert, and C. A. Elvehjem, *J. Nutr.* **28**, 219 (1944).
20. R. W. Engle, *J. Nutr.* **24**, 441 (1943).
21. S. M. McIntire, B. S. Schweigert, and C. A. Elvehjan, *J. Nutr.* **28**, 219 (1944).
22. J. J. Wurtman, *Nutrition and the Brain*, Vol. **5**, 1979.
23. Ger. Pat. 193,449 (Dec. 22, 1906), J. D. Reidel (to iengesellschaft Berlin).
24. U.S. Pat. 2,774,759 (1956), Blackett and Soliday (to American Cyanamid Co.).
25. U.S. Pat. 2,198,629 (Apr. 30, 1940), R. T. Major and H. T. Bonnett (to Merck & Co., Inc.).
26. Fr. Pat. 736,107 (Apr. 29, 1932), F. Korner.
27. U.S. Pat. 2,137,314 (Nov. 22, 1938), H. Ulrich and E. Ploetz (to I. G. Farbenindustrie A.G.).
28. I. Sakakibaia and T. Yoshinaga, *J. Biochem. (Japan)* **23**, 211 (1936).
29. D. Glick, *J. Biol. Chem.* **156**, 643 (1945).
30. I. Hanin, *Handbook of Chemical Assay Methods*, Raven Press Publishers, New York, 1974.
31. S. E. Valdes Martinez, *Analyst (London)* **108**(1290), 1114–1119 (1983).
32. A. S. Atwal, N. A. M. Eskin, and M. Vaisey-Genser, *Cereal Chem.* **57**, 367–369 (1980).
33. J. P. Fletcher, C. H. Best, and O. M. Solandt, *Biochem. J.* **29**, 2278 (1935).
34. M. Takayama, S. Itoh, T. Nagasaki, and I. Tanimizu, *Clin. Chim. Acta* **79**, 93–98 (1977).
35. H. C. Hodge, *Proc. Soc. Exptl. Biol. Med.* **58**, 212 (1945).
36. M. W. Neumann and H. C. Hodge, *Proc. Soc. Exptl. Biol. Med.* **58**, 87 (1945).
37. V. K. Tsiagbe, C. W. Kang, and M. L. Sunde, *Poult. Sci.* **61**(10), 2060–2064 (1982).
38. M. L. Sunde, *Proc. Cornell Nutr. Conf. Feed Manuf.*, 67–73 (1982).
39. Jpn. Kokai 108 787 (1981), Yakult.
40. R. A. Erdman, R. D. Shaver, and J. H. Vandersall, *J. Dairy Sci.* **67**(2), 410–415 (1984).
41. T. S. Rumsey and R. R. Oltjen, *J. Anim. Sci.* **41**, 416 (1975).

42. E. Fourneau and J. Page, *Bull. Soc. Chim. (France)* **15**, 544 (1914).
43. J. R. Cooper, F. E. Bloom, and R. H. Roth, *The Biochemical Basis of Neuropharmacology*, 3rd ed., Oxford University Press, New York, 1978.
44. M. A. Lipton, A. DiMascio, and K. F. Killam, eds., *Psychopharmacology—A Generation of Progress*, Raven Press, New York, 1978.
45. Ref. 4, p. 14.
46. Ref. 4, p. 935.
47. Ref. 6, p. 838.

CURTIS E. GIDDING
DuCoa

Related Articles

Lecithin; Feeds and feed additives