CHAPTER 30

Choline-containing phospholipids and treatment of adult-onset dementia disorders

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List of abbreviations

ACh Acetylcholine
AChR Acetylcholine receptor
AD Alzheimer's disease
CCPL Choline-containing phospholipids
ChAT Choline acetyltransferase
ChE Cholinesterase
GPC Choline alphoscerate
PL Phospholipids

Mini-dictionary of terms

Acetylcholine ACh is an organic chemical that acts as a neurotransmitter in the brain and in the body of many different animals, including humans.

Acetyltransferase acetyltransferase (also knows as *transacetylase*) is a kind of transferase enzyme that transfers an acetyl group.

Cholinergic neurons a cholinergic neuron is a nerve cell that primarily uses ACh as a neurotransmitter to send its messages. Many neurological systems in our body are cholinergic. Cholinergic neurons represent the primary source of ACh for the cerebral cortex.

Neurotransmitter neurotransmitters are endogenous chemicals that allow neurotransmission. They are a type of chemical messenger that transmits signals across a chemical synapse from one neuron to another neuron, muscle cell, or gland cell.

Phospholipids PLs are lipids that are a major component of all cell membranes. They can form lipid bilayers thanks to their amphiphilic characteristic. The structure of the PL molecule commonly consists of two hydrophobic fatty acid "tails" and a hydrophilic "head" consisting of a phosphate group.

Introduction

Choline, the main precursor of the brain and autonomic neurotransmitter acetylcholine (ACh), is a quaternary ammonium salt (Fig. 30.1). Choline is an essential component of various membrane phospholipids (PLs) and contributes to the structural integrity of cell membranes. Choline-containing PLs (CCPLs) include phosphatidylcholine (PC),

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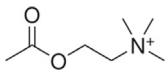

Figure 30.1 Acetylcholine. The structural formula of acetylcholine is shown.

phosphatidylserine, sphingomyelin (SM), cytidine 5′-diphosphocholine (CDP—choline or citicoline), and choline alphoscerate (GPC). PC is the major PL in most eukaryotic cells and is involved in SM synthesis, choline/choline metabolite regeneration, and fatty acid/GPC formation. Moreover, the role of CCPLs in the biosynthesis of various neurotransmitters, ACh included, has been extensively demonstrated.

This chapter summarizes the main preclinical and clinical data on the use of CCPLs in the treatment of adult-onset dementia and suggests that, if properly used, these compounds could still have a place in the pharmacotherapy of dementia disorders.

Cholinergic neurotransmission in Alzheimer's disease and the cholinergic approach in the treatment of Alzheimer's disease

Dr. Aloysius Alzheimer observed feeble dementia in his patient Auguste Deter in 1901, and there are still no specific treatments for the disease, although several approaches have been proposed to alleviate its cognitive and noncognitive symptoms (Amenta, Parnetti, Gallai, & Wallin, 2001; Parnetti, Mignini, Tomassoni, Traini, & Amenta, 2007).

ACh, the first neurotransmitter ever identified, is largely diffused in the central, peripheral, and autonomic nervous systems. In the central nervous system, the cholinergic neurons are widely distributed. Nerve cell bodies are located in the spinal cord, hindbrain, medial habenula, mesopontine region, basal forebrain, striatum, olfactory tubercle, and islands of Calleja complex. From these nerve cell bodies originate projections innervating almost all brain areas.

The cholinergic system plays a role in learning and memory processes. Endogenous ACh released by neurons located in the basal forebrain (the *nucleus basalis magnocellularis* of Meynert and in the septal nuclei) is involved in the modulation of acquisition, encoding, consolidation, extinction, and retrieval of memory. Cholinergic projections originating from the nucleus basalis magnocellularis supply the cerebral cortex, whereas those originating from septal nuclei supply the hippocampus. The hippocampus is a cerebral region involved in a variety of functions, including learning and memory processes. It plays an important role in the elaboration of mapping and working memory, in the improvement of selective attention, and in the inhibition of inappropriate behavioral responses. The hippocampus shows a high sensitivity to aging and is involved in the pathogenesis of learning and memory dysfunction occurring with age and in adult-onset dementia disorders (Amenta et al., 2001; Parnetti et al., 2007).

The important role of the cholinergic neurons originating from the nucleus basalis of Meynert and from septal nuclei in memory is highlighted by the fact that a specific degeneration of these neurons takes place in Alzheimer's disease (AD) and contributes

to the memory loss exhibited by AD patients (Amenta et al., 2001). The observation of a loss of the ACh biosynthetic enzyme choline acetyltransferase (ChAT) in the cerebral cortex of AD patients stimulated the development of cholinergic strategies to counter cognitive dysfunction typical of adult-onset dementia, including AD itself (Amenta et al., 2001).

The observation that administration of the muscarinic antagonist scopolamine to healthy young subjects induced a cognitive impairment resembling that found in adult-onset dementia, and the subsequent demonstration of a remarkable decrease in ChAT activity in the cerebral cortex and hippocampus in AD patients, contributed to the development of the cholinergic hypothesis of geriatric memory dysfunctions (Bartus, Dean, Pontecorvo, & Flicker, 1985). The cholinergic system is not the only neurotransmitter system affected in adult-onset cognitive impairment, AD included. However, changes in cholinergic function are implicated in the pathogenesis of learning and memory alterations occurring in adult-onset dementia (Avery, Baker, & Asthana, 1997; Muir, 1997). An analysis of the involvement of cholinergic receptors in cognitive functions has shown that central muscarinic and nicotinic cholinergic receptors might be involved in learning and memory through complex mechanisms. Studies of the brain of aged subjects or of patients suffering from AD have shown a marked loss of ChAT and of nicotinic cholinergic receptors (Davies & Maloney, 1976; Hellström-Lindahl, Mousavi, Zhang, Ravid, & Nordberg, 1999). A correlation between the loss of cortical cholinergic synapses and cognitive decline was demonstrated, as well as the existence of a close relationship between this loss and the decrease in high-affinity cholinergic receptors. The different pharmacological treatments proposed or tested included intervention with ACh precursors, stimulation of ACh release, use of muscarinic or nicotinic receptor agonists, and acetylcholinesterase (AChE) inhibition (Amenta et al., 2001; Parnetti et al., 2007).

Choline-containing phospholipids and the precursor-loading approach for treating age-related dementia disorders

PLs (i.e., PC, phosphatidylserine, phosphatidylethanolamine, and SM) play an important role in cellular constitution and metabolism and are relevant components of various cell membranes (i.e., cells, mitochondria, endoplasmic reticulum, Golgi apparatus, peroxisomes, and lysosomes). Choline is an essential nutrient with a complex role in the body. Choline is necessary for the biosynthesis of the neurotransmitter ACh, cell-membrane signaling PLs, lipid transport (lipoproteins), and methyl-group metabolism (plasmatic homocysteine reduction). Choline is also the major dietary source of methyl groups via the synthesis of *S*-adenosylmethionine (Tayebati & Amenta, 2013).

Cholinergic precursor-loading therapy was the first approach tried to restore deficient cholinergic neurotransmission and to relieve cognitive impairment in dementia disorders.

Given the controversial nature of the existence of a direct role of choline in ACh release, and given that an increase in brain free choline does not always imply an increase in brain ACh (Tayebati & Amenta, 2013), it was CCPLs that were initially proposed for treating adult-onset dementia disorders.

ACh is a neurotransmitter derived from choline and acetyl-coenzyme A (Fig. 30.2). The biosynthesis is catalyzed by the enzyme ChAT, and the biosynthesis of the cosubstrate acetyl-coenzyme A is not specific for cholinergic neurons. Nervous tissue is unable to synthesize choline, which derives from the diet and is delivered to neurons through the bloodstream. ACh released from cholinergic synapses is hydrolyzed by AChE into choline and acetyl-coenzyme A (Fig. 30.2). Approximately 50% of choline derived from ACh hydrolysis is recovered through a high-affinity transporter. Neurons require, therefore, a further supply of choline to synthesize ACh (Amenta et al., 2001).

Cholinergic neurons probably have a particular avidity for choline, and it has been hypothesized that when the provision of choline is insufficient, neurons obtain it by hydrolyzing membrane PLs. This mechanism, known as autocannibalism, may render cholinergic neurons more susceptible to injury and may contribute to cholinergic neuron degeneration (Wurtman, Blussztajn, & Ulus, 1990).

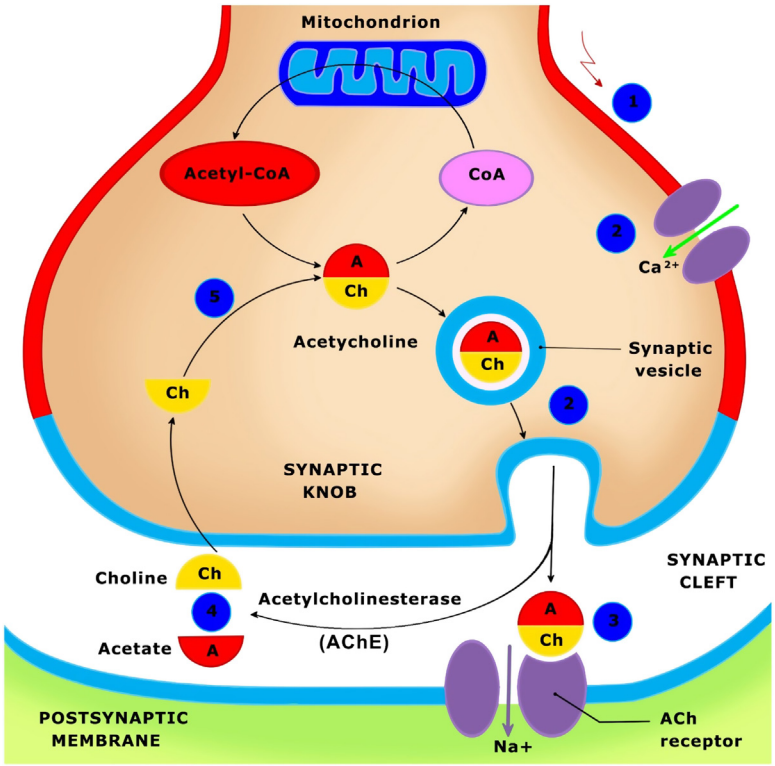

Figure 30.2 Cholinergic synapsis. Acetylcholine processing in a cholinergic synapse is shown. After the release, acetylcholine is broken down by the enzyme acetylcholinesterase.

The CCPLs proposed for the treatment of age-related dementia disorders include lecithin, phosphatidylserine, CDP—choline, and GPC. The main evidence of their activity is reviewed next.

Lecithin

Lecithin is a CCPL representing the main source of choline. Lecithin has been shown to increase serum choline levels more effectively than orally administered choline (Wurtman et al., 1990). Apparently, lecithin may accelerate ACh synthesis in the brain through enhanced availability of choline. Lecithin has been tried out in the treatment of dementia, alone or in combination with an acetylcholinesterase inhibitors (AChEI). Lecithin is largely available as a nutraceutical.

The role of this compound in dementia and in cognitive impairment was the focus of a Cochrane Review in 2009 (Higgins & Flicker, 2003). In the field of dementia and cognitive impairment, lecithin was investigated in 21 studies. Eight studies used the compound alone, others in association with tacrine, physostigmine, and piracetam.

The Cochrane analysis selected 12 randomized trials involving patients with AD (265 patients), parkinsonian dementia (21 patients), and individual memory problems (90 patients). No trials reported any clear clinical benefit of lecithin for AD or parkinsonian dementia. Only a few trials have led to data for meta-analyses. The only statistically significant result was in favor of placebo for adverse events, based on one single trial. Relevant results in favor of lecithin were obtained in a trial of subjects with individual memory problems (Higgins & Flicker, 2003).

Phosphatidylserine

Phosphatidylserine (Fig. 30.3) is a CCPL largely diffused in animal tissues (brain, liver, kidney, heart, and spleen), bluefish, soy, and egg yolk. Phosphatidylserine is a nutraceutical that was originally obtained from the distillation of bovine brains, which show a high

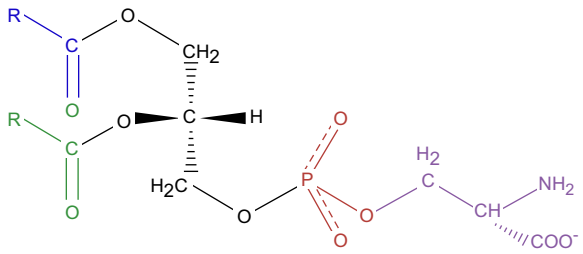

Figure 30.3 Phosphatidylserine. The structural formula of phosphatidylserine is shown. The different components of the phosphatidylserines are colored as described below: Blue and green, variable fatty acid groups; black, glycerol; red, phosphate; purple, serine.

concentration of this PL. After the epidemics of bovine spongiform encephalopathy, soy lecithin became the main source of phosphatidylserine.

Supplementation of this PL is based on the observation that reduced levels of DHA—phosphatidylserine in the cerebral cortex are associated with conversion of mild cognitive impairment to overt dementia (Olivera-Pueyo & Pelegrín-Valero, 2017). It has been suggested that phosphatidylserine content decreases in aging in parallel to the increase in cholesterol in neuronal membranes. These changes modify cell membrane viscosity by reducing enzymatic catalysis with consequent higher susceptibility of nerve cells to injury (Olivera-Pueyo & Pelegrín-Valero, 2017).

Seven clinical trials have investigated the effects of phosphatidylserine administration in AD. These studies, performed between 1986 and 1994, suggested some beneficial effects of the compound, though without providing conclusive evidence. More recent studies using phosphatidylserine as a supplement in patients with mild cognitive impairment or very early stage dementia have reported significant improvement in learning, memory, and verbal fluency, as well as visual learning, attention, communication, initiative, and socialization. Another study has reported reduced apathy, increased motivation and interest, and improved memory (Olivera-Pueyo & Pelegrín-Valero, 2017).

The limited number of patients investigated in the aforementioned studies and the lack of established diagnostic criteria do not allow one to draw a certain conclusion about a possible role of phosphatidylserine as a therapeutic agent in dementia.

CDP-choline

CDP—choline, or citicoline, is a CCPL composed of cytidine and choline linked together by a diphosphate bridge (Fig. 30.4). Citicoline is the international nonproprietary name of CDP—choline. Citicoline is marketed as a prescription-only drug in several European countries and in Japan, and as a nutraceutical in the United States and in Europe. CDP—choline is an intermediate in the synthesis of PC in Kennedy's pathway. PC is a cell membrane component that is degraded during cerebral ischemia to free fatty acids and free radicals (Parnetti et al., 2007). The functions of CDP—choline include the repair of the neural membrane through the synthesis of PC, the reduction of accumulated

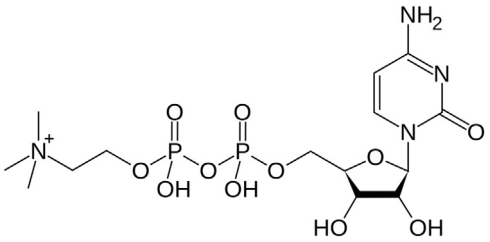

Figure 30.4 Citicoline. The structural formula of citicoline is shown.

fat responsible for increased cognitive deficit, and an increase in ACh levels. In poststroke patients, a neuroprotective effect of CDP—choline, contributing to significant improvement in temporal orientation, attention tasks, and executive function, has been reported, along with an ability, albeit in an experimental setting, to assist in neural repair. Citicoline reduced the duration of hospitalization and improved the recovery of neurological function and level of consciousness (Colucci et al., 2012). The activity of citicoline on the cognitive domain was also analyzed in a meta-analysis evaluating 13 studies carried out from the 1970s to the early 2000s, which demonstrated the effectiveness of the compound in the treatment of cognitive, emotional, and behavioral disorders (Colucci et al., 2012). These effects were attributed to the effect of citicoline on metabolic activation and were noticeable in patients with dementia of degenerative and/or vascular origin and in patients with cerebrovascular disease (Colucci et al., 2012).

More recent studies have reviewed the pharmacological profile, kinetics, and possible new uses of citicoline (Gareri et al., 2015). The CITIRIVAD Study investigated the effects of a combined treatment with the AChEI rivastigmine and citicoline in AD and in mixed dementia. This showed the greater effectiveness of a combined administration of citicoline plus rivastigmine (an AChEI) versus AChEI alone (Castagna, Cotroneo, Ruotolo, & Gareri, 2016). A 2017 trial investigated the effects of an oral 9-month treatment with citicoline in addition to AChEI treatment, compared with the AChEI alone, in AD patients (Citicholinage Study). An increase in the Mini-Mental State Examination (MMSE) scores was noticeable in CDP—choline-treated patients (Gareri et al., 2017). The Citicholinage Study concluded that coadministration of citicoline and AChEI supports the combined administration in managing the disease, by slowing disease progression (Gareri et al., 2017).

The effectiveness of citicoline was proposed in patients with traumatic brain injury, probably due to its effect on edema (Colucci et al., 2012). However, more recent studies did not confirm this effect. Citicoline is a compound that has been available in the pharmaceutical market since the beginning of the 1980s; more recently it has also been marketed as a dietary supplement. Several studies have demonstrated positive effects of the compound on cognition, whereas other investigations failed to confirm the positive results in the cognitive domain. In view of these discrepancies, additional clinical studies are necessary to confirm the potential benefits of citicoline in the treatment of adult-onset dementia disorders.

Choline alphoscerate

GPC, or α-glycerylphosphorylcholine (Fig. 30.5), is a semisynthetic derivative of lecithin. Following oral administration, it is converted to phosphorylcholine, a metabolically active form of choline able to reach the cholinergic nerve terminals, where it increases ACh synthesis, levels, and release (Amenta & Tayebati, 2008; Amenta et al., 2014).

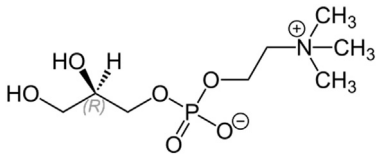

Figure 30.5 Choline alphoscerate. The structural formula of α-GPC (choline alphoscerate) is shown.

Although GPC has been on the pharmaceutical market since 1987, its interest seems to have suffered since the introduction of the therapy with cholinesterase inhibitors. Since 2010 there has been renewed attention paid to this compound, with preclinical studies, clinical investigations, and review articles published in the literature (Traini, Bramanti, & Amenta, 2013).

GPC interferes with brain PL metabolism and increases brain choline and ACh levels and release in rat hippocampus, facilitates learning and memory in experimental animals, improves brain transduction mechanisms, and decreases the age-dependent structural changes occurring in the rat frontal cortex and hippocampus. Moreover, this compound contributes to anabolic processes responsible for membrane PL and glycerolipid synthesis, positively influencing membrane fluidity. GPC was shown to ameliorate cognitive deficit in experimental models of aging brain and to reverse mnemonic deficits induced by scopolamine administration (Traini et al., 2013).

A restorative role of GPC on the central cholinergic system was documented by studies performed on old rodents. Neuroprotective effects of GPC were also reported in a rodent model of altered cholinergic neurotransmission caused by lesions of the nucleus basalis magnocellularis, which constitutes the main source of cholinergic innervation of the cerebral neocortex. A positive effect of treatment with GPC on hippocampus microanatomy and glial reaction was documented in spontaneously hypertensive rats. In a test of the cholinergic precursors lecithin, CDP—choline, and GPC, the last elicited the most relevant stimulation of vesicular ACh and choline transporters in the same model of brain vascular injury. This suggests that GPC is an enhancer of central cholinergic neurotransmission (Traini et al., 2013).

The activity of CCPLs on brain PL biosynthesis may influence brain metabolism and different neurotransmitter systems (Fig. 30.6). Based on the observation that the CCPL CDP—choline has a monoaminergic profile, an investigation was carried out on the activity of GPC on brain dopamine and serotonin levels and on dopamine plasma membrane transporter, vesicular monoamine transporters 1 and 2, serotonin transporter, and norepinephrine transporter (Traini et al., 2013). Administration of the compound increased dopamine levels in frontal cortex and cerebellum and serotonin levels in frontal cortex and striatum. It also stimulated the dopamine plasma membrane transporter in the frontal cortex and cerebellum. This investigation concluded that GPC also possesses a monoaminergic profile and interferes to some extent with brain monoamine transporters (Traini et al., 2013).

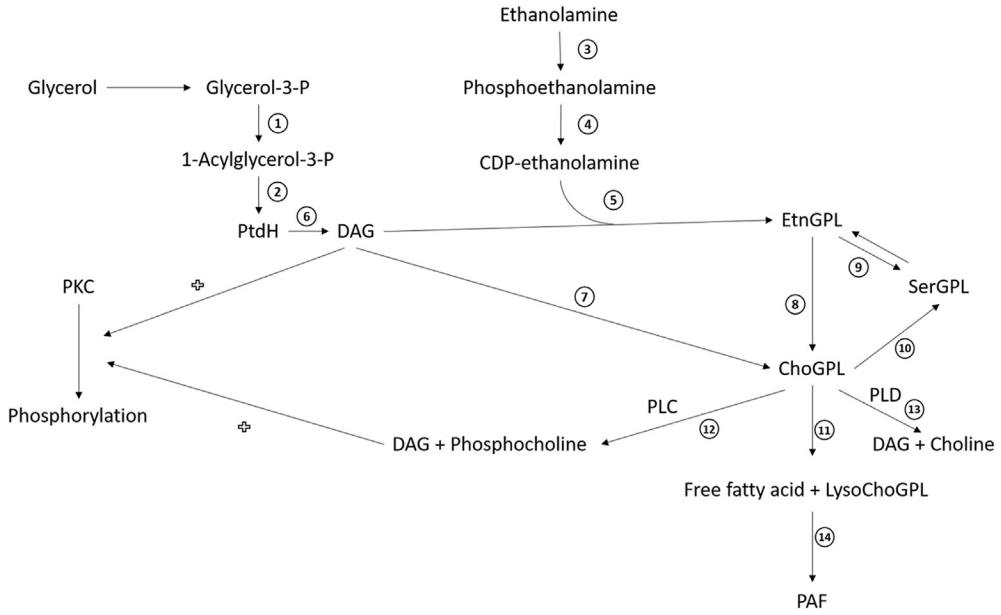

Figure 30.6 Pathways of biosynthesis of glycerophospholipids. Glycerol-3-phosphate acyltransferase (1); lysophosphatidic acid acyltransferase (2); ethanolamine kinase (3); ethanolamine cytidylyltransferase (4); CDP—phosphoethanolamine cytidylyltransferase (5); phosphatidic acid phosphatase (6); CDP—phosphocholine cytidylyltransferase (7); phosphatidylethanolamine methyltransferase (8); phosphatidylserine decarboxylase (9); phosphatidylserine synthase (10); phospholipase A2 (11); phospholipase C (*PLC*) (12); phospholipase D (*PLD*) (13); PAF synthesizing enzymes (14). *CDP—ethanolamine*, cytidine 5′-diphosphoethanolamine; *ChoGPL*, choline glycerophospholipid; *DAG*, diacylglycerol; *EtnGPL*, ethanolamine glycerophospholipid; *LysoChoGPL*, choline lysoglycerophospholipid; *PKC*, protein kinase C; *PtdH*, phosphatidic acid; *SerGPL*, serine glycerophospholipid.

The majority of clinical studies available on the effect of GPC on cognitive function in neurodegenerative and cerebrovascular disorders were detailed in two review articles from our group (Parnetti et al., 2007; Traini et al., 2013). Studies published before 2001 investigated 1570 patients, of which 854 were in controlled trials. Patients examined were affected by dementia of degenerative, vascular, or combined origin, such as dementia of the Alzheimer's type, vascular dementia, and acute cerebrovascular diseases (transitory ischemic attack and stroke) (Amenta et al., 2001). Test batteries for assessing the effect of GPC on cognitive domains were primarily the MMSE for disorders of neurodegenerative origin and the Sandoz Clinical Assessment Geriatric scale for disorders of vascular origin (e.g., vascular dementia) (Traini et al., 2013). The activity of GPC was also investigated in 789 patients with cognitive impairment of vascular origin. Three homogeneous-case trials evaluated 408 patients, while three combined-case trials included 381 patients with vascular dementia. Treatment with GPC improved memory and attention, as well as affective and somatic symptoms (fatigue, vertigo). The effects of GPC were greater than those of placebo and of the same extent as or superior to those of reference compounds (Amenta et al., 2001).

Preclinical studies showed that the association of GPC with (acetyl)cholinesterase inhibitors enhances the effects of both drugs on cholinergic neurotransmission. This prompted the development in Italy of an independent trial known as "Effect of association between a cholinesterase inhibitor and GPC on cognitive deficits in AD associated with cerebrovascular impairment" (ASCOMALVA). This controlled, randomized, and double-blind multicenter (two neurological units in Naples and Mantua) study was designed to assess whether the association between the AChEI donepezil (at the daily dose of 10 mg) and GPC (at the daily dose of 1200 mg) was accompanied by changes in MMSE; Alzheimer's Disease Assessment Scale, Cognitive subscale (ADAS-Cog); Basic Activities of Daily Living; Instrumental Activities of Daily Living; and Neuropsychiatric Inventory (NPI) scores. The last included the evaluation of measures of severity and of caregiver stress.

The patients were between 56 and 91 years of age (mean 75 ± 10 years) and were included in the protocol with an MMSE score between 15 and 23. They showed ischemic brain damage documented by neuroimaging (MRI and CT scan), with a score ≥2 in at least one subfield (white matter or basal ganglia), according to the new rating scale for Age-Related White Matter Changes. Recruited patients were then randomly allotted to an active treatment group (donepezil + GPC) or to a reference treatment group (donepezil + placebo) and were treated for 48 months; examination took place at recruitment and after 3, 6, 9, 12, 18, 24, 36, and 48 months of treatment. Consistent with literature data, in patients allotted to the reference treatment group (donepezil + placebo), a slight time-dependent worsening of MMSE and ADAS-Cog scores was found. Treatment with donepezil + GPC (active treatment) countered the decline in MMSE and ADAS-Cog scores. The effect of the association on psychometric tests was statistically significant after 12 months of treatment (Figs. 30.7 and 30.8) (Amenta et al., 2014).

Another aspect explored by the ASCOMALVA trial was the influence of GPC on apathy. Apathy is a common symptom in AD and has a significant impact on several outcomes. No treatment has proven to be effective against apathy, although the administration of cholinesterase inhibitors has been associated with modest improvements in the short term. The ASCOMALVA trial measured apathy at baseline and at 3, 6, 9, 12, 18, and 24 months, through the apathy subtest of the NPI, in 113 mild–moderate AD patients. Two matched groups were compared: group A (n = 56 subjects), treated with donepezil plus GPC, and group 2B (n = 57 subjects), treated with donepezil alone. The combination of donepezil plus GPC was more effective than donepezil alone in countering symptoms of apathy in AD. This suggests that the availability in the brain of a higher amount of ACh may affect apathy in AD subjects with spared executive functions (Amenta & Tayebati, 2008).

Behavioral and psychological symptoms of dementia (BPSD) are a group of psychological reactions, psychiatric symptoms, and behaviors commonly found in AD.

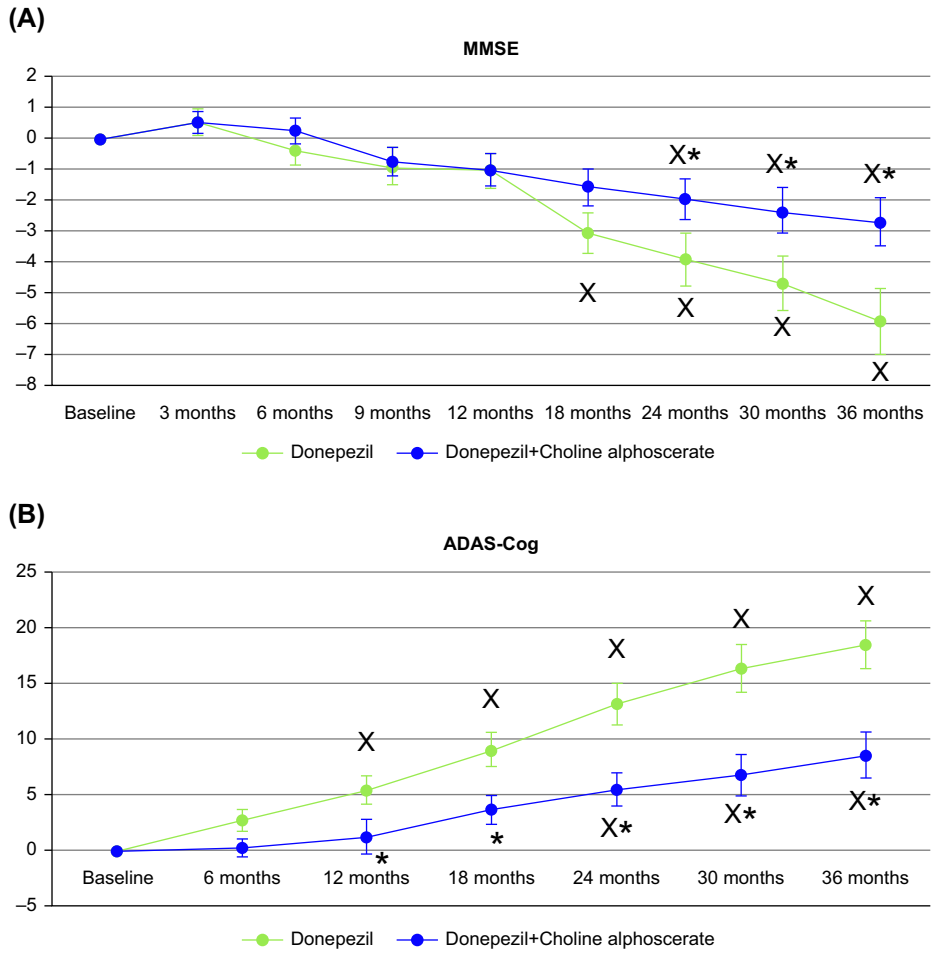

Figure 30.7 MMSE and ADAS-Cog scores in the 36 months of observation of the ASCOMALVA trial. The first results of the ASCOMALVA trial after 12 months of treatment are shown, including evaluation of cognitive tests. Data are the means of the difference in the scores from baseline ± SEM × $P > .05$ versus baseline; *$P < .05$ of donepezil versus associated therapy. *ADAS-Cog*, Alzheimer's Disease Assessment Scale, Cognitive subscale; *MMSE*, Mini-Mental State Examination.

Four clusters of BPSD have been described: mood disorders (depression, anxiety, and apathy), psychotic symptoms (delusions and hallucinations), aberrant motor behaviors (pacing, wandering, and other purposeless behaviors), and inappropriate behaviors (agitation, disinhibition, and euphoria). Most of them are attributed to ACh deficiency. The ASCOMALVA trial investigated the influence of the addition of GPC to donepezil on BPSD at the baseline and after 24 months in 113 mild–moderate AD patients. BPSD were analyzed through the NPI. NPI data revealed a significant decrease in BPSD severity and caregiver distress in patients of group A compared with group B. Mood

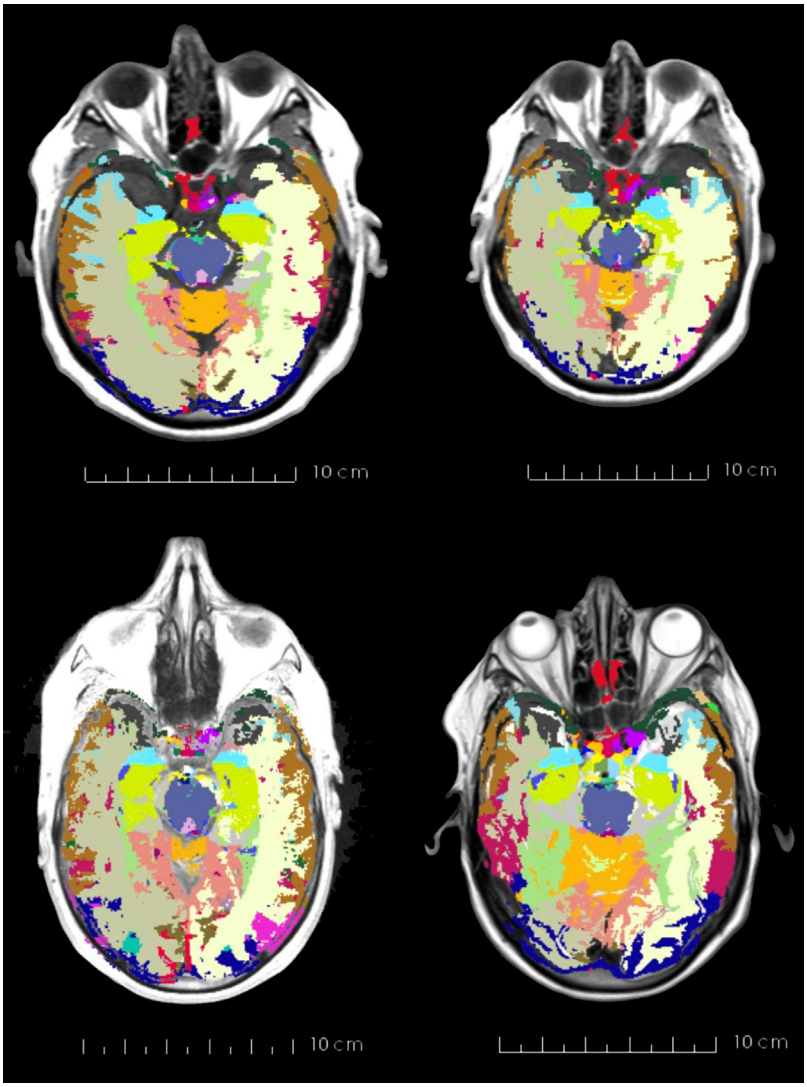

Figure 30.8 Nuclear magnetic resonance images of two patients from the ASCOMALVA study. Images from the ASCOMALVA study, relating to two patients belonging respectively to the donepezil + placebo group (first row) and to the donepezil + choline alphoscerate group (second row) are shown. The hippocampal area is indicated in the parceling from the yellow area. *(The images are original and are modified MRI used in morphometric evaluation of brain volume.)*

disorders (depression, anxiety, and apathy) were significantly decreased in subjects treated with donepezil and GPC, while their severity and frequency were increased in the other group. In conclusion, patients treated with donepezil plus GPC showed a lower level of behavioral disturbances than subjects treated with donepezil only, suggesting that the combination may have beneficial effects (Carotenuto et al., 2017).

A further contribution of the ASCOMALVA trial was the analysis of the influence of a treatment with GPC associated with a treatment with donepezil on brain atrophy in AD. Cerebral atrophy is a common feature of neurodegenerative disorders, such as AD. This pathology includes a loss of gyri and sulci in the temporal lobe and parietal lobe and in parts of the frontal cortex and of the cingulate gyrus. Participants of the ASCOMALVA trial underwent yearly MRI for diagnostic purposes. In 56 patients who achieved 3 years of therapy, brain MRI was analyzed by voxel morphometry techniques. After 3 years of treatment, in patients treated with donepezil plus GPC, a reduction in the volume loss of the gray matter (with a concomitant increase in the volume of the ventriculi and cerebrospinal fluid space) was observed, compared with the reference group, treated with donepezil only. The areas in which brain atrophy was sensitive to combination treatment with GPC were the frontal and temporal lobes, hippocampus, amygdala, and basal ganglia. In other areas, no significant differences were noticeable between the two groups. Morphological data were confirmed by neuropsychological assessment performed alongside the trial.

In conclusion, a cholinergic precursor-loading strategy with GPC in combination with donepezil counters to some extent the atrophy occurring in some brain areas of AD patients. The parallel observation of an improvement in cognitive and functional tests in those patients suggests that morphological changes observed may also have functional relevance.

Conclusions and future perspectives

One option for improving impaired cerebrocortical cholinergic neurotransmission consists in the inhibition of endogenous ACh degradation by using ChE inhibitors (Amenta et al., 2001; Parnetti et al., 2007).

Unfortunately, based on available data from clinical trials, the usefulness of cholinergic precursors in the treatment of AD appears doubtful. The dietary supplementation of free choline, for instance, does not increase the synthesis or the release of ACh in the brain (Amenta & Tayebati, 2008; Sigala et al., 1992; Traini et al., 2013). However, some CCPLs involved in ACh synthesis play an important role in cholinergic neurotransmission, and increase the availability and release of ACh. These PLs include phosphatidylserine, CDP—choline, and GPC. Among these CCPLs, GPC is the compound inducing the most marked release of ACh in animal models. Clinical trials performed with CDP—choline and GPC have shown that these compounds have a more pronounced activity than choline GPC. The results of clinical trials with these compounds were, however, obtained on relatively small numbers of patients. Nonetheless, the results are encouraging to further studies on these molecules.

The ASCOMALVA trial is also of interest as it demonstrated that a cholinergic precursor-loading strategy with GPC and donepezil counters to some extent the atrophy occurring in some brain areas of AD patients. The parallel observation of an improvement

in cognitive and functional tests in those patients suggests that morphological changes observed may also have functional relevance.

It is worth mentioning that the main effects of CCPLs were observed after 1–3 years of treatment. This suggests a reconsideration of the time of observation needed for identifying the therapeutic effects of drugs in a brain disorder as complex and as long lasting as AD.

Key facts about choline and phospholipids

- Choline and PLs are important, not only for structural purposes within the lipid bilayer, but also for preventing certain diseases and general disorders at the cellular level.
- PLs are present in various foods, such as milk, chicken eggs, soy, fish eggs, and sunflower seeds.
- PC, phosphatidylserine, and phosphatidylethanolamine are three of the numerous forms of PLs with specific and specialized functions.
- PC is the most abundant PL in the human body; it performs functions that aid in neural processes, including memory, reasoning, and learning.
- Phosphatidylserine is produced by almost every cell in the body, but also needs to be included in the diet to ensure that all cells receive an adequate supply.
- Phosphatidylethanolamine is known for playing a significant role in cellular membrane formation, cognition, and memory.
- A proper daily dietary intake of choline is necessary to decrease the risk of many problems such as fatty liver, liver damage, cognitive decline, nerve damage, fatigue, and memory loss.
- Choline is responsible for good cognitive abilities. Therefore, diets consisting of fish, leafy green vegetables, and soy are essential for maintaining good health.

Summary points

- Cholinergic neurons, for example, ACh-delivering neurons, are essentially associated with the pathogenesis of AD.
- Phosphatidylserine and, in addition, CDP–choline increase ACh substance and discharge.
- ACh and choline are very important for memory and intellectual capacity.
- Choline takes part in various neurochemical processes. It is an antecedent and a metabolite of ACh, it has a role in single-carbon digestion, and it is a fundamental segment of various film PLs.
- Hypoxia/ischemia leads to hydrolysis of PC and a further breakdown of glycerophosphocholine.
- An expansion of free choline does not generally imply expansion of ACh.

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