Presynaptic Control of Release of Amine Neurotransmitters by Precursor Levels

Richard J. Wurtman

The amounts of such aminergic neurotransmitters as serotonin, dopamine, norepinephrine, and acetylcholine that are released into synapses, spontaneously and when the neurons fire, can be affected by the concentrations of their nutrient precursors tryptophan, tyrosine, and dopamine and can thus be influenced by eating "real" foods or taking the pure precursors. Simple laws can apparently enable the investigator to predict when precursor levels will or will not have such effects.

Introduction

That there can be wide variations in the amounts of acetylcholine, serotonin, and the catecholamines that neurons release, spontaneously or by firing, is now well established. One process that causes such variations involves receptors on the neuron's own presynaptic terminals. When activated by neurotransmitter molecules that the neuron has released into a synapse, by concurrently released neuromodulators (like adenosine), or by transmitters (like the enkephalins) secreted by different neurons at axoaxonal synapses, these receptors initiate intracellular events that diminish subsequent transmitter release.

This article describes another type of process that modulates the release of amine neurotransmitters: changes in their rates of synthesis, caused by variations in the availability of the precursor substances, the concentrations of which control these rates. The precursors themselves are nutrients, and their concentrations in blood and neural tissue can be raised or lowered physiologically (by eating certain "real" foods) or experimentally (by administering the pure nu-

R. J. Wurtman is in the Department of Brain and Cognitive Sciences, and the Clinical Research Center at Massachusetts Institute of Technology, Building E25-604, Cambridge, MA 02139, USA.

trients). Such variations in precursor levels may uniformly affect the outputs of all of the neurons that release a particular aminergic transmitter or, in the case of dopamine and norepinephrine, only those neurons that happen to be undergoing prolonged periods of physiological activity.

Unlike the receptor-mediated modulations of transmitter release, this type of modulation depends primarily on metabolic events that occur outside the brain and that are influenced by a particular type of behavior, i.e., eating. Indeed, the primary physiological role of this dependency may be sensory, i.e., to provide the omnivore's brain with on-line data about plasma nutrient levels, so that the brain can then decide what and when to eat next. However, because precursor-dependent neurotransmitters are involved in a wide variety of normal (and pathological) brain mechanisms besides those controlling the intake and utilization of nutrients, this relationship may have broad physiological implications. Moreover, as discussed below, aminergic neurotransmitters may turn out to be one of the groups of brain constituents, the syntheses of which vary with plasma composition. For example, choline's availability to cholinergic neurons may affect their production of both the neurotransmitter acetyl-

choline and the membrane constituent phosphatidylcholine.

Food consumption, tryptophan availability, and brain serotonin synthesis

The initial observation that physiological changes in precursor availability (i.e., after food consumption) could affect neurotransmitter synthesis was made in studies on rats performed in 1971 [4]. Animals were allowed to eat a test diet that contained carbohydrates and fat but lacked protein. Soon after the start of the meal, brain levels of the essential (and scarce) amino acid tryptophan were found to have risen, thus increasing the substrate saturation of the enzyme tryptophan hydroxylase that controls serotonin synthesis. The resulting increase in brain serotonin levels was associated with an increase in brain levels of serotonin's chief metabolite, 5-hydroxyindole acetic acid (5-HIAA), suggesting that serotonin release had also been enhanced. (Direct evidence that physiological variations in brain tryptophan concentrations affect serotonin release was not obtained, however, until 1987.)

The rise in brain tryptophan levels after consumption of this test diet was accompanied by a small increase (rats) or no change (humans) in plasma tryptophan levels. Both of these changes had been unanticipated, since the insulin secretion elicited by dietary carbohydrates was known to lower plasma levels of most of the other amino acids. However, the unusual response of plasma tryptophan to insulin was soon recognized as resulting from the amino acid's unusual propensity to bind loosely to circulating albumin. Insulin causes nonesterified fatty acid (NEFA) molecules to dissociate from albumin and to enter adipocytes. This dissociation increases the protein's capacity to bind circulating tryptophan; hence, whatever reduction insulin causes in "free" plasma tryptophan levels is compensated by a rise in the portion bound to albumin, yielding, in humans, no net change in total plasma tryptophan levels [7]. (Because this binding is of

low affinity, the albumin-bound tryptophan is almost as able as free tryptophan to be taken up into the brain.)

Considerably more difficult to explain were the data obtained subsequently on what happened to brain tryptophan and serotonin levels after rats consumed a meal rich in protein. Although plasma tryptophan levels rose, reflecting the contribution of some of the tryptophan molecules in the protein, brain tryptophan and serotonin levels either failed to rise or, if the meal contained sufficient protein, actually fell (8). The explanation for this paradox was found to lie in the kinetic properties of the transport systems that carry tryptophan across the blood-brain barrier (10) and into neurons. The endothelial cells that line central nervous system capillaries contain various macromolecules that shuttle specific nutrients or their metabolites between the blood and the brain's extracellular space. One such macromolecule mediates the transcapillary flux (by facilitated diffusion) of tryptophan and other large neutral amino acids (LNAA); others move choline, basic or acidic amino acids, hexoses, monocarboxylic acids, adenosine, adenine, and various vitamins. The amount of any LNAA transported by the macromolecule depends on its ability to compete with the other circulating LNAA. Thus the ability of circulating tryptophan molecules to enter the brain is increased when plasma levels of the other LNAA fall (as occurs after insulin is secreted) and is diminished when the other LNAA rise, even if plasma tryptophan levels remain unchanged. Since all dietary proteins are considerably richer in the other LNAA than in tryptophan (only 1.0–1.5% of most proteins), consumption of a protein-rich meal decreases the plasma tryptophan ratio (the ratio of the plasma tryptophan concentration to the summed concentrations of its major circulating competitors for brain uptake: tyrosine; phenylalanine; the branched-chain amino acids leucine, isoleucine, and valine; methionine). This, in turn, decreases tryptophan's transport into the brain and slows its conversion to serotonin. (Similar competitive mechanisms also mediate the passage of trypto-phan on other LNAA between the brain's extracellular space and neurons. Moreover, similar plasma ratios predict brain levels of each of the other LNAA after treatments that modify plasma amino acid patterns.)

The fact that giving pure tryptophan could increase brain serotonin synthesis and could thereby affect various serotonin-dependent brain functions (e.g., sleepiness, mood) had been known at least since 1968. What was novel and perhaps surprising about the above findings was their demonstration that brain tryptophan levels, and serotonin synthesis, normally undergo important variations in response, for example, to the decision to eat a carbohydrate-rich vs. a protein-rich breakfast. It remained possible, however, that mechanisms might exist external to the serotonin-releasing neuron that kept precursor-induced increases in serotonin's synthesis from causing parallel changes in the amounts released into the synapses. Indeed, it was known that if rats were given very large doses of tryptophan, sufficient to raise brain tryptophan levels well beyond their normal range, the firing frequencies of their serotonin-releasing raphe neurons decreased markedly; this was interpreted as reflecting the operation of a feedback system designed to keep serotonin release within a physiological range. (Similar decreases in raphe firing had also been observed in animals given drugs, like monoamine oxidase (MAO) inhibitors or serotonin-reuptake blockers, that cause persistent increases in intrasynaptic serotonin levels.) However, if rats were given small doses of tryptophan, sufficient to raise brain tryptophan levels but not beyond their normal peaks, or if they consumed a carbohydrate-rich meal, which raised brain tryptophan levels physiologically, no decreases in raphe firing occurred. Hence, food-induced changes in serotonin synthesis are able to affect the amounts of serotonin released per firing without slowing the neuron's firing frequencies and thus are "allowed" to modulate the net output of information from serotoninergic neurons.

The ability of supplemental tryptophan to enhance serotonin turnover within the human central nervous system (i.e., to elevate cerebrospinal fluid 5-HIAA levels) was first shown in 1970 (3); apparently, no neurochemical data are available concerning the human brain's responses to carbohydrate intake. Numerous behavioral and neurological effects have been associated with tryptophan administration, starting with Smith and Prockop's (11) original observation that it caused drowsiness and euphoria. Most of these effects have been reviewed extensively elsewhere (e.g., Ref. 7) and are not discussed further here.

Brain serotonin, nutrient choice, and carbohydrate craving

If rats are allowed to pick from foods in two pans presented concurrently that contain differing proportions of protein and carbohydrate, they choose among the two so as to obtain fairly constant (for each animal) amounts of these macronutrients. However, if before dinner they receive either a carbohydrate-based snack or a drug that facilitates serotoninergic neurotransmission, they quickly modify their food choice, selectively diminishing their intake of carbohydrates. These observations support the hypothesis that the responses of serotoninergic neurons to food-induced changes in the relative concentrations of plasma amino acids allow these neurons to serve a special function as sensors in the brain's mechanisms governing nutrient choice (13). Perhaps they participate in a feedback loop through which the composition of breakfast (that is, its proportions of protein and carbohydrate) can, by increasing or decreasing brain serotonin levels, influence the choice of lunch.

A similar mechanism may operate in humans. Subjects housed in a research hospital were allowed to choose from six different isocaloric foods (containing varying proportions of protein and carbohydrate but constant amounts of fat) at each meal, taking as many small portions as they liked; they also had continuous access to a computer-driven vending machine, stocked with mixed carbohydrate-rich and protein-rich isocaloric snacks. It was observed (e.g., Ref. 12) that the basic parameters of each person's food intake (total number of calories, grams

of carbohydrate and protein, number and composition of snacks) tended to vary only within a narrow range, day to day, and to be unaffected by placebo administration.

To assay the involvement of brain serotonin in maintaining this constancy of nutrient intake, pharmacological studies were undertaken in individuals in whom the putative feedback mechanism might be impaired. These were obese people who claimed to suffer from carbohydrate craving, manifested as their tendency to consume large quantities of carbohydrate-rich snacks, usually at a characteristic time of day or evening (12). Subjects were given D-fenfluramine (Isomerid), a drug that had been found to decrease carbohydrate intake in normal rats and to cause weight loss in obese people by a mechanism involving the release of brain serotonin. Administration of relatively low doses (15 mg twice daily) caused a major reduction in snack carbohydrate intake, a smaller reduction in mealtime carbohydrates, and no significant changes in mealtime protein nor fat intake (12). (Too few protein-rich snacks were consumed by the subjects to allow assessment of the drug's effect on this source of calories.) Other drugs also thought to enhance serotonin-mediated neurotransmission selectively, for example, the antidepressants zymelidine, fluvoxamine, and fluoxetine, also have been found to cause weight loss; this contrasts with the weight gain (and carbohydrate craving) often associated with less chemically specific antidepressants, such as amitriptyline. It has not yet been determined whether these drugs also selectively suppress carbohydrate intake in humans.

Severe carbohydrate craving is also characteristic of patients suffering from seasonal affective disorder syndrome (SADS), a variant of bipolar clinical depression associated with a fall onset, a higher frequency in populations living far from the equator, and concurrent hypersomnia and weight gain. A reciprocal tendency of many obese people to suffer from affective disorders (usually depression) has also been noted. Since serotoninergic neurons apparently are involved in the actions of both appetite-reducing and antidepressant drugs, they might constitute the link between a patient's appetitive and affective symptoms. Some patients with disturbed serotoninergic neurotransmission might seem to their physicians to have a problem with obesity, reflecting their overuse of dietary carbohydrates to treat their dysphoria. (The carbohydrates, by increasing intrasynaptic serotonin, would mimic the neurochemical actions of bona fide antidepressant drugs like the MAO inhibitors and tricyclic compounds.) Other patients might complain of depression, and their carbohydrate craving and weight gain would be perceived as secondary problems. Another group of patients, the non-anorexic bulimics, might seek medical assistance because of their concurrent food binges and depression or anxiety. Yet another group might include women with the premenstrual syndrome, which have late luteal phase mood disturbances, weight gain, carbohydrate craving, and sometimes fluid retention. The participation of serotoninergic neurons in a large number of brain functions besides nutrient choice regulation might have the effect of making such functions hostages to eating (seen in the sleepiness that can, for example, follow carbohydrate intake) just as it could cause mood-disturbed individuals to consume large amounts of carbohydrates for reasons related neither to the nutritional value nor taste of these foods. In support of this view, we have recently found that the serotoninergic drug D-fenfluramine can be an effective treatment for both the affective and the appetitive symptoms of SADS (unpublished observations).

Under what circumstances will nutrient intake affect neurotransmission?

On the basis of the tryptophan-serotonin relationship, one can formulate a sequence (15) of biochemical processes that would have to occur for any nutrient-precursor to affect the synthesis of its neurotransmitter product and the additional steps that would be required for the nutrient to also affect the release of the neurotransmitter.

1) Plasma levels of the precursor (and of other circulating compounds, such as the LNAA that affect tryptophan's availability to the brain) must be allowed to increase after its administration (or after its consumption as a constituent of foods). That is, plasma levels of tryptophan or the other LNAA or of choline cannot be under tight homeostatic control comparable to, for example, plasma calcium or osmolarity. In actuality, plasma levels of tryptophan, tyrosine, and choline do vary severalfold after the consumption of normal foods, and those of the branched-chain amino acids may vary by as much as five- or sixfold.

2) The brain level of the precursor must be dependent on its plasma level, i.e., there must not be an absolute blood-brain barrier for circulating tryptophan, tyrosine, or choline. In fact, such absolute barriers do not exist; rather, facilitated diffusion mechanisms operate that allow these compounds to enter the brain at rates that depend on the plasma levels of these ligands.

3) The rate-limiting enzyme within presynaptic nerve terminals that initiates the conversion of the precursor to its neurotransmitter product must, similarly, be unsaturated with this substrate so that when presented with more tryptophan, tyrosine, or choline it can accelerate synthesis of the neurotransmitter. [Tryptophan hydroxylase and choline acetyltransferase (CAT) do indeed have very poor affinities for their substrates tryptophan and choline. As discussed below, tyrosine hydroxylase activity becomes tyrosine limited when neurons containing the enzyme have been activated and the enzyme has been phosphorylated.]

4) The activity of this enzyme cannot be subject to local end-product inhibition, i.e., the products of tryptophan's hydroxylation, 5-hydroxy-tryptophan and serotonin itself, may not appreciably suppress tryptophan hydroxylase activity nor may acetylcholine levels within cholinergic nerve terminals affect CAT activity. Tyrosine hydroxylase activity probably is subject to some end-product inhibition when the enzyme protein is in its nonphosphorylated state; however, once the enzyme is phos-

phorylated, it apparently is freed from this constraint.

Available evidence suggests that only some of the neurotransmitters present in the human brain are likely to be subject to such precursor control, principally the monoamines mentioned above (serotonin; the catecholamines, dopamine, norepinephrine, and epinephrine; acetylcholine) and, possibly, histidine and glycine. Pharmacological doses of the amino acid histidine do elevate histamine levels within nerve terminals, and the administration of threonine, a substrate for the enzyme that normally forms glycine from serine, can elevate glycine levels within spinal cord neurons. One large family of neurotransmitters, the peptides, almost certainly is not subject to precursor control. Brain levels of these compounds have never been shown to change with variations in brain amino acid levels; moreover, there are sound theoretical reasons why it is unlikely that brain peptide synthesis would respond. The immediate precursor for a brain protein or peptide is not an amino acid per se, as is the case for some of the monoamine neurotransmitters, but the amino acid molecule attached to its particular species of tRNA. In brain tissue, the enzymes characterized to date that catalyze the coupling of an amino acid to its tRNA have very high affinities for their amino acid substrates, such that their ability to operate at full capacity, in vivo, is probably unaffected by amino acid levels (except possibly in pathological states, such as phenylketonuria, which are associated with major disruptions in brain amino acid patterns).

Little information is available concerning the possible precursor control of the nonessential amino acids, such as glutamate, aspartate, and γ-aminobutyric acid (GABA), that are probably the most abundant neurotransmitters in the brain, for the reason that it is very difficult to do experiments on these relationships. Even though glutamate and aspartate can be formed at various organs in the body via many different biochemical pathways, the precise pathways that synthesize these compounds within the terminals of neurons that use them as their neuro-

transmitters are not well established. In the case of GABA, although its precursor glutamate is well established, brain levels of that amino acid apparently cannot be raised experimentally without sorely disrupting normal brain functions. The macromolecule that transports acidic amino acids like glutamate and aspartate across the blood-brain barrier is unidirectional and secretes these compounds by an active-transport mechanism from the brain into the blood (10). Hence, administration of even an enormous dose of monosodium glutamate will not affect brain glutamate levels unless it elevates plasma osmolarity to the point of disrupting the blood-brain barrier, in which case the experimenter finds himself with a different experiment from the one that he had intended to perform.

In order for an increase in a neurotransmitter's synthesis, caused by administering a food or the transmitter's precursor, to affect its release, the neuron that releases it must continue to fire at its normal frequency. This may be kept from happening by receptor-mediated feedback processes that are activated soon after release of the transmitter has been increased. One such process, described briefly above, involves the presynaptic autoreceptors present on many monoaminergic terminals that are activated by intrasynaptic transmitter molecules. The other process involves chains of neurons, including at least one that makes an inhibitory neurotransmitter. The precursor-dependent neurotransmitter now interacts with postsynaptic receptors, ultimately causing the neuron that releases it to receive fewer excitatory (or more inhibitory) impulses and to fire less frequently. As described below, the administration of tyrosine or choline to normal individuals produces few detectable changes in brain function, probably because of the operation of these two feedback mechanisms. In contrast, serotonin-releasing neurons do not appear to decrease their firing frequencies when brain tryptophan levels are increased, unless the increase transcends the normal range, which is quite broad. Hence, brain serotonin synthesis is responsive to physiolog-

ical changes in brain tryptophan levels.

Tyrosine effects on dopamine and norepinephrine synthesis

Because tyrosine administration had not been shown to increase brain dopamine or norepinephrine levels in otherwise untreated animals, it was assumed until fairly recently that the catecholamine neurotransmitters were not under precursor control, in spite of the facts that plasma tyrosine levels do increase severalfold after protein intake or tyrosine administration; that the LNAA transport system does ferry tyrosine, like tryptophan, across the blood-brain barrier; and that tyrosine hydroxylase, which catalyzes the rate-limiting step in catecholamine synthesis, is unsaturated in vivo (13). It seemed possible that a pool of neuronal dopamine or norepinephrine might exist, the synthesis of which was indeed responsive to tyrosine but that the size of which was too small, in relation to total catecholamine levels, to be detected.

Hence, studies were performed to determine whether catecholamine synthesis or release, assessed independently of brain catecholamine levels, could be affected by changes in brain tyrosine concentrations. Catecholamine synthesis was estimated by following the rate at which dopa, the product of tyrosine's hydroxylation, accumulated in brains of rats treated acutely with a drug that blocks the next enzyme in catecholamine formation (aromatic L-amino acid decarboxylase); tyrosine administration increased dopa accumulation, whereas other LNAAs decreased both it and brain tyrosine levels (15). Catecholamine release was then estimated by measuring brain levels of metabolites of dopamine [homovanillic acid (HVA); dihydroxyphenylacetic acid (DOPAC)] or norepinephrine [methoxyhydroxyphenylglycol sulfate (MHPH-SO₄)]. Administration of even large doses of tyrosine had no consistent effect on these metabolites. However, if the experimental animals were also given an additional treatment designed to accelerate the firing of dopaminergic or noradrenergic tracts (e.g., dopamine receptor blockers,

cold exposure, partial lesions of dopaminergic tracts, reserpine), the supplemental tyrosine now caused a marked augmentation of catecholamine release (15). These initial observations formed the basis for the hypothesis that catecholaminergic neurons become tyrosine sensitive when they are physiologically active and lose this capacity when they are quiescent.

The biochemical mechanism that couples a neuron's firing frequency to its ability to respond to supplemental tyrosine involves phosphorylation of the tyrosine hydroxylase enzyme protein, a process that occurs when the neurons fire. This phosphorylation, which is short-lived, enhances the enzyme's affinity for its cofactor (tetrahydrobiopterin) and makes it insensitive to end-product inhibition (by norepinephrine and other catechols): these changes allow its net activity to depend on the extent to which it is saturated with tyrosine. An additional mechanism underlying this coupling may be an actual depletion of tyrosine within nerve terminals, as a consequence of its accelerated conversion to catecholamines. If slices of rat caudate nucleus are superfused with a standard Krebs Ringer solution that lacks tyrosine or other amino acids and are depolarized repeatedly, they are unable to sustain their release of dopamine; concurrently, their contents of tyrosine, but not of other LNAA, decline markedly. The addition of tyrosine to the superfusion solution enables the tissue to continue releasing dopamine at initial rates and also protects it against depletion of its tyrosine. The concentrations of tyrosine needed for these effects are proportional to the number of times the neurons are depolarized. (Of course, the intact brain is continuously perfused with tyrosine-containing blood, making it highly unlikely that tyrosine levels fall to a similar extent, even in continuously active brain neurons; however, they might decline somewhat, since tyrosine is poorly soluble in aqueous media, and diffuses relatively slowly.)

More recently, in vivo dialysis techniques have been used to assess tyrosine's effects on dopamine release from the rat's corpus striatum. When otherwise untreated animals receive the amino acid systematically, there is, after 20–40 min, a substantial increase in dopamine output, unaccompanied by detectable increases in levels of dopamine's metabolites DOPAC or HVA. However, this effect is short-lived, and dopamine release returns to basal levels after 20–30 min. This latter response probably reflects receptor-mediated decreases in the firing frequencies of the striatal neurons (to compensate for the increase in dopamine release that occurs per firing) or perhaps local presynaptic effects. If animals are given haloperidol, a dopamine-receptor blocking agent, before or along with the tyrosine, the supplemental tyrosine continues to amplify dopamine output for prolonged periods.

The tight coupling of tyrosine responsiveness to neuronal firing frequency (and tyrosine hydroxylase phosphorylation) probably explains tyrosine's paradoxical effects on blood pressure. The amino acid elevated blood pressure (and sympathoadrenal catecholamine release) in hypotensive animals but lowers blood pressure (without affecting sympathoadrenal catecholamines) in hypertensive animals (15). (It fails to affect blood pressure at all in normotensive animals or humans.) Tyrosine's blood pressure lowering effect in hypertensive animals probably results from its conversion to norepinephrine in brain stem neurons, which, when active, suppress sympathetic outflow; these neurons presumably are activated in the varieties of hypertension in which tyrosine is effective and may be participants in the brain's attempts to deal with the hypertension. As might be anticipated, tyrosine administration elevates brain levels of MHPG-SO$_4$ in these animals but has little or no effect on brain MHPG-SO$_4$ in those with normal or low blood pressure.

Effects of choline on synthesis of acetylcholine and phosphatidylcholine

The amounts of acetylcholine released by physiologically active cholinergic neurons depend on the concentrations of choline available. In the absence of supplemental free choline, the neurons will continue to release fairly constant quantities of the transmitter (8); however, when choline is made available (in concentrations bracketing the physiological range), a clear dose relationship is observed between its concentration and acetylcholine release (2, 8). (The biochemical mechanism that couples a cholinergic neuron's firing frequency to its choline responsiveness awaits discovery.) When no free choline is available, the source of the choline used for acetylcholine synthesis is the cells' own membranes (1, 8). Membranes are very rich in endogenous phosphatidylcholine (PC), and this phospholipid serves as a reservoir of free choline, much as bone and albumin serve as reservoirs for calcium and essential amino acids. It has been suggested that a prolonged imbalance between the amounts of free choline that are available to a cholinergic neuron and the amounts needed for acetylcholine synthesis might alter the composition of the neuron's membranes or its ability to make normal amounts of membranes to the point of interfering with normal neuronal functioning, [autocannibalism (2, 14)]. In that event, providing the brain with supplemental choline would serve two purposes: it would both enhance acetylcholine release from physiologically active neurons, and it would replenish the choline-containing phospholipids in their membranes.

Neurons can draw on three sources of free choline for acetylcholine synthesis: that stored as PC in their own membranes, that formed intrasynaptically from the hydrolysis of acetylcholine (and taken back up into the presynaptic terminal by a high-affinity process estimated to be 30–50% efficient in the brain), and that present in the blood stream (and taken into the brain by a specific blood-brain barrier transport system). PC in foods (e.g., liver, eggs) is rapidly hydrolyzed to free choline in the intestinal mucosa (or broken down more slowly after passage into the lymphatic circulation). Consumption of adequate quantities of PC can lead to severalfold elevations in plasma choline levels, thereby increasing brain choline and the substrate saturation of CAT.

The PC molecules consumed in the diet, as well as those formed

endogenously in neuronal membranes, are very heterogeneous with respect to their fatty acid compositions. Some PCs (e.g., those in soy beans and nerve terminals) are relatively rich in polyunsaturated fatty acids; others (e.g., in eggs) are highly saturated. PCs are also heterogeneous with reference to their mode of synthesis. Brain neurons produce PC by three distinct biochemical pathways: the sequential methylation of phosphatidylethanolamine (PE), the incorporation of preexisting free choline via the CDP-choline cycle, or the incorporation of free choline via the base-exchange pathway [in which a choline molecule substitutes for the ethanolamine in PE or the serine in phosphatidylserine (PS)]. Quite possibly, the different varieties of PC may subserve distinct functions; conceivably, one type of PC, distinguished by its fatty acid composition or its mode of synthesis, might be preferentially utilized to provide a choline source for acetylcholine synthesis, it might be formed preferentially during the processes of cell division or synaptic remodeling, or it might be involved in the pathogenesis of particular degenerative diseases afflicting cholinergic neurons (e.g., Alzheimer's disease).

Supplemental choline or PC has been used with success in the treatment of tardive dyskinesia. A recent summary of related publications (9) concluded that choline and the cholinesterase-inhibitor physostigmine were about equally efficacious and that choline was less toxic. Most patients exhibited some improvement in the frequency of abnormal movements, but in only a few was there complete cessation of the movements. Choline sources have also been tried in the treatment of Alzheimer's disease. Most well-controlled studies have treated subjects for relatively short intervals ($\leq$6–8 wk) and have focused on younger subjects, with little or no success. A single double-blind study administered the PC for 6 mo (6). Improvement was noted in about one-third of the subjects; the average age of the responders was 83 and that of nonresponders 73, a relationship thought to be compatible with evidence that Alzheimer's disease may be more restricted to cholinergic

neurons in subjects who become symptomatic at a later age. It seems important that additional long-term studies be done on the possible utility of PC in very old Alzheimer's patients. Occasional reports have also described useful effects of choline or PC in treating mania, ataxia, and myasthenic syndromes.

References

1. Blusztajn, J. K., M. Liscovitch, and U. I. Richardson. Synthesis of acetylcholine from choline derived from phosphatidylcholine in a human neuronal cell line. Proc. Natl. Acad. Sci. USA 84: 5474–5478, 1987.
2. Blusztajn, J. K., and R. J. Wurtman. Choline and cholinergic neurons. Science Wash. DC 221: 614–620, 1983.
3. Eccleston, D., G. W. Ashcroft, T. B. B. Crawford, J. B. Stanton, D. Wood, and P. H. McTurk. Effect of tryptophan administration on 5-HIAA in cerebrospinal fluid in man. J. Neurol. Neurosurg. Psychiatry 33: 269, 1970.
4. Fernstrom, J. D., and R. J. Wurtman. Brain serotonin content: physiological dependence on plasma tryptophan levels. Science Wash. DC 173: 149–152, 1971.
5. Fernstrom, J. D., and R. J. Wurtman. Brain serotonin content: physiological regulation by plasma neutral amino acids. Science Wash. DC 178: 414–416, 1972.
6. Little, A., R. Levy, P. Chaqui-Kidd, and D. Hand. A double-blind placebo controlled trial of high-dose lecithin in Alzheimer's disease. J. Neurol. Neurosurg. Psychiatry 48: 736–742, 1985.
7. Madras, B. K., E. L. Cohen, R. Messing, H. N. Munro, and R. J. Wurtman. Relevance of serum-free tryptophan to tissue tryptophan concentrations. Metabolism 23: 1107–1116, 1974.
8. Maire, J.-C., and R. J. Wurtman. Effects of electrical stimulation and choline availability on release and contents of acetylcholine and choline in superfused slices from rat striatum. J. Physiol. Paris 80: 189–195, 1985.
9. Nasrallah, H. A., F. L. Dunner, R. E. Smith, M. McCalley-Whitters, and A. D. Sherman. Variable clinical response to choline in tardive dyskinesia. Psychol. Med. 14: 697–700, 1984.
10. Pardridge, W. M. Regulation of amino acid availability to the brain. In: Nutrition and the Brain, edited by R. J. Wurtman and J. J. Wurtman. New York: Raven, 1977, vol. 7, p. 141–204.
11. Smith, B., and D. J. Prockup. Central-nervous-system effects of ingestion of L-tryptophan by normal subjects. N. Engl. J. Med. 267: 1338–1341, 1962.
12. Wurtman, J. J., R. J. Wurtman, S. Mark, R. Tsay, W. Gilbert, and J. Growdon. D-Fenfluramine selectively suppresses carbohydrate snacking by obese subjects. Int. J. Eating Disord. 4: 89–99, 1985.
13. Wurtman, R. J. Behavioural effects of nutrients. Lancet 1: 145–147, 1983.
14. Wurtman, R. J. Alzheimer's disease. Sci. Am. 252: 62–75, 1985.
15. Wurtman, R. J., F. Hefti, and E. Melamed. Precursor control of neurotransmitter synthesis. Pharmacol. Rev. 32: 315, 1980.
16. Young, S. The clinical psychopharmacology of tryptophan. In: Nutrition and the Brain, edited by R. J. Wurtman and J. J. Wurtman. New York: Raven, 1985, vol. 7, p. 49–88.